TRENDS OF COLIFORM BACTERIA IN HEMLOCK BROOK UPSTREAM OF MARGARET LINDLEY PARK

Independent Research

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I. Introduction

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three years, high levels of coliform bacteria a probl**em** in the pond in Williamstown's Margaret Counts of total coliform in the pond have reached 150,000 organisms per 100 ml, and counts in lower Hemlock Brook which feeds the pond have been as hing as 25,000 organisms ml. (Muzyka, 1990) These levels far the Massachusetts criteria for bathing waters -As a result, for periods over the last 100 ml. three summers, the pond, which is used by Williamstown as an pool, you with say outdoor swimming hole, has been either closed for swimming or extensively chlorinated. The pond in Margaret Lindley not naturally filled by the brook; during the summer months, the

of the brook is diverted to fill the pond.

the time it reaches Margaret Lindley Park, Hemlock Brook has drained approximately 3.75 square miles. Its Taconic Crest consist of three major tributaries: Treadwell Hollow Brook, Berlin Mountain Brook, and southern flowing past the Mt. Carmel Novitiate. (See Maps Treadwell Hollow Brook and Berlin Mountain Brook elevation of 1080 feet where Treadwell Hollow Road meets Berlin Road. The Mt. Carmel Branch meets this brook just west of intersection of Oblong and Torey Woods Road, at about 900 feet.

coliform family consists of small, forming, facultative anaerobic, gram-negative,

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rod-shaped bacteria. Escherichia coli, known as fecal coliform, originates exclusively in the intestines of humans warm-blooded animals, while Aerobacter aerogenes. coliform, may be found naturally in soil. However, the grain species is also abundant in human and animal waste. Although these species themselves are not hazardous, high numbers of coliform bacteria indicate polluted waters which would most likely contain other, less easily detected, hazardous bacteria. Consequently, their concentration is used to evaluate bacteriological quality of water.

Previous coliform testing in Hemlock Brook conducted in the spring of 1989 by Williams students Elizabeth Rogers and Rebecca Foster was unsuccessful in identifying the source of the summer coliform contamination. This field survey found (very) low levels of total coliform - all were below 70 organisms per 100 ml - and moderate to low levels of fecal coliform (See Table 2, Map 3). A more extensive survey, conducted in November of 1988 by Williamstown Environmental Inspector Charles Kochenour, found levels of total coliform exceeding bathing standards (1000 organisms per 100 ml) downstream from 395 Berlin Road, and 147 and 149 Oblong Road. (Table 3, Map 2) However, in both studies, levels in Margaret Lindley Park were low, and it was unclear whether these relatively slightly elevated levels upstream would be sufficient to elevate pond levels during summer months.

Consequently, the only way to pimpoint the exact source of the coliform bacteria is through extensive summer sampling when levels of coliform are known to be high in Margaret Lindley Park.

This study sought to identify locational trends in coliform levels, to investigate the relationships between coliform and nitrate and chloride ions which are generally associated with human-generated pollution, and to determine if relationships existed between coliform and rainfall and temperature.

II. Methods

were collected on the morning of March 18, 1991 under light rain with temperatures in the upper 30's. Fifteen water samples were collected from locations in Hemlock Brook and its tributaries. Three of these locations were upstream from the levels of human influence - locations TW2, TW5, and 02, Explant ton and four were located in Margaret Lindley Park.(See Location Ke) 2) Samples were tested (membrane filter method) for total and fecal coliform bacteria, and levels of chloride were tested with the ion chromatograph. Samples were nitrate collected again at the same locations late in the afternoon on After 11, 1991. two days of moderate rain, conditions with temperatures in the 60's prevailed. samples were also tested for levels of coliform, nitrate, and chloride.

Total coliform cultures were conducted using 10:100 Fecal coliform cultures dilutions. were run using undiluted and chloride samples. Nitrate tests were run on the ion chromatograph with undiluted samples, and 10:100 dilutions test for chloride in approximately 60% of the samples

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whose chloride was above 3 mg/l, exceeding the instrument's standard.

