Preservation and survival of E. coli in well water samples submitted for routine analyses. [DNR-166] [2002]

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Preservation and Survival of *E. coli* in Well Water Samples Submitted for Routine Analyses

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Background

Total coliform testing to determine potability of drinking water has been performed routinely on Wisconsin well waters since the beginning of the 20th century. The data derived from this testing has driven the development and continuous improvement of the administrative codes that regulate well construction and maintenance. The quality assurance program associated with laboratory testing for coliforms establishes maximum sample holding times (APHA, 1998)[1]. For public water supply testing, these holding times are established by the United States Environmental Protection Agency (USEPA, 1989)[2]. The agency uses published data and expert opinion as the basis for setting the required holding times. Currently the holding time is set at 30 hours. In Wisconsin, the Laboratory of Hygiene (WSLH) did an extensive study that demonstrated coliforms actually survive quite well for up to 48 hours at ambient temperatures in typical Wisconsin well water samples (Standridge, 1983)[3]. Subsequent to this study, the USEPA granted an exception for the WSLH to allow compliance testing for total coliforms up to 48 hours after collection. This exception has permitted a high compliance rate for required testing of drinking waters, even from remote areas of the state where delivery to the laboratory can often take two days.

During the mid nineties, changes in available technology moved the testing of drinking water ahead by a quantum leap. The introduction of enzyme based assays allowed for low cost, rapid testing for *E. coli* simultaneously with total coliform analysis. (Edberg, 1989, McCarty, 1992). The advent of this new technology has allowed public health officials to rethink how they look at indicator testing of drinking water aimed at reducing the incidence of waterborne disease. While total coliform testing has been the cornerstone of lab based water supply protection activities, the reality is that the test often sounds false alarms. (Standridge, 1996). The intent of coliform testing is to detect situations where fecal material has compromised a water source. Since coliforms can exist in many non-fecal environments, a large percentage of the positive tests occur in situations where no fecal contamination is present. This false positivity often results in poor expenditures of resources to deal with problems that are not a real public health threat. On the other hand the detection of *E. coli* provides direct unequivocal evidence of fecal contamination and does warrant a strong response from public health experts. The ability to easily detect *E. coli* has precipitated actual changes and proposed changes in how officials respond to coliform/*E. coli* testing results. For regulated public water supplies, the USEPA has removed total coliform positivity as a trigger for emergency (boil water) or acute violations where "boil water" advisories are issued. The only way a total coliform result can trigger a boil water order is if it is coupled with another test from the same time period that was *E. coli* positive. For privately owned water wells the Wisconsin Department of Natural Resources has discussed a move to a two level interpretation of bacteriological water tests. Wells testing positive for total coliforms only could be classified as "at risk" and wells with both total coliform and *E. coli* present would be classified as "unsafe".

This general move towards decision making based on *E. coli* test results, creates a need for understanding whether or not *E. coli* survives as well as the total coliform group while samples are in transit to the laboratory. Previously published work in this area is incomplete since it focused primarily on surface waters and studied a limited array of temperatures and storage times. The work described in this report provides information to more completely understand the issue of *E. coli* survival in water samples. The topic of establishing holding times for water samples submitted for *E. coli* testing is extremely important and timely. If *E. coli* dies off during transit to the laboratory, unsafe water supplies could be classified as safe. Decisions aimed at increasing the emphasis placed on *E. coli* positivity are close at hand. The data presented in this study provides information for making data based decisions regarding sample preservation requirements.

Experimental Design

The purpose of this study was to provide data on the survival of *E. coli* in water samples, to be used in setting storage a handling requirements. The ability of a bacterium to survive in a water environment is dependent on temperature, time, available nutrients, competing microbial flora and the presence of toxicants. (Le Chevallier, 1990, Smith, 1989).

Temperature

Most drinking water samples submitted as a part of a monitoring program are collected and shipped to the laboratory with no attempt to control transit temperatures. In Wisconsin, an attempt is made to hold ambient temperature by shipping samples in individual styrofoam™ mailers. While a properly collected groundwater sample may begin its trip to the lab at 10-15^{O}C, it often arrives at the laboratory as warm as 30^{O}C. At the other end of standard practice, there are some laboratories that require samples be cooled during transit, where proper cooling of the sample is defined as a final transit temperature below 10^{O}C. This is usually accomplished by submitting the sample in some sort of insulated "cooler" with pre-frozen ice packs. There is ample evidence showing that the enzymatic activities that may lead to the death of a bacterium essentially stop at temperatures less than 4^{O}C. (Standridge, 1983 and 1987). To achieve this temperature, samples must be submitted in an insulated cooler in direct contact with wet ice where the ice mass must exceed the mass of the sample. With any attempts to cool samples, care must be taken to assure that the samples do not freeze, as freezing of bacterial cells will often lyse the cell wall. Given this variety of temperatures routinely used by laboratories, the researchers chose to include four different temperature variables; 4^{O}C, 10^{O}C, 20^{O}C and 30^{O}C.

Concerns were raised between using strictly controlled temperatures, with refrigerators and incubators, verses use of coolers and ice as is used in actual practice. The researchers decided that to more clearly understand the effects of specific temperatures and to produce consistent and reliable data it would be advantageous to use precise temperature control achieved with thermostatically controlled refrigerators and incubators.

Time

Opinions and practice on the maximum allowable storage time for water samples prior to bacteriological analysis varies substantially. Some scientists feel that testing must be done immediately after collection, as is the practice for larger utilities that have their own laboratories. At the WSLH, 90% of all samples are tested within 30 hours of collection, with the remaining samples tested within 48 hours. Other Wisconsin laboratories testing private water supplies accept samples up to 72 hours after collection. The range of holding times selected for this study was based on actual practice for Wisconsin labs; immediately after collection/preparation, eight hours, 30 hours, 48 hours and 72 hours after collection.

Chemical make-up and microbial flora

Another critical factor that affects survival of a bacterium in water is the overall chemical make-up of the water. This is a complex issue that includes many factors such as pH,

buffering capacity, carbon nitrogen and phosphorous content, heavy metal toxicants, and organic toxicants. (Standridge, 1983, Le Chevallier, 1990, Smith 1989).

Due to the difficulty in preparing waters that truly represent naturally occurring chemical conditions, the researchers chose to use a variety of natural Wisconsin waters, collected across the seasons, that truly represented the actual conditions seen in Wisconsin groundwater's. This approach also allowed testing on a variety of microbial floras that might affect coliform survival.

Experimental Protocol

Sample collection/preparation:

FifNineteen water samples were collected from southern Wisconsin to provide a variety of chemically and microbiologically diverse waters representing different geological formations, well depths and types of well construction. An attempt was made to collect samples from naturally contaminated wells. Contaminated wells were chosen based on previously determined positive results from groundwaters submitted to the WSLH for routine testing. Phone calls were made to these locations to acquire permission to take water samples. Unfortunately, only two naturally contaminated samples were successfully collected and tested. Most of the samples were not contaminated and required spiking with laboratory cultures. Additionally, samples were collected from polluted surface water sources to provide "worst case" scenario water types. Characterizations of the samples are provided in Table 1.

Table 1

Site ID	County	Site Description	Date Collected
G W	Dane	Private well in heavily farmed area	4/23/2001
K W	Dane	18 foot sand point well 50 feet from the Yahara river in a residential area	7/09/2001
W W	Dane	Agricultural area dog fecal material observed near well	7/16/2001
St W	Waupaca	Shallow sand point well 150 feet from Bear Lake in a residential/lightly farmed area.	7/31/2001

Site ID	County	Site Description	Date Collected
So W	Dane	6 inch, cased 100 foot drilled well in residential area	10/31/2001
E M	Iowa	Convenience store in heavy agricultural area	12/12/2001
Oetf W	Waushara	Machine operators facility in a non-farmed rural area near Coloma	2/5/2002
Cp W	Sauk	Sand point well , gas station/convenience store in light commercial development/agriculture area	2/05/2002
N W	Clark	Motel in non-ag rural area. Very old well construction in disrepair.	3/29/2002
V W	Dane		3/29/2002
Blcw W	Waukesha	church located in village of Wales near light commercial land uses	4/15/2002
Bumc W	Sauk	Church in a rural heavily farmed area	4/23/2002
Ol W	Dane	Drilled well in agricultural area	4/26/2002
Lwdp	Dane	Spring fed pond source water to Lake Wingra	6/22/01
l M	Dane	Urban/rural eutrophic lake	5/01/2002

Sampling sites were chosen to be within two hours driving distance of Madison. On the day before each experiment, a sample was collected and an initial determination was made to estimate background levels of *E. coli*. On the day of the experiment, a driver was dispatched to the sampling site to collect a 20-liter sample, which was iced and returned to the laboratory. Within 2 hours of collection aliquots of the well mixed sample were placed in sterile plastic sample bottles. The uncontaminated samples were immediately spiked with two known concentrations of *E. coli.* The two concentrations of organism used were 10 to 15 *E. coli* bacteria/100 ml and 100 to 150 *E. coli* bacteria/100 ml. These numbers were chosen to represent organism concentration levels normally seen in contaminated drinking water samples. A single passage master culture of an environmental isolate *of E. coli* was aliquoted to freezing media vials and frozen at –70° C for use as the spiking organism. The strain was verified as *E. coli* using the API 20E enteric bacteria identification system and conventional biochemicals. For each water sample tested, one frozen vial was thawed and grown in trypticase soy broth and incubated at 35°C overnight. This master culture was then serially diluted. To reach the desired number of organisms, the McFarland Equivalence Turbidity Standard was used. A McFarland 0.5 turbidity standard represents a solution with approximately 1.5 X 10^8 organisms per milliliter. Using a manual spectrophotometer, the master *E. coli* culture was diluted to reach that of the 0.5 McFarland standard using phosphate buffer as a diluent. From the McFarland 0.5 standard dilution, three more successive dilutions of 1:1000, 1:1000 and 1:10 were made. The second 1:1000 (≈1.5 X 10^2 *E. coli* /ml) and the 1:10 (≈1.5 X *10 E. coli*/ml) were used to spike the water samples. For the unspiked samples, 60 bottles were prepared (3 replicates x 4 holding temperatures x 5 time intervals). For the spiked samples this number was doubled due to the two spiking concentrations.

Sample holding condition:

Appropriate numbers of samples from each group were placed in refrigerators or incubators designed to hold closely to the predetermined temperatures. Temperatures were monitored using Thermochron iButtons™. The Thermochron iButton™ integrates a thermometer, a clock/calendar, a thermal history log and 512 bytes of additional memory into a small stainless steel case the size of approximately 5 dimes. The thermometer measures temperature from –40°C to +85°C in 0.5 increments, while the clock measures seconds to years accurately to ±1 minute per month from 0°C to 45°C. (Thermochron, 2000)[7]. Temperature monitoring with the iButton was done by using a separate bottle filled with 100 ml of sterile water and placing the iButton in the bottle. The iButtons™ were programmed to collect temperature data every 15 minutes. This bottle was then refrigerated or incubated along with the rest of the bottles for each of the four temperatures. After 72 hours the iButton was removed from the bottle and the data was downloaded to a personal computer which archived the data and created a thermograph of sample temperature versus time.

E. coli testing:

All testing for E. coli was done using the MMO-MUG defined substrate (Colilert™) methodology with Quantitray 2000™ most probable number enumeration following Standard Methods procedures. This technique resulted in a numeric value for E. coli

levels. (Edberg, 1989, APHA, 1998)[8]. Due to the low precision of the most probable number enumeration, all tests were done in triplicate to increase the robustness of the database thus reducing the standard error around the data points.

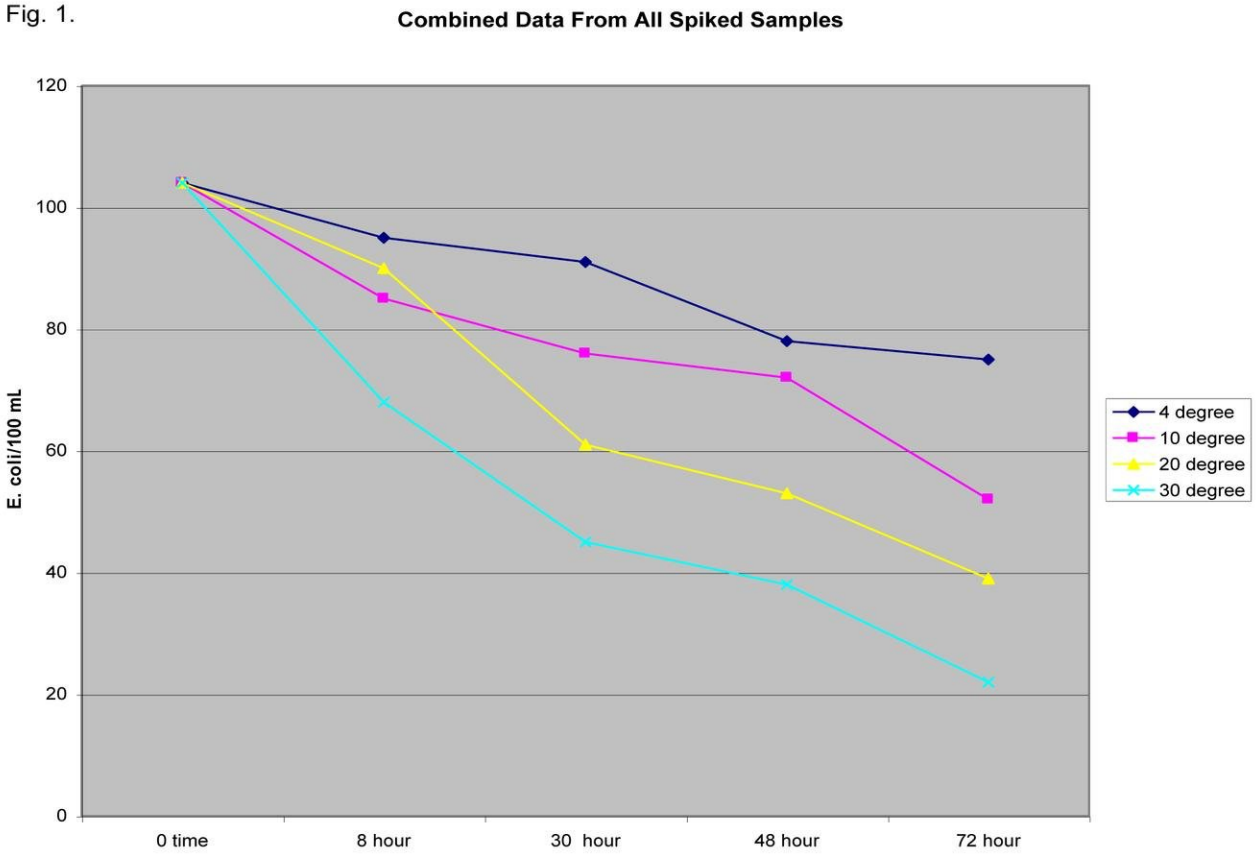
Quality assurance/ Quality control:
A Quality Assurance (QA) plan was designed for this study to ensure that all data are of known quality. The Wisconsin State Laboratory of Hygiene Water Microbiology unit is certified by both the USEPA and the Wisconsin Department of Agriculture and Consumer Protection for all the analyses performed for this study. All aspects of the project were managed with a team approach using tools such as timed agendas, flow charting, brainstorming, time lines, "plan do check act" action plans, "equal air time" meeting processes, cause and effect diagrams, extensive use of data in decision processes and continuous focus on the customer's (Ground water Coordinating Council) needs and expectations. The team included a quality assurance data check as part of each team meeting. All experiments stayed within control limits except where noted. Temperature holding refrigerators and incubators were occasionally adjusted.

Results:

Note: In order to facilitate any possible future uses of the data created from this study, all of the raw data from each sampling event are presented in appendices 1-15.

With 1,620 *E. coli* measurements, the data set from this study is fairly large. One way to begin understanding the data is to look at it in summary form. To begin with we have combined all of the data from the 12 samples that were spiked with *E. coli*. All of the counts from all of the holding temperatures from each sample run have been averaged and are then plotted against each time interval. This four line summary graph is presented below as Figure 1. This graph shows two general trends. First, *E. coli* concentrations in a sample decrease over time, and secondly, *E. coli* concentrations decrease more rapidly at warmer temperatures.

Fig. 1.



While the elucidation of these general trends is helpful, it does not really answer the question of "how long, and at what temperature can water samples be held prior to analysis for *E. coli*?". For the same group of spiked samples summarized above, we can look at each one of the holding temperatures separately and apply some basic statistical analysis. It should be noted that for one of the sampling sites rapid die-off occurred. The well had been batch chlorinated on the day before the sample was collected. Since no attempt was made to test the sample for the presence of chlorine or to de-chlorinate the sample, it is likely that some chlorine was present. For this sample even testing the water at 8 hours would have resulted in a negative test. This data set was considered an outlier, and was not used in subsequent analysis.

Once again the data can be presented graphically with the average *E. coli* counts for the remaining 11 spiked samples represented as a separate bar for each holding time. Additionally the 95%confidence interval can be calculated for each of these averages and represented as error whiskers on the bars. When comparing one time frame against another, if the average value, represented as the top of the bar, falls within the error whiskers range of the other average, you can be 95% assured that the two averages are statistically the same. The bar graphs for the four holding temperatures are presented in Figures 2-5 below.

The current standard for holding of water samples prior to *E. coli* analysis is the USEPA maximum allowable time of eight hours with a suggestion that samples should be iced or refrigerated. Consequently it makes sense to compare the effects of holding time periods to the currently acceptable eight hours. Looking at the combined 4 degree data for the 11 runs spiked with high levels of organisms, (Fig. 2) it is clear that while the counts go down slightly over time, there is no statistical difference between numbers of *E. coli* detected even with the samples held for 72 hours. For samples held at 10 degrees (Fig. 3), there is no statistically significant difference between 8 hours and 30 or 48 hours. *E. coli* levels in samples held at 10 degrees for 72 hours decrease to the point where they are statistically lower than the eight hour numbers. For samples held at 20 degrees (Fig. 4), all of the levels in samples stored longer than 8 hours are significantly lowered. Looking further at the raw data used to create figure 4, it can be noted that the die-off occurred in three of the eleven samples. Interestingly, the samples stored at 30 degrees (Fig. 5.) while generally showing numbers lower than storage at cooler temperatures, demonstrate no statistical difference between samples stored 8 hours, 30 hours.