III. General Results and Discussion

Coliform levels measured this spring were dramatically lower than levels at the same locations during the summer. Coliform levels tested by the town in the spring of 1989 were also lower aways than summer levels, suggesting that this is a seasonal variation. See Table 1) At all locations, total coliform was below the limit for bathing standards. Several contributing factors could be responsible for this. First, the water temperature at all sample locations on March 18, 1991 was 2 degrees Celsius. water temperatures may inhibit the survival of coliform organisms, particularly fecal coliform, who are acclimated to a warm body temperature near 35 degrees Celsius. Also, spring melt flows and higher precipitation with less evaporation tend to increase the discharge of streams, so spring dilution would reduce even constant numbers of coliform, while lower volumes of effectively the would increase water in summer concentration. April levels were also low; the water temperature then was 8 degrees Celsius.

Nitrate levels were significantly higher in April than in March. March nitrate concentrations ranged from 2.29 mg/l at to 0.86 at TW5, with the average concentration being 1.59. April, the average rose to 2.51, with a high of 3.66 at 02 and a low of .74 at TW5. Most likely, this reflected an increase in

by lasteria, which ordised more No releasing

organic activity in plants which reduced more nitrogen, releasing more in soluble form into the streams. (Dethier, 1991)

The average chloride levels, however, decreased in April, from an average of 7.92 mg/l in March to 6.37. March had a broad range, from 21.1 mg/l at TW4 to 0.86 at TW5. Sixty percent of the March samples had over 9 mg/l of chloride, while the remaining 40% had less than 1.5 mg/l. The lack of intermediate levels suggests that the presence of large amounts of chloride is correlated to a definite source. All five of the lowest levels had one thing in common - none were downstream from a town road. Party of town roads are salted with sodium chloride during bad weather; spring rains, like the one falling on March 18, washing the sodium chloride into roadside streams, could at least be partially responsible for the elevated levels.

However, levels of chloride in April did reach 16.6 mg/l, and there were intermediate values, indicating that another chloride existed. Although most downstream concentration sites were unlikely that road salt could be entirely responsible for the chloride this late in the season. Most of the high concentration sites were downstream from houses, and chloride is known to be associated with human pollution. However, the second highest of chloride and another high level were found at TW4 and work whater were ... **INDO** TW2 respectively, sites which are upstream from human a lymbor. ... woultn't a lymbor which are upstream from human establishments, and it is difficult to establish the source of human this chloride.

IV. Coliform Relationships with Nitrate and Chloride

chloride levels were measured as indicators of and human influence to correlate with coliform levels. Nitrate, in particular, is associated with human and animal waste, the only source of fecal coliform. Although chloride was largely correlated with human presence, it is not related to levels of either fecal or total coliform (Fig. 1). Nitrate levels also showed no correlation with fecal or total coliform. (Fig 2) In addition, no correlation between nitrate and locational human presence existed - location TW2, with the third-highest March nitrate level of 1.86 mg/l, and a high April rate of 2.7 mg/l, (+ www.) (+ of human influence. Thus, any March levels below 1.86, and any April levels below 2.7, could not be conquisively attributed to human causes, since some natural phenomenon was elevating nitrate Preliminary research in forests on Mount Greylock has found evidence of forests bleeding nitrate in the spring. (Dethier, 1991) It is probable that this also occurs in the Mt. Berlin forests of the Hemlock Brook watershed and is affecting nitrate levels in the stream in the spring. At this time, at these levels, there is too much background nitrate for it to be used as a "tracer" for sewage contamination.