Fig. 2

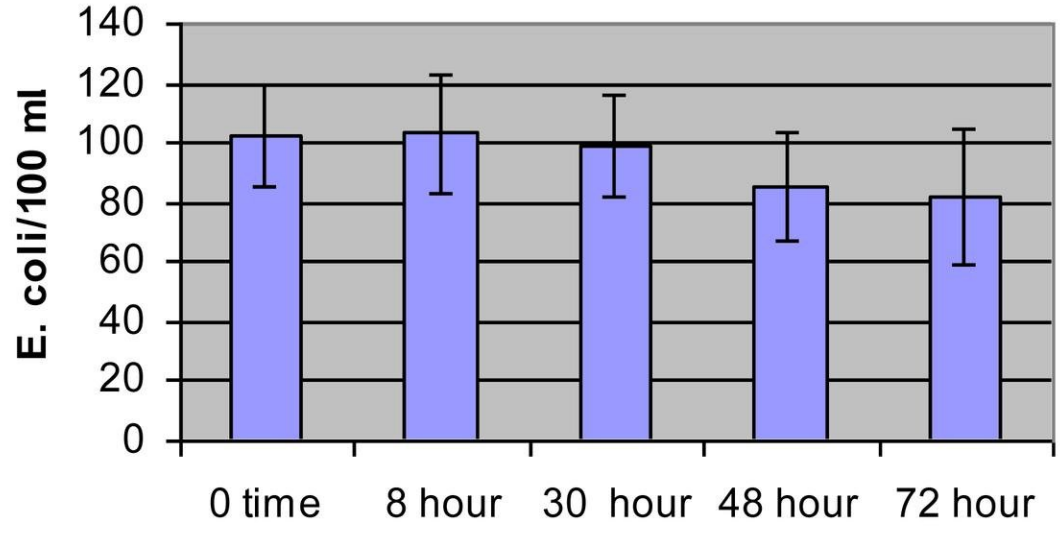

4 degrees holding Temperature
E. coli/100 ml
140
120
100
80
60
40
20
0
0 time
8 hour
30 hour
48 hour
72 hour

Fig. 3

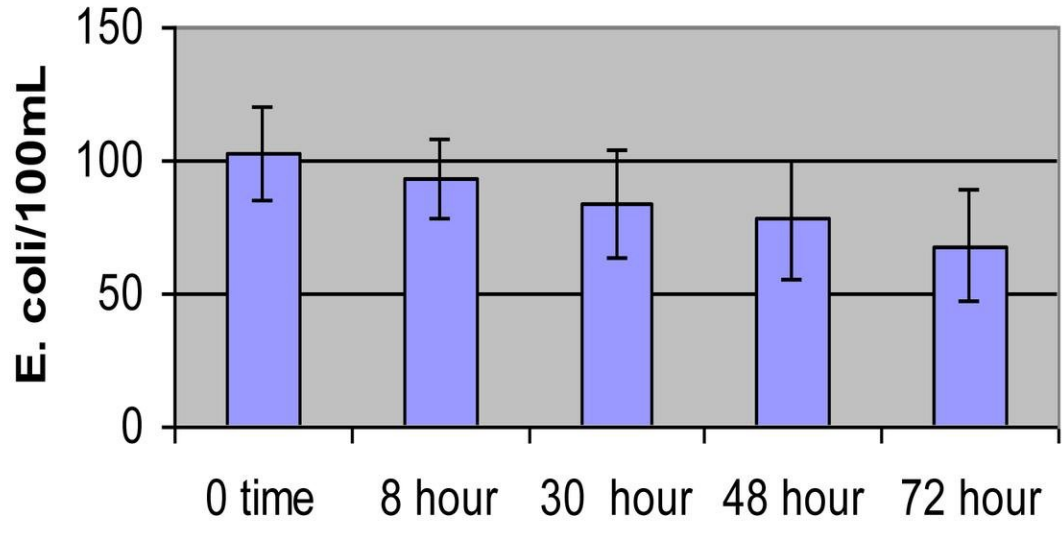

10 degree Holding Temperature
E. coli/100mL
150
100
50
0
0 time
8 hour
30 hour
48 hour
72 hour

Fig. 4.

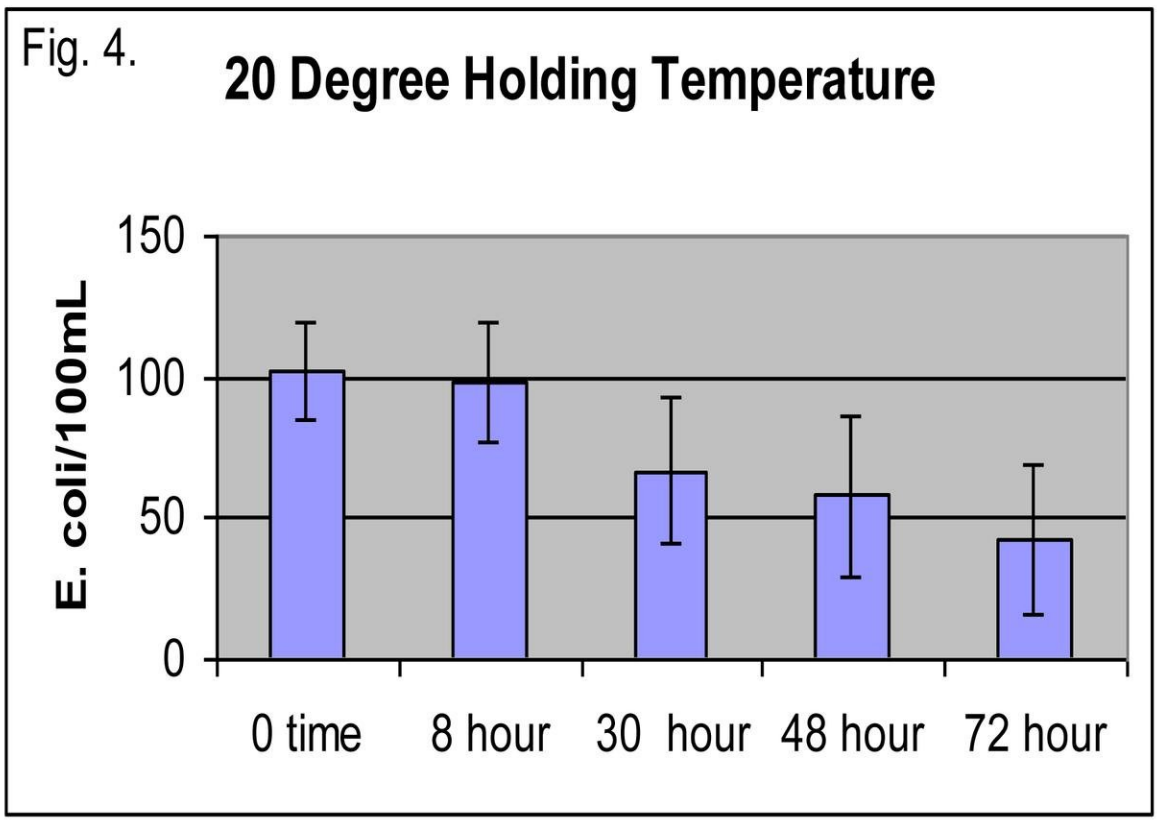
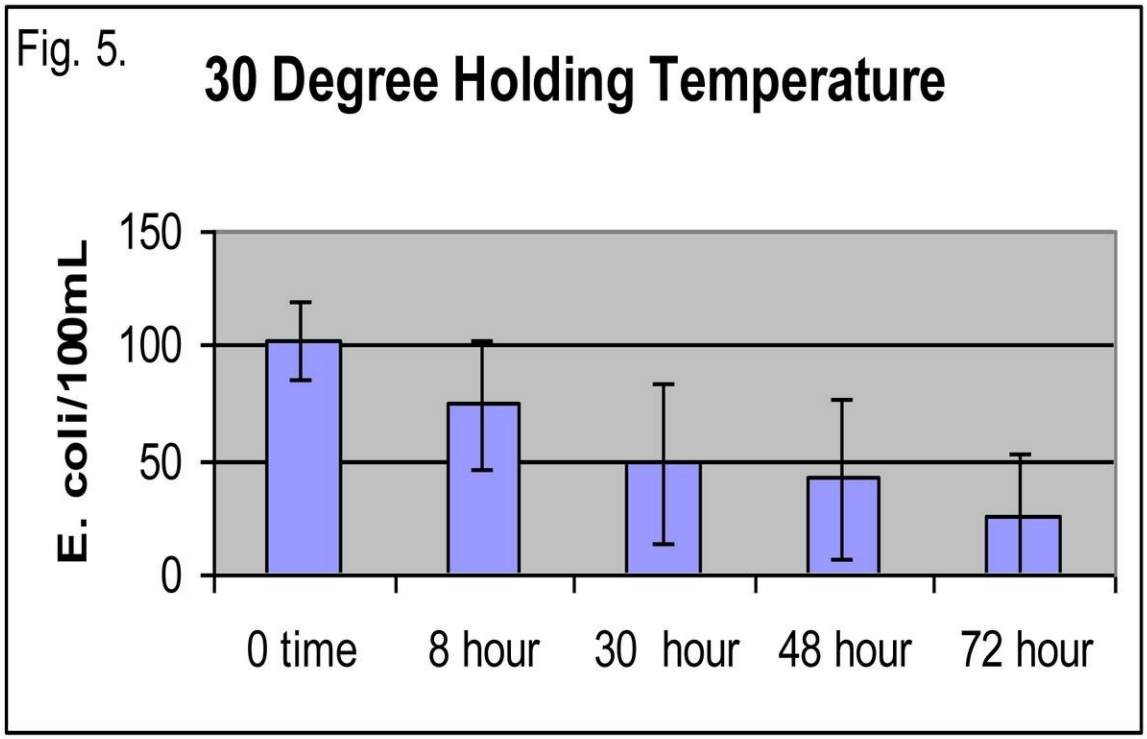

20 Degree Holding Temperature
E. coli/100mL
150
100
50
0
0 time
8 hour
30 hour
48 hour
72 hour

Fig. 5.