V. Locational Trends

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Although the levels of nitrate and chloride changed between March and April, the relative concentrations at specific locations generally did not, as an examination of the Berlin and Carmel Branch downstream progressions shows (Figs. 3 -10). The nitrate correlations between the two samplings in the Berlin Branch had a regression of 0.7829, and the correlation in the Carmel Branch had a regression of 0.9004. Chloride had similarly strong correlations with regressions of 0.8555 in the Berlin Branch and .9991 in the Carmel Branch.

levels of total coliform However. showed almost correlation between samplings: regressions were insignificant 0.0222 for the Berlin Branch, and 0.0337 Branch. This may mean that sources of total coliform are highly variant at this time of year, or it may reflect variance within the precision range of the total coliform test. To test Witeo (M) of of the method, three bottles water were Km collected consecutively from ML6 in April. Three tests for total UNDLICE conducted on each bottle, (1.e. 4-16 an deliteral) (Oll) results (Table 4) ranged in underested un what who suggesting that the lack of repeatability was due THOME ... OND HOPE testing method. This variance means that comparison of bacteria and locations within this range is only valid increase/decrease in coliform) occurs trend (ie. sharp samplings. (Since a high peak of 310 was detected March, the highest probable number, 160, rather than the average, (12., one widencounts was used for all graphs including this data point.)

Still, some of the variance may reflect actual changes. The significant increase in total coliform between March and April at TW2 was out of the precision range outlined above. Additionally, it was also accompanied by a significant increase in chloride, although in most other locations, the chloride level decreased. (Nitrate increased as well, but not more than the base increase noted at all of the other locations.) This suggests that some source of low-level coliform and chloride pollution which was not present in March entered the stream some time around April 18. A variety of "natural" explanations exist. The most probable one is that an animal (perhaps a deer or coyote) defecated in or near the stream and the coliform bacteria and chloride entered the stream, elevating levels.

Given the variance of the total coliform test, the only locational data which can be interpreted from the total coliform samplings is the presence of an increase in total coliform at ML6, TW6, and probably ML7.

The precision of fecal the coliform coliform, like although. repeatability with regressions of 0.0612 and 0.0589 in the Berlin and Carmel Branches, respectively. Still, this may be du varying sources. One trend did repeat in both of the samples. however. suggesting at least some pattern. Fecal coliform increased appreciably between TW1 and ML7 in the Berlin Branch.

From this survey, two locations emerge as possibly suspicious. Location ML7 had relatively high levels of total and fecal coliform in both samplings. Location ML6 had high levels

of total coliform in both samplings; however, its fecal coliform was low in March, and there was no fecal coliform at all in April.

VI. Relationships Between Coliform, Rainfall and Temperature

The single sampling site from Hemlock Brook precludes locational comparison for seeking the source of the coliform; however, it provides a constant, frequent source of data for comparison with precipitation, temperature, and levels in the pond.

In general, precipitation does not appear to be related to total coliform levels in Hemlock Brook. A three-day precipitation average of data from Hopkins Forest was plotted against total coliform at the Hemlock Brook site, both directly and on a time trend graph, to determine if high precipitation

wash bacteria out of soil, send manure runoff from farms into the stream, or elevate the water table, causing flushing of bacteria from septic leach fields. All of these would lead to higher coliform levels. Neither graph showed a significant correlation. If rain does bring additional bacteria into the stream, it dilutes the concentrations of the colonies to levels?

Also, precipitation does not appear to affect levels of fecal coliform in the brook (Fig. 13); however, it does appear to be related to levels of fecal coliform in the pond inlet. Fig 14 a close relationship between precipitation and fecal coliform entering the pond. Peaks in fecal coliform occur only with peaks of precipitation, although all precipitation peaks are accompanied by peaks in fecal coliform. Furthermore, the increases in fecal coliform at the inlet are not "preceded" in the brook by higher levels. It seems quite logical that rain increasing the volume of water in the brook could additional fecal coliform into the inlet, but it is difficult to envision this scenario with brook fecal coliform at lower levels than the inlet. Perhaps fecal coliform "resides" somewhere between the brook test site and the inlet and is flushed out without "registering" in the brook census. A thorough "flushing out" of the fecal coliform would also explain why closely following storms are not accompanied by high inlet coliform counts, a trend noted on the graph.

Water work of the

Long Long Temperature does not appear to have a significant impact on summer coliform fluctuations. (Fig. 13)

VII. Conclusions

- 1. Locational trends are difficult to identify in the recent surveys of Hemlock Brook. Low levels of coliform in the spring, and the lack of precision of the total coliform test make tracking sources difficult. The only suspicious source of high levels of both total and fecal coliform identified in this investigation was ML7, off Torey Woods Road 0.10 mile west of its intersection with Oblong Road.
- 2. Chloride and nitrate showed no relationship to coliform in the spring, although chloride may be an indicator of human acitvity in a location. Nitrate, at this time, shows no relationship with human presence. Therefore, it is a poor index of sewage/waste pollution. (A... wet to work Sewage)
- 3. In general, coliform fluctuations in the brook are not dependent on precipitation or temperature. Levels in the pond inlet do appear to show some correlation to precipitation perhaps because rain surgeS in the stream helps flush out a resident population.
- 4. Nitrate and chloride levels are not good indicators for tracking the entry of pollution, nor is precipitation a useful tool predict coliform levels. A conclusive determination of the source of the elevated coliform levels could be accomplished only through thorough sampling and coliform testing during the elevated levels in the summer.

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FIGURES

TRENDS OF COLIFORM BACTERIA
IN HEMLOCK BROOK
UPSTREAM OF MARGARET LINDLEY PARK





LOCATION KEY FOR COLIFORM WATER SAMPLING

- Tomey Woods Road, Northwest of intersection with Oblong Road, where the brook crosses from the west to east side of the road. Sample from west side of road.
- TW2 Torrey Woods/Bee Hill Road, 1 mile up (west) of junction of East Branch of Hemlock Brook with the Bee Hill Road Branch.
- TW3 On East Branch, north off Toney Woods Road at farm drive along stream. Sample from 4th bridege after pond, roughly 1600 feet from road.
- TW4 Just before junction of East Branch and Bee Hill Road Branch, from the East Branch
- TW5 Between TW1 and Oblong Road, a small brook crosses road from west to east side. Sample from west side.
- TW6 Junction of Torey Woods and Oblong Road, on lower side of culvert.
- O1 Where stream crosses Oblong Road, on downstream side.
- On Carmelite Property, just before corduroy section of road where an old road/path crosses the creek and curves up the hill. Sample location approximately 550 feet west of main building.
- ML1 Margaret Lindley Park, 1/2 way between the dam and the road.
- ML2 Margaret Lindley Park, just below little falls over quartzite, about 100 feet below the bridge.
- ML3 Margaret Lindley Park, from West Branch, just before merge with Bee Hill Road Branch.
- ML4 Margaret Lindley Park, from Bee Hill Road Branch, just before merge with West Branch.
- ML5 Margaret Lindley Park, 1/3 of the way from ML4 to ML2 at sharp curve where many dead branches are across.
- ML6 Oblong Road, about 200 feet upstream from culvert 01, by houses.
- ML7 Torey Woods Road, 1/10 mile east of intersection with Oblons Road, 20 feet downstream from pond.