30 Degree Holding Temperature
E. coli/100mL
150
100
50
0
0 time
8 hour
30 hour
48 hour
72 hour

While the high level spikes provide numbers of organisms in a range usable for statistical analysis, samples spiked with low numbers of organisms more closely represent the real world. These data can be examined from a presence/absence view point. That is, the public health significance of a sample test result for *E. coli* is based solely on whether or not the organism can be detected. Therefore a holding time and temperature combination would be unacceptable only if it resulted in a test result of no *E. coli* detected. This data is presented in Table 2.

Table 2.

Number of Samples Out of Eleven Runs Testing Positive for *E. coli*
On Samples Spiked with @ 10 Organisms/100mL

Temperature	0 Time	8 hours	30 hours	48 hours	72 hours
4 degrees	11	11	11	11	11
10 degrees	11	11	11	10	10
20 degrees	11	11	10	8	8
30 degrees	11	10	6	6	5

The most striking observation from this data is that *E. coli* in all the samples held at 4 degrees, and the majority of samples held at 10 degrees is still detectable up to 72 hours. Even at room temperature, the *E. coli* is still detectable in 10 of the 11 samples at 30 hours. At the 30 degree temperature, significant die-off occurs after eight hours.

Since laboratory grown *E. coli* spikes may behave differently than wild type strains, attempts were made to sample sites that were naturally contaminated. Our original hope was that we could find a least 10 naturally contaminated wells. In reality we were only able to find two wells, both of which had very low levels of contamination. The results are presented below in Table 3. All *E. coli* counts are presented as the average of the three replicates described above. Although this data set is small, it is consistent with the spiked sample data, and suggests that Wild type *E. coli* also tolerate storage over time particularly when the samples are kept cold.

Table 3.

Clark County well					
	E. coli	E. coli	E. coli	E. coli	E. coli
Storage	per 100mL	per 100mL	per 100mL	per 100mL	per 100mL
Temperature	0 time	8 hours	30 hours	48 hours	
4	0.6	1.3	0.3	0	0.6
10	0.6	1	0	0	0.3
20	0.6	0	0	0.3	0
30	0.6	0.6	0	0	0
Dane County well					
	E. coli	E. coli	E. coli	E. coli	E. coli
Storage	per 100mL	per 100mL	per 100mL	per 100mL	per 100mL
Temperature	0 time	8 hours	30 hours	48 hours	72 hours
4	6.7	4.1	1.7	2.4	0
10	6.7	3.1	2.4	2	1.7
20	6.7	1.7	0.33	1	1
30	6.7	2.6	0.33	1.4	1

Preliminary analysis of the work funded by this study precipitated a USEPA study to further evaluate holding times and temperatures for *E. coli* in surface water samples. The protocols for the EPA study were the same as those employed for the GWCC study with the exception that the storage temperatures and holding times were altered somewhat. The work performed for this aspect of the study is presented separately in Table 4. below. The results from these tests are very similar to the GWCC results. Little change in *E. coli* concentrations were observed for samples held at both 4 and 10 degrees for up to 48 hours. Some die-off did occur at higher storage temperatures.

Table 4.
USEPA study site 1, Wisconsin River below the Dells dam.*

Temp	0 time	8 hours	24 hours	30 hours	48 hours
4°	79	65	81	61	38
10°	79	53	65	52	32
20°	79	57	32	21	10
35°	79	59	27	7	4

* Data taken from Quantitray method only

USEPA study site 2, Rainbow Lake-Wisconsin Veterans Home.*

Temp	0 time	8 hours	24 hours	30 hours	48 hours
4°	76	77	72	86	81
10°	76	78	74	75	59
20°	76	93	45	41	15
35°	76	18	6	5	0.33

* Data taken from Quantitray method only

USEPA study site 3, Lincoln Creek.*

Temp	0 time	8 hours	24 hours	30 hours	48 hours
4°	264	255	240	331	281
10°	264	271	212	207	153
20°	264	283	223	167	120
35°	264	261	171	96	40

* Data taken from Quantitray method only

USEPA study site 4, Lake Winnebago Oshkosh water intake.*

Temp	0 time	8 hours	24 hours	30 hours	48 hours
4°	214	211	220	227	183
10°	214	238	161	162	206
20°	214	244	167	133	131
35°	214	211	151	183	135

* Data taken from Quantitray method only

Conclusions: While sample holding time criteria is in place for drinking water samples submitted for total coliform analysis, currently the USEPA has no guidelines for sample holding times and shipping temperatures for drinking water samples submitted for *E. coli* testing. For surface water samples the guidelines state that samples must be refrigerated in transit and tested within eight hours of collection. The general move in the regulation of drinking water towards decision making based on *E. coli* test results, creates a need for understanding whether or not *E. coli* survives as well as the total coliform group while samples are in transit to the laboratory. The work described in this report provides much of this information. The data provides a strong basis for a decision to expand the allowable storage time of water samples submitted for *E. coli* analysis beyond the current eight hour limit as well as the basis for supporting only one

recommended preservation protocol for both surface waters and drinking water samples.

All samples including those with very low levels of bacteria can safely be preserved for at least 48 hours if held at 4 degrees C, the temperature usually achieved by shipping samples packed in wet ice. 48 hour package delivery to Madison can easily be achieved from all areas of Wisconsin. Thus, water samples shipped in coolers packed with wet ice could be accurately analyzed up to 48 hours after collection. The data also shows that in all the trials except one, *E. coli* can be preserved for 48 hours when held at 10 degrees C, and all samples can be preserved at 10 degrees C for 30 hours. 10 degree shipping temperatures can be achieved with the use of "blue ice" freezer packs in coolers, which simplifies the shipping process as compared to dealing with wet ice. The current practice of shipping drinking water samples in Styrofoam boxes to hold the temperature at approximately 20 degrees C would be valid for almost all samples for up to 30 hours. The data clearly shows that samples held at 30 degrees C are unsuitable for *E. coli* testing. This finding suggests that samples submitted during the summer months, with no attempt at preservation through cooling would not be suitable for *E. coli* testing.

This data suggests that the USEPA established holding time of eight hours for surface water samples submitted for *E. coli* testing, is overly stringent. A change to a maximum holding time of chilled samples for up to 30 hours could easily be supported by the data presented in this study. The data also suggests that the current practice of allowing up to 48 hours for drinking water samples submitted with no attempt to cool the samples may be too lax. A reduction in the time period to 30 hours, or a requirement to ship the samples at less than 10 degrees C, could be supported by the data.

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Appendix 1.

Water source	GW
Date Collected	4/23/01
Date/time TSB inoc.	4/22 3:30pm
Organism used	E coli
Date/time tests inoc.	4/23 2:00pm
Low Concentration	
Zero time	
Rep 1	5.2
Rep 2	6.3
Rep 3	14.5
average	8.7

Sample background E. coli levels (zero time)

Rep 1	0
Rep 2	0
Rep 3	0
average	0.0

High Conc.

Zero time	
Rep 1	95.9
Rep 2	83.6
Rep 3	98.7
average	92.7

4 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	15.6	Rep 1	12.2	Rep 1	5.2	Rep 1	5.2
Rep 2	13.2	Rep 2	7.4	Rep 2	9.6	Rep 2	3.1
Rep 3	12	Rep 3	8.6	Rep 3	5.2	Rep 3	5.2
average	13.6	average	9.4	average	6.67	average	4.5

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	101.4	Rep 1	110.6	Rep 1	93.3	Rep 1	87.8
Rep 2	79.8	Rep 2	114.5	Rep 2	81.6	Rep 2	108.1
Rep 3	141.4	Rep 3	146.7	Rep 3	77.1	Rep 3	104.3
average	108	average	124	average	84	average	100

10 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	7.4	Rep 1	12	Rep 1	7.4	Rep 1	13.2
Rep 2	4.1	Rep 2	9.8	Rep 2	13.4	Rep 2	6.3
Rep 3	5.2	Rep 3	10.9	Rep 3	9.6	Rep 3	6.3
average	5.57	average	10.9	average	10.1	average	8.6

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	73.3	Rep 1	88.2	Rep 1	88.6	Rep 1	67.7
Rep 2	64.4	Rep 2	69.7	Rep 2	101.7	Rep 2	76.6
Rep 3	125	Rep 3	56.3	Rep 3	60.5	Rep 3	71.7
average	87.6	average	71.4	average	83.6	average	72

20 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	13.4	Rep 1	7.4	Rep 1	2	Rep 1	0
Rep 2	3.1	Rep 2	14.6	Rep 2	4.1	Rep 2	6.3
Rep 3	9.8	Rep 3	7.4	Rep 3	3.1	Rep 3	10.8
average	8.8	average	9.8	average	3.1	average	5.7

20 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	115.3	Rep 1	68.3	Rep 1	75.4	Rep 1	78.9
Rep 2	139.6	Rep 2	93.2	Rep 2	88.4	Rep 2	63.8
Rep 3	123.6	Rep 3	86	Rep 3	48	Rep 3	66.9
average	126	average	82.5	average	70.6	average	69.9

30 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	13.2	Rep 1	6.3	Rep 1	3	Rep 1	1
Rep 2	16.8	Rep 2	12.2	Rep 2	0	Rep 2	1
Rep 3	9.8	Rep 3	5.2	Rep 3	3.1	Rep 3	4.1
average	13.3	average	7.9	average	2.03	average	2.03

30 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	98.4	Rep 1	1	Rep 1	1	Rep 1	5.2
Rep 2	98.7	Rep 2	13.4	Rep 2	2	Rep 2	17.3
Rep 3	96	Rep 3	37.9	Rep 3	21.8	Rep 3	8.6
average	97.7	average	17.4	average	8.27	average	10.4

Appendix 2.