SUMMARY OF SPRING '91 DATA

SITE	TOTAL COLIFORM		FECAL COLIFORM		NITRATE (mg/l)		CHLORID{E (mg/l)	
	MARCH	APRIL	MARCH	APRIL	MARCH	APRIL	MARCH	APRIL
TW1	50	0	1	0	1.34	2.3	10.2	7.9
TW2	10	190	0	1	1.85	2.7	0.65	4.8
TW3	50	90	1	0	1.17	2.08	20.5	16.6
TW4	0	50	1	0	1.09	2.06	21.1	14.3
TW5	30	30	1	0	0.86	0.74	0.44	0.5
TW6	80	120	3	2	1.22	2.28	9.3	8.4
01	20	80	4	0	2.21	3.47	1.13	0.69
02	80	70	2	1	2.29	3.66	0.55	0.35
ML1	50	170	0	2	1.37	2.4	10.1	8.2
ML2	70	90	10	2	1.63	2.37	9.8	8
ML3	20	70	3	3	2.24	3.28	1.5	1.2
ML4	20	120	3	10	1.38	2.26	12.7	7.9
ML5	40	90	6	4	1.56	2.43	9.5	8
ML6	310	*	3	0	2.34	3.49	0.48	0.4
ML7	200	110	6	7	1.25	2.15	10.9	E.8

+ =

Table #2 - Data (See Map 3)

	Fecal	Total			Standard	EPA
<u>Site#</u>	Coliform#	Coliform#	Cond.	<u>Hq</u>	Alkalinity	Alkalinity
1	0,4	4	106	7.26	12.0	11.0
2	1,2	2	100	7.14	17.5	15.0
3 -	1	34	109	7.10	9.5	7.5
4	2	65	106	7.05	8.0	4.5
5	0	11	45	7.04	9.0	7.0
6	0	7	42	6.96	13.0	8.0
7	0	0	38	6.84	3.5	3.0
8	0,4	6	153	7.69	56.0	55.0
9		8	60	7.21	10.0	8.0
10	2	1	166	6.92	7.0	5.0
T1*	. 5	42	62	7.05	12.0	10.0
2*	20	56	42	7.01	11.0	9.0
3*	2	25	121	6.73	7.5	5.0

It should be noted that sites showing two fecal coliform counts had two separate samples taken about a week apart. In addition, sites 3 and 10 had fecal tests done on 48-hour old samples. The fecal test for site #9 drowned and was unable to be redone; there is a high probability that the results would have indicated very low, if any, fecal coliform levels.

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Hemlock Brook Survey

November 15 & 16, 1988

A survicity of Coliform colonies found upstream and downstream of each sample point.

Location	ຂວຸປັ	tream	Downs	tream
	Total	Pecal	<u>Total</u>	Tecal
1. Hargaret Lindley Park	27	C	710	2
2. 150 Torrey Toods Road (Roger Gould)	960	3	360	73
3. 190 Torrey Joods Road (James Heekin)	58	17	340	1
h. 167 Colong Road (Faul Rosenthal)	180	32	THIC	L30
5. 119 Colong Road (Frederick Scarborough)	30	<u>1.</u>	1300	72C
6. 239 Colong Road (Carmelite Fathers)	28	1	ć	2
7. 237 Oblong Road (Eric Thite)	11	3	30	11
8. 55 Colong Read (Arthur George)	29	2	1.2	0
9. 85 Tresdwell Hollow Road (Tim Stinson)	3£0	1	280	, 0
10. 191 Herlin Road (John Franz)	110	7	500	38
11. South Branch of Hemlock Prook	90	3	350	1
12. 270 Berlin Road (Roland Ethier)	500	10	190	<u>1.</u>
13. 331 Berlin Road (Karen Baker, Trustee)	210	<u>1.</u>	7 <u>1</u> 0	0
15. 395 Berlin Road (Floyd Rosenburg)	003	7	1200	5