	K W		
Date Collected	7/9/01		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	7/8 12 noon	Rep 1	0
Organism used	E coli	Rep 2	0
Date/time tests inoc.	7/9/01 9:30	Rep 3	0
Low Conc.		average	0

Zero time	
Rep 1	21.6
Rep 2	11
Rep 3	12.2
average	14.9

High Conc.

Zero time	
Rep 1	116.2
Rep 2	172.5
Rep 3	186
average	158.2

4 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	13.4	Rep 1	10.9	Rep 1	13.4	Rep 1	17.5
Rep 2	9.8	Rep 2	14.5	Rep 2	12.1	Rep 2	16
Rep 3	6.3	Rep 3	7.3	Rep 3	12.1	Rep 3	14.6
average	9.8	average	10.9	average	12.5	average	16.0

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	178.5	Rep 1	178.2	Rep 1	83.9	Rep 1	161.6
Rep 2	218.7	Rep 2	118.7	Rep 2	155.3	Rep 2	160.7
Rep 3	135.4	Rep 3	133.3	Rep 3	131.3	Rep 3	135.4
average	177.5	average	143.4	average	123.5	average	152.5

10 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	16	Rep 1	12.1	Rep 1	21.8	Rep 1	8.5
Rep 2	14.6	Rep 2	14.3	Rep 2	17.5	Rep 2	9.8
Rep 3	21.6	Rep 3	14.3	Rep 3	13.2	Rep 3	12.1
average	17.4	average	13.6	average	17.5	average	10.1

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	129.6	Rep 1	156.5	Rep 1	120.1	Rep 1	118.7
Rep 2	151.5	Rep 2	148.3	Rep 2	146.7	Rep 2	90.5
Rep 3	122.3	Rep 3	157.6	Rep 3	135.4	Rep 3	129.6
average	134.5	average	154.1	average	134.1	average	112.9

20 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	16	Rep 1	18.7	Rep 1	18.7	Rep 1	17.1
Rep 2	5.2	Rep 2	11	Rep 2	20.1	Rep 2	18.3
Rep 3	14.6	Rep 3	14.5	Rep 3	8.6	Rep 3	16.1
average	11.9	average	14.7	average	15.8	average	17.2

20 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	118.7	Rep 1	140.1	Rep 1	172.3	Rep 1	155.3
Rep 2	185	Rep 2	156.5	Rep 2	162.4	Rep 2	131.3
Rep 3	146.7	Rep 3	148.3	Rep 3	118.7	Rep 3	127.4
average	150.1	average	148.3	average	151.1	average	138

30 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	12.1	Rep 1	6.3	Rep 1	9.8	Rep 1	15.8
Rep 2	7.4	Rep 2	24.3	Rep 2	11	Rep 2	7.4
Rep 3	20.3	Rep 3	12.1	Rep 3	9.8	Rep 3	6.3
average	13.2	average	14.2	average	10.2	average	9.8

30 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	133.4	Rep 1	172.3	Rep 1	148.3	Rep 1	111.2
Rep 2	172.2	Rep 2	143	Rep 2	272.3	Rep 2	148.3
Rep 3	186	Rep 3	119.8	Rep 3	142.1	Rep 3	127.4
average	163.9	average	145	average	187.6	average	129

Appendix 3.

Water source	W W
Date Collected	7/16/01
Date/time TSB inoc.	NA
Organism used	NA
Date/time tests inoc.	

Sample background E. coli levels (zero time)	
Rep 1	3.1
Rep 2	6.3
Rep 3	10.8
average	6.7

Zero time	
Rep 1	3.1
Rep 2	6.3
Rep 3	10.8
average	6.73

4 Degrees

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	4.1	Rep 1	1	Rep 1	3.1	Rep 1	0
Rep 2	5.2	Rep 2	3.1	Rep 2	2	Rep 2	0
Rep 3	3	Rep 3	1	Rep 3	2	Rep 3	0
average	4.1	average	1.7	average	2.36666667	average	0

10 Degrees

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	2	Rep 1	3.1	Rep 1	1	Rep 1	2
Rep 2	3.1	Rep 2	1	Rep 2	2	Rep 2	2
Rep 3	4.1	Rep 3	3.1	Rep 3	3.1	Rep 3	1
average	3.0	average	2.4	average	2.0	average	1.7

20 Degrees

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	3	Rep 1	0	Rep 1	1	Rep 1	1
Rep 2	1	Rep 2	0	Rep 2	1	Rep 2	0
Rep 3	1	Rep 3	1	Rep 3	1	Rep 3	2
average	1.7	average	0.3	average	1	average	1

30 Degrees

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	0	Rep 1	1	Rep 1	3.1	Rep 1	1
Rep 2	3.1	Rep 2	0	Rep 2	1	Rep 2	2
Rep 3	4.7	Rep 3	0	Rep 3	0	Rep 3	0
average	2.6	average	0.3	average	1.4	average	1

Appendix 4.

Water source St W
Date Collected 7/31/01

Sample background E. coli levels (zero time)

Date/time TSB inoc.	Rep 1	0
Organism used	Rep 2	0
Date/time tests inoc.	Rep 3	0

Low Concentration		average	0
Zero time			
Rep 1	12.2		
Rep 2	11.8		
Rep 3	9.7		
average	11.2333333		
High Concentration			
Zero time			
Rep 1	118.7		
Rep 2	151.5		
Rep 3	131.7		
average	133.966667		

4 degrees low concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	17.1	Rep 1	12.1	Rep 1	8.6	Rep 1	9.7
Rep 2	12.1	Rep 2	7.4	Rep 2	13.4	Rep 2	7.4
Rep 3	13.5	Rep 3	14.3	Rep 3	8.5	Rep 3	10.7
average	14.2	average	11.3	average	10.2	average	9.3

4 degrees high concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	161.6	Rep 1	93.3	Rep 1	93.3	Rep 1	66.3
Rep 2	108.1	Rep 2	118.7	Rep 2	117.8	Rep 2	72.7
Rep 3	137.4	Rep 3	116.9	Rep 3	93.3	Rep 3	68.9
average	135.7	average	109.6	average	101.5	average	69.3

10 degrees low concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	9.7	Rep 1	11.8	Rep 1	7.3	Rep 1	8.6
Rep 2	12.1	Rep 2	14.5	Rep 2	12.1	Rep 2	6.2
Rep 3	9.5	Rep 3	13.4	Rep 3	13.4	Rep 3	8.6
average	10.433	average	13.23	average	10.933	average	7.8

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	149.7	Rep 1	108.6	Rep 1	93.3	Rep 1	70.3
Rep 2	98.4	Rep 2	78.5	Rep 2	81.6	Rep 2	81.6
Rep 3	108.6	Rep 3	135.4	Rep 3	108.6	Rep 3	64.4
average	118.9	average	107.5	average	94.5	average	72.1

20 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	14.6	Rep 1	12	Rep 1	21.8	Rep 1	1
Rep 2	8.5	Rep 2	9.8	Rep 2	10.8	Rep 2	0
Rep 3	12.2	Rep 3	10.9	Rep 3	4.1	Rep 3	2
average	11.8	average	10.9	average	12.2	average	1

20 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	135.4	Rep 1	53	Rep 1	68.3	Rep 1	29.9
Rep 2	114.5	Rep 2	83.6	Rep 2	54.6	Rep 2	26.2
Rep 3	145	Rep 3	71.7	Rep 3	81.3	Rep 3	28.8
average	131.6	average	69.4	average	68.1	average	28.3

30 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	9.8	Rep 1	5.2	Rep 1	1	Rep 1	0
Rep 2	8.6	Rep 2	3	Rep 2	0	Rep 2	0
Rep 3	9.8	Rep 3	11	Rep 3	0	Rep 3	0
average	9.4	average	6.4	average	1.3	average	0

30 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	85.5	Rep 1		Rep 1	4.1	Rep 1	0
Rep 2	74.9	Rep 2	16	Rep 2	0	Rep 2	1
Rep 3	110.6	Rep 3	7.3	Rep 3	0	Rep 3	0
average	90.3333333	average	11.65	average	1.36666667	average	0.3333333

Appendix 5.

Water source	So W		
Date Collected	10/31/01		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	10/30/01	Rep 1	0
Organism used	E. coli	Rep 2	0
Date/time tests inoc.	10/31/01	Rep 3	0
Low Conc.		average	0

Zero time	
Rep 1	8.6
Rep 2	8.6
Rep 3	10.9
average	9.4
High Conc.	

Zero time	
Rep 1	67.6
Rep 2	84.4
Rep 3	99.0
average	83.7

4 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	7.4	Rep 1	5.2	Rep 1	6.3	Rep 1	4.1
Rep 2	4.1	Rep 2	8.6	Rep 2	5.2	Rep 2	3.1
Rep 3	3	Rep 3	7.4	Rep 3	9.8	Rep 3	6.3
average	4.8	average	7.1	average	7.1	average	4.5

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	68.3	Rep 1	101.7	Rep 1	56.5	Rep 1	47.2
Rep 2	65.7	Rep 2	76.6	Rep 2	63.8	Rep 2	73.3
Rep 3	55.6	Rep 3	61.7	Rep 3	64.4	Rep 3	69.7
average	63.2	average	80	average	61.6	average	63.4

10 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	11.0	Rep 1	1	Rep 1	3.1	Rep 1	2
Rep 2	9.7	Rep 2	7.4	Rep 2	2	Rep 2	7.4
Rep 3	8.5	Rep 3	3	Rep 3	4.1	Rep 3	1
average	9.7	average	3.8	average	3.07	average	3.5

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	110.0	Rep 1	82.3	Rep 1	35.4	Rep 1	19.7
Rep 2	76.3	Rep 2	74.9	Rep 2	34.1	Rep 2	22.6
Rep 3	66.3	Rep 3	71.7	Rep 3	60.1	Rep 3	29.4
average	84.2	average	76.3	average	43.2	average	23.9

20 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	5.2	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	7.4	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	5.2	Rep 3	1	Rep 3	0	Rep 3	0
average	5.9	average	0.3	average	0	average	0

20 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	72.2	Rep 1	2	Rep 1	1	Rep 1	0
Rep 2	57.1	Rep 2	10.8	Rep 2	0	Rep 2	0
Rep 3	52.0	Rep 3	3.1	Rep 3	0	Rep 3	0
average	60.4	average	5.3	average	0.3	average	0

30 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	3.1	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	4.1	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	2.0	Rep 3	0	Rep 3	0	Rep 3	0
average	3.1	average	0	average	0	average	0

30 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	55.6	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	52.1	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	39.9	Rep 3	0	Rep 3	0	Rep 3	0
average	49.2	average	0	average	0	average	0

Appendix 6

Water source	Em W
Date Collected	12/12/01
Date/time TSB inoc.	12/11 12pm
Organism used	E. coli
Date/time tests inoc.	12/12 10am
Low Conc.	

Sample background E. coli levels (zero time)

Rep 1	0
Rep 2	0
Rep 3	0
average	0

Zero time

Rep 1	7.4
Rep 2	8.5
Rep 3	4.1
average	6.7

High Concentration Zero time

Rep 1	45.7
Rep 2	49.6
Rep 3	66.3
average	53.9

4 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	9.8	Rep 1	1	Rep 1	3.1	Rep 1	8.6
Rep 2	9.8	Rep 2	4.1	Rep 2	2	Rep 2	5.1
Rep 3	9.7	Rep 3	4.1	Rep 3	3	Rep 3	4.1
average	9.8	average	3.1	average	2.7	average	5.9

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	60.1	Rep 1	54.7	Rep 1	50.4	Rep 1	55.6
Rep 2	52.5	Rep 2	66.9	Rep 2	52.8	Rep 2	58.3
Rep 3	82	Rep 3	54.6	Rep 3	48.7	Rep 3	56.6
average	64.9	average	58.7	average	50.6	average	56.8

10 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	10.9	Rep 1	3.1	Rep 1	3	Rep 1	3.1
Rep 2	6.3	Rep 2	4.1	Rep 2	1	Rep 2	7.4
Rep 3	16.1	Rep 3	5.2	Rep 3	6.3	Rep 3	0
average	11.1	average	4.1	average	3.4	average	3.5

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	49.5	Rep 1	54.7	Rep 1	46.4	Rep 1	41.3
Rep 2	82.3	Rep 2	59.1	Rep 2	50.4	Rep 2	42.5
Rep 3	62.4	Rep 3	68.9	Rep 3	57.3	Rep 3	42.2
average	64.7	average	60.9	average	51.4	average	42

20 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	8.6	Rep 1	3.1	Rep 1	2	Rep 1	0
Rep 2	8.6	Rep 2	1	Rep 2	2	Rep 2	0
Rep 3	12.1	Rep 3	5.2	Rep 3	0	Rep 3	0
average	9.8	average	3.1	average	1.3	average	0

20 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	62.2	Rep 1	48	Rep 1	13.2	Rep 1	2
Rep 2	48.7	Rep 2	35.4	Rep 2	10.8	Rep 2	0
Rep 3	65.1	Rep 3	58.8	Rep 3	13.2	Rep 3	1
average	58.7	average	47.4	average	12.4	average	1

30 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	2	Rep 1	0	Rep 1	0	Rep 1	0	
Rep 2	7.4	Rep 2	0	Rep 2	0	Rep 2	0	
Rep 3	9.8	Rep 3	0	Rep 3	0	Rep 3	0	
average	6.4	average	0	average	0	average	0	
30 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	47.1	Rep 1	0	Rep 1	0	Rep 1	0	
Rep 2	52.1	Rep 2	0	Rep 2	0	Rep 2	0	
Rep 3	43.2	Rep 3	0	Rep 3	0	Rep 3	0	
average	47.5	average	0	average	0	average	0	

Appendix 7.

Water source	Oetf W		
Date Collected	2/5/02		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	2/4 1:30pm	Rep 1	0
Organism used	E coli	Rep 2	0
Date/time tests inoc.	2/5/02 10am	Rep 3	0
Low Conc.		average	0
Zero time			
Rep 1	10.9		
Rep 2	9.8		
Rep 3	5.1		
average	8.6		
High Conc.			
Zero time			
Rep 1	90.7		
Rep 2	90.7		
Rep 3	81.3		
average	87.7		

4 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	12	Rep 1	10.9	Rep 1	2	Rep 1	4.1	
Rep 2	15.8	Rep 2	1	Rep 2	11	Rep 2	2	
Rep 3	13.1	Rep 3	5.2	Rep 3	6.3	Rep 3	2	
average	13.6	average	5.7	average	6.4	average	2.7	
4 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	129.6	Rep 1	166.4	Rep 1	88.2	Rep 1	59.1	
Rep 2	101.4	Rep 2	101.7	Rep 2	73.3	Rep 2	51.2	
Rep 3	150	Rep 3	113	Rep 3	78.5	Rep 3	39.3	
average	127	average	127.0	average	80	average	49.9	
10 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	6.2	Rep 1	4.1	Rep 1	4.1	Rep 1	1	
Rep 2	13.1	Rep 2	7.4	Rep 2	5.2	Rep 2	2	
Rep 3	5.2	Rep 3	11.9	Rep 3	5.2	Rep 3	8.6	
average	8.2	average	7.8	average	4.8	average	3.9	
10 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	104.3	Rep 1	59.4	Rep 1	54.7	Rep 1	8.6	
Rep 2	98.7	Rep 2	45.7	Rep 2	24.3	Rep 2	16	
Rep 3	59.4	Rep 3	67.7	Rep 3	33.5	Rep 3	18.3	
average	87.5	average	57.6	average	37.5	average	14.3	
20 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	16.1	Rep 1	2	Rep 1	0	Rep 1	0	
Rep 2	4.1	Rep 2	1	Rep 2	0	Rep 2	0	
Rep 3	16	Rep 3	1	Rep 3	0	Rep 3	0	
average	12.1	average	1.3	average	0	average	0	

20 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	82.3	Rep 1	13.2	Rep 1	0	Rep 1	0
Rep 2	107.6	Rep 2	29.8	Rep 2	0	Rep 2	0
Rep 3	101.9	Rep 3	13.5	Rep 3	0	Rep 3	0
average	97.3	average	18.8	average	0	average	0

30 degrees low concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	11	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	5.2	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	16.9	Rep 3	0	Rep 3	0	Rep 3	0
average	11.0	average	0	average	0	average	0

30 degrees high concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	59.4	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	60.5	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	61.3	Rep 3	0	Rep 3	0	Rep 3	0
average	60.4	average	0	average	0	average	0

Appendix 8.

Water source	Cp W
Date Collected	2/5/02
Date/time TSB inoc.	2/4 12pm
Organism used	E coli
Date/time tests inoc.	2/5 10am
Low Conc.	

Sample background E. coli levels (zero time)	
Rep 1	0
Rep 2	0
Rep 3	0
average	0

Zero time	
Rep 1	10.9
Rep 2	9.8
Rep 3	5.1
average	8.6

High Conc.

Zero time

Rep 1	90.7
Rep 2	90.7
Rep 3	81.3
average	87.6

4 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	11	Rep 1	1	Rep 1	1	Rep 1	0
Rep 2	6.3	Rep 2	5.2	Rep 2	3.1	Rep 2	3.1
Rep 3	5.2	Rep 3	5.2	Rep 3	3.1	Rep 3	3
average	7.5	average	3.8	average	2.4	average	2.0

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	101.7	Rep 1	56.5	Rep 1	21.6	Rep 1	16
Rep 2	103.9	Rep 2	38.4	Rep 2	23.1	Rep 2	7.4
Rep 3	93.3	Rep 3	51.2	Rep 3	21.6	Rep 3	14.5
average	99.6	average	48.7	average	22.1	average	12.6

10 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	11	Rep 1	1	Rep 1	0	Rep 1	0
Rep 2	10.8	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	6.3	Rep 3	0	Rep 3	0	Rep 3	0
average	9.4	average	0.3333333	average	0	average	0

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	75.9	Rep 1	11	Rep 1	1	Rep 1	0
Rep 2	60.1	Rep 2	9.8	Rep 2	3.1	Rep 2	0
Rep 3	65	Rep 3	9.8	Rep 3	0	Rep 3	na
average	67	average	10.2	average	1.4	average	0

20 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	3.1	Rep 1	0	Rep 1	0	Rep 1	0	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	0	
Rep 3	10	Rep 3	0	Rep 3	0	Rep 3	0	
average	4.4	average	0	average	0	average	0	
20 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	41.3	Rep 1	0	Rep 1	0	Rep 1	0	
Rep 2	18.9	Rep 2	0	Rep 2	0	Rep 2	0	
Rep 3	31.7	Rep 3	0	Rep 3	0	Rep 3	0	
average	30.6	average	0	average	0	average	0	
30 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	0	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	0	
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	0	
average	0	average	0	average	0	average	0	
30 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	0	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	0	
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	na	
average	0	average	0	average	0	average	0	

Appendix 9.

Water source	N W		
Date Collected	3/29/02		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	na	Rep 1	2
Organism used	na	Rep 2	0

Date/time tests inoc.	3/29/02 1pm	Rep 3	0
		average	0.666667

4 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	2	Rep 1	1	Rep 1	0	Rep 1	2
Rep 2	2	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	0
average	1.3	average	0.	average	0	average	0.7

10 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	1	Rep 1	0	Rep 1	0	Rep 1	1
Rep 2	1	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	1	Rep 3	0	Rep 3	0	Rep 3	0
average	1	average	0	average	0	average	0.3

20 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	0	Rep 2	0	Rep 2	1	Rep 2	0
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	0
average	0	average	0	average	0.3	average	0

30 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	1	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	1	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	0
average	0.7	average	0	average	0	average	0

Appendix 10.

Water source	V W
Date Collected	3/29/02
Date/time TSB inoc.	na
Organism used	na
Date/time tests inoc.	3/29/02 1pm

Sample background E. coli levels (zero time)

Rep 1	0
Rep 2	0
Rep 3	0
average	0

Stopped test due to no results at 48 hours

4 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	
average	0	average	0	average	0	average	0

10 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	
average	0	average	0	average	0	average	0

20 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	
average	0	average	0	average	0	average	0

30 degrees

8 hours	MPN/100 ml	24 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	0	Rep 1	0	Rep 1	0	Rep 1	
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	
Rep 3	0	Rep 3	0	Rep 3	0	Rep 3	
average	0	average	0	average	0	average	0

Appendix 11.

Water source	Blcw W		Note: wrong times and temps run
Date Collected	4/15/02		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	4/14 12pm	Rep 1	0
Organism used	E. coli	Rep 2	0
Date/time tests inoc.	4/15 11am	Rep 3	0
Low Conc.		average	0

Zero time	
Rep 1	7.4
Rep 2	8.5
Rep 3	5.2
average	7.0

High Conc.

Zero time	
Rep 1	83.3
Rep 2	139.6
Rep 3	101.4
average	108.1

4 degrees low concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	13.2	Rep 1	12.1	Rep 1	8.6	Rep 1	4.1
Rep 2	9.8	Rep 2	6.3	Rep 2	9.7	Rep 2	9.4
Rep 3	12.1	Rep 3	6.3	Rep 3	6.3	Rep 3	5.2
average	11.7	average	11.21	average	8.2	average	6.2

4 degrees high concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	105	Rep 1	133.4	Rep 1	98.7	Rep 1	73.8
Rep 2	78.9	Rep 2	62.4	Rep 2	75.2	Rep 2	75.4
Rep 3	66.9	Rep 3	75.4	Rep 3	101.7	Rep 3	88.9
average	83.6	average	90.4	average	91.9	average	79.4

10 degrees low concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	8.5	Rep 1	9.7	Rep 1	7.3	Rep 1	9.7
Rep 2	7.4	Rep 2	5.2	Rep 2	3.1	Rep 2	3
Rep 3	6.3	Rep 3	13.5	Rep 3	8.6	Rep 3	6.3
average	7.4	average	9.5	average	6.3	average	6.3

10 degrees high concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	93.3	Rep 1	103.9	Rep 1	110	Rep 1	78.5
Rep 2	81.3	Rep 2	93.2	Rep 2	83.3	Rep 2	78.8
Rep 3	71.2	Rep 3	85.5	Rep 3	77.6	Rep 3	75.4
average	81.9	average	94.2	average	90.3	average	77.6

20 degrees low concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	7.4	Rep 1	11	Rep 1	7.4	Rep 1	3.1
Rep 2	5.2	Rep 2	12.2	Rep 2	5.2	Rep 2	3.1
Rep 3	11	Rep 3	11	Rep 3	5.2	Rep 3	6.3
average	7.9	average	11.4	average	5.9	average	4.2

20 degrees high concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	46.5	Rep 1	90.7	Rep 1	82	Rep 1	61.3
Rep 2	86	Rep 2	68.4	Rep 2	60.1	Rep 2	51.2
Rep 3	67.6	Rep 3	63.7	Rep 3	77.6	Rep 3	40.4
average	66.7	average	74.3	average	73.2	average	51.0

35 degrees low concentration

8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	1	Rep 1	0	Rep 1	0	Rep 1	0
Rep 2	0	Rep 2	0	Rep 2	0	Rep 2	0
Rep 3	0	Rep 3	0	Rep 3	3.1	Rep 3	0
average	0.3	average	0	average	0	average	0

35 degrees high concentration								
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1	1	Rep 1	2	Rep 1	0	Rep 1	1
Rep 2	1	Rep 2	0	Rep 2	2	Rep 2	0
Rep 3	2	Rep 3	7.4	Rep 3	2	Rep 3	2
average	1.3	average	3.13	average	1.3	average	1

Appendix 12.

Water source	Bumc W		
Date Collected	4/23/02		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	4/22 12pm	Rep 1	0
Organism used	E. coli	Rep 2	0
Date/time tests inoc.	4/23 11am	Rep 3	0
Low Conc.		average	0

Zero time	
Rep 1	6.3
Rep 2	14.8
Rep 3	24
average	15.0

High Conc.

Zero time	
Rep 1	125.9
Rep 2	131.3
Rep 3	95.9
average	117.7

4 degrees low concentration								
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1		Rep 1		Rep 1		Rep 1	
Rep 2		Rep 2		Rep 2		Rep 2	
Rep 3		Rep 3		Rep 3		Rep 3	
average	0	average	0	average	0	average	0

4 degrees high concentration								
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1		Rep 1		Rep 1		Rep 1	
Rep 2		Rep 2		Rep 2		Rep 2	
Rep 3		Rep 3		Rep 3		Rep 3	
average	0	average	0	average	0	average	0
10 degrees low concentration							
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1		Rep 1		Rep 1		Rep 1	
Rep 2		Rep 2		Rep 2		Rep 2	
Rep 3		Rep 3		Rep 3		Rep 3	
average	0	average	0	average	0	average	0
10 degrees high concentration							
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1		Rep 1		Rep 1		Rep 1	
Rep 2		Rep 2		Rep 2		Rep 2	
Rep 3		Rep 3		Rep 3		Rep 3	
average	0	average	0	average	0	average	0
20 degrees low concentration							
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1		Rep 1		Rep 1		Rep 1	
Rep 2		Rep 2		Rep 2		Rep 2	
Rep 3		Rep 3		Rep 3		Rep 3	
average	0	average	0	average	0	average	0
20 degrees high concentration							
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml
Rep 1		Rep 1		Rep 1		Rep 1	
Rep 2		Rep 2		Rep 2		Rep 2	
Rep 3		Rep 3		Rep 3		Rep 3	
average	0	average	0	average	0	average	0

35 degrees low concentration								
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	
Rep 1		Rep 1		Rep 1		Rep 1		
Rep 2		Rep 2		Rep 2		Rep 2		
Rep 3		Rep 3		Rep 3		Rep 3		
average	0	average	0	average	0	average		0
35 degrees high concentration								
8 hours	MPN/100 ml	24 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	
Rep 1		Rep 1		Rep 1		Rep 1		
Rep 2		Rep 2		Rep 2		Rep 2		
Rep 3		Rep 3		Rep 3		Rep 3		
average	0	average	0	average	0	average		0

Appendix 13.

Water source	O W		
Date Collected	4/26/02		
		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	4/25 12PM	Rep 1	0
Organism used	E coli	Rep 2	0
Date/time tests inoc.	4/26 8AM	Rep 3	0
Low Conc.		average	0
Zero time			
Rep 1	12		
Rep 2	4.1		
Rep 3	9.5		
average	8.533		
High Conc.			
Zero time			
Rep 1	54.7		
Rep 2	90.6		
Rep 3	71.7		
average	72.3		

4 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	6.3	Rep 1	7.3	Rep 1	14.8	Rep 1	4.1
Rep 2	5.2	Rep 2	10.8	Rep 2	10.9	Rep 2	5.1
Rep 3	11	Rep 3	13.5	Rep 3	9.7	Rep 3	8.6
average	7.5	average	10.5	average	11.8	average	5.9

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	63.1	Rep 1	66.9	Rep 1	83.9	Rep 1	77.6
Rep 2	65	Rep 2	98.7	Rep 2	58.1	Rep 2	70.3
Rep 3	73.3	Rep 3	69.7	Rep 3	81.3	Rep 3	69.1
average	67.1	average	78.4	average	74.4	average	72.3

10 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	10.9	Rep 1	12.2	Rep 1	5.2	Rep 1	7.4
Rep 2	4.1	Rep 2	8.6	Rep 2	9.7	Rep 2	9.8
Rep 3	13.5	Rep 3	6.3	Rep 3	6.3	Rep 3	4.1
average	9.5	average	9.0	average	7.1	average	7.1

10 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	79.8	Rep 1	111.9	Rep 1	59.1	Rep 1	81.3
Rep 2	64.4	Rep 2	95.9	Rep 2	65	Rep 2	74.9
Rep 3	83	Rep 3	76.6	Rep 3	68.9	Rep 3	79.8
average	75.7	average	94.8	average	64.3	average	78.7

20 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	8.5	Rep 1	21.1	Rep 1	6.3	Rep 1	2
Rep 2	12.2	Rep 2	24	Rep 2	3.1	Rep 2	5.2
Rep 3	8.6	Rep 3	21.3	Rep 3	7.4	Rep 3	4.1
average	9.8	average	22.1	average	5.6	average	3.8

20 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	129.6	Rep 1	75.4	Rep 1	62.4	Rep 1	25.9
Rep 2	110.6	Rep 2	101.2	Rep 2	70.6	Rep 2	35.9
Rep 3	83.3	Rep 3	56.5	Rep 3	65	Rep 3	27.2
average	108	average	77.7	average	66	average	30

30 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	6.3	Rep 1	14.6	Rep 1	9.5	Rep 1	5.2
Rep 2	4.1	Rep 2	17.3	Rep 2	4.1	Rep 2	2
Rep 3	17.3	Rep 3	17.5	Rep 3	7.4	Rep 3	0
average	9.2	average	16.57	average	7	average	2.4

30 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	90.6	Rep 1	75.4	Rep 1	31.3	Rep 1	23.8
Rep 2	102.2	Rep 2	59.4	Rep 2	60.9	Rep 2	22.8
Rep 3	84.2	Rep 3	88.4	Rep 3	52.9	Rep 3	19.4
average	92.3	average	74.4	average	48.4	average	22

Appendix 14.

Water source	Lwdp
Date Collected	6/22/01
Date/time TSB inoc.	6/21/01 15:00
Organism used	E coli
Date/time tests inoc.	6/22 8:30am
Low Conc.	

Sample background E. coli levels (zero time)

Rep 1	2
Rep 2	0
Rep 3	1
average	1

Zero time

Rep 1	8.6
Rep 2	9.8
Rep 3	12.2
average	10.2

High Conc.

Zero time

Rep 1	118.7
Rep 2	107.6
Rep 3	110.6
average	112.3

4 degrees low concentration

8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	8.6	Rep 1	10.9	Rep 1	15.8	Rep 1	13.5
Rep 2	6.3	Rep 2	12.1	Rep 2	17.3	Rep 2	10.9
Rep 3	12.1	Rep 3	14.5	Rep 3	13.5	Rep 3	12.2
average	9	average	12.5	average	15.5	average	12.2

4 degrees high concentration

8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	115.3	Rep 1	95.9	Rep 1	113	Rep 1	122.2
Rep 2	122.3	Rep 2	107.6	Rep 2	128.1	Rep 2	86.2
Rep 3	88.2	Rep 3	125	Rep 3	107.6	Rep 3	135.4
average	108.6	average	109.5	average	116.2	average	114.6

10 degrees low concentration

8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	13.5	Rep 1	13.4	Rep 1	9.6	Rep 1	11
Rep 2	13.4	Rep 2	7.4	Rep 2	6.3	Rep 2	5.2
Rep 3	14.6	Rep 3	16.1	Rep 3	16	Rep 3	13.4
average	13.8	average	12.3	average	10.6	average	9.87

10 degrees high concentration

8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	123.6	Rep 1	101.4	Rep 1	98.5	Rep 1	90.9
Rep 2	98.7	Rep 2	119.8	Rep 2	131.3	Rep 2	115.3
Rep 3	98.5	Rep 3	86.7	Rep 3	118.7	Rep 3	88.6
average	107	average	103	average	116.	average	98.3

20 degrees low concentration								
8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	14.5	Rep 1	16	Rep 1	18.7	Rep 1	13.2	
Rep 2	9.8	Rep 2	13.4	Rep 2	19.7	Rep 2	9.8	
Rep 3	14.5	Rep 3	13.4	Rep 3	12.1	Rep 3	7.3	
average	12.9	average	14.3	average	16.8	average	10.1	
20 degrees high concentration								
8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	113.7	Rep 1	113.4	Rep 1	116.9	Rep 1	111.2	
Rep 2	156.5	Rep 2	93.2	Rep 2	119.8	Rep 2	112.4	
Rep 3	106.3	Rep 3	87.6	Rep 3	71.2	Rep 3	98.8	
average	125.5	average	98.1	average	102.6	average	107	
30 degrees low concentration								
8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	18.5	Rep 1	13.4	Rep 1	13.2	Rep 1	10.9	
Rep 2	17.1	Rep 2	14.8	Rep 2	16	Rep 2	9.8	
Rep 3	21.3	Rep 3	25.6	Rep 3	16.1	Rep 3	17.1	
average	19.0	average	17.9	average	15.1	average	12.6	
30 degrees high concentration								
8 hours	MPN/100ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml	
Rep 1	119.8	Rep 1	139.6	Rep 1	111.9	Rep 1	104.6	
Rep 2	83.3	Rep 2	118.7	Rep 2	79.8	Rep 2	123.6	
Rep 3	129.6	Rep 3	113.7	Rep 3	129.1	Rep 3	88.2	
average	110.9	average	124	average	107	average	105	

Appendix 15.

Water source	Lm
Date Collected	5/15/01

		Sample background E. coli levels (zero time)	
Date/time TSB inoc.	5/14/01 2 p.m.	Rep 1	1
Organism used	E. coli	Rep 2	0
Date/time tests inoc.	5/15/01 10 a.m.	Rep 3	1
Low Conc.		average	0.666667

Zero time	
Rep 1	14.3
Rep 2	18.9
Rep 3	12.2
average	15.1
High Conc.	
Zero time	
Rep 1	143
Rep 2	129.6
Rep 3	133.4
average	135.

4 degrees low concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	7.4	Rep 1	17.3	Rep 1	11	Rep 1	14.6
Rep 2	15.6	Rep 2	17.3	Rep 2	13.5	Rep 2	13.5
Rep 3	9.8	Rep 3	12.2	Rep 3	11	Rep 3	8.5
average	10.9	average	15.6	average	11.8	average	12.2

4 degrees high concentration

8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	93.2	Rep 1	122.3	Rep 1	116.2	Rep 1	95.9
Rep 2	101.4	Rep 2	160.7	Rep 2	115.3	Rep 2	191.8
Rep 3	101.7	Rep 3	81.3	Rep 3	156.5	Rep 3	101.4
average	98.8	average	121.4	average	129	average	130

10 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	14.6	Rep 1	8.5	Rep 1	15.5	Rep 1	9.7
Rep 2	8.6	Rep 2	8.6	Rep 2	17.1	Rep 2	17.1
Rep 3	15.6	Rep 3	9.8	Rep 3	10.9	Rep 3	11
average	12.9	average	9.0	average	14.5	average	12.6
10 degrees high concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	146.7	Rep 1	120.1	Rep 1	65	Rep 1	123.6
Rep 2	145	Rep 2	123.6	Rep 2	172.3	Rep 2	125.9
Rep 3	111.9	Rep 3	0	Rep 3	131.3	Rep 3	98.7
average	135	average	81	average	123	average	116

20 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	17.9	Rep 1	9.8	Rep 1	8.6	Rep 1	6.3
Rep 2	9.7	Rep 2	13.4	Rep 2	7.4	Rep 2	5.2
Rep 3	14.6	Rep 3	4.1	Rep 3	16	Rep 3	5.2
average	14.1	average	9.1	average	11	average	5.6
20 degrees high concentration							
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	159.7	Rep 1	133.4	Rep 1	101	Rep 1	46.4
Rep 2	101.4	Rep 2	110.6	Rep 2	93.2	Rep 2	41.3
Rep 3	101.7	Rep 3	96	Rep 3	83.3	Rep 3	48.7
average	121	average	113	average	92.5	average	45.5

30 degrees low concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	16.1	Rep 1	6.3	Rep 1	12.2	Rep 1	4.1
Rep 2	17.1	Rep 2	11	Rep 2	16	Rep 2	1
Rep 3	20.1	Rep 3	18.3	Rep 3	7.4	Rep 3	1
average	17.8	average	11.9	average	11.9	average	2.0

30 degrees high concentration								
8 hours	MPN/100 ml	30 hours	MPN/100 ml	48 hours	MPN/100 ml	72 hours	MPN/100 ml
Rep 1	135.4	Rep 1	152.9	Rep 1	64.5	Rep 1	2419.2
Rep 2	101.4	Rep 2	116.9	Rep 2	115.3	Rep 2	27.2
Rep 3	96	Rep 3	156.5	Rep 3	101.7	Rep 3	14.6
average	110.9	average	142.1	average	93.8	average	820.