* Shown on Map

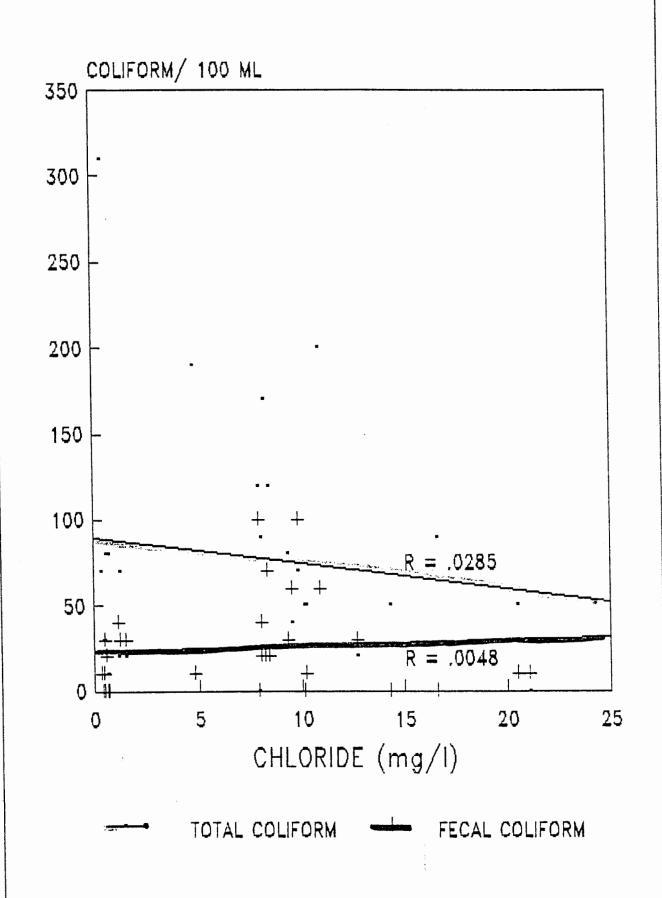
Prepared by Charles Kochenour Environmental Inspector Movember 17, 1988

ML6 TOTAL	LCOLIFORM	RESULTS
SAMPLE	COLIFORM	
A1	160	
A2 A3	60	
A3	50	
B1	40	
B2	40	
B3	60	
C1	80	
C2 C3	50	
СЗ	130	

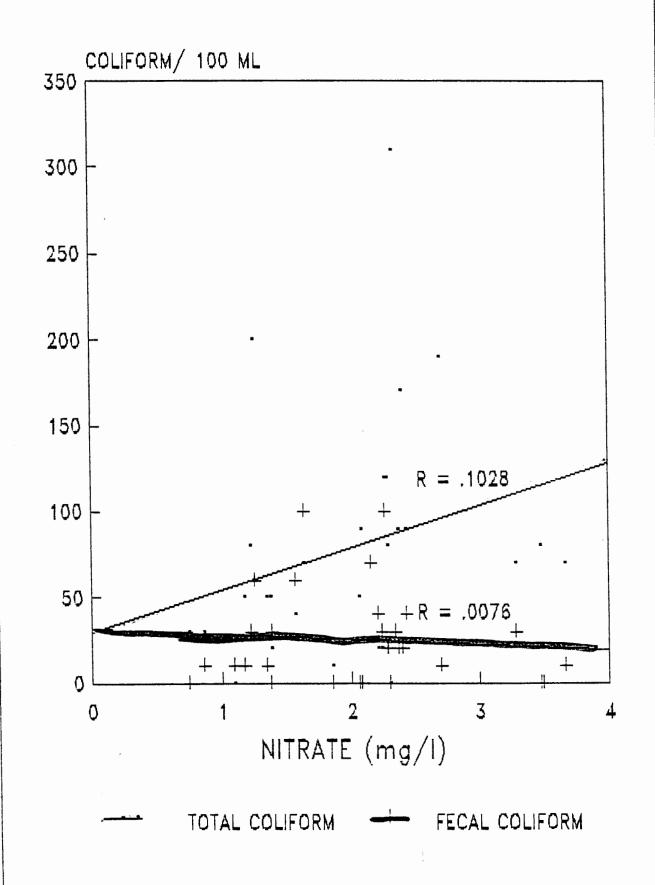
COLIFORM IN MARGARET LINDLEY PARK, AUGUST 1988										
	DATE		HEMLOCK		BATHING E		POND INL		POND OUT	
			TOTAL	FECAL	TOTAL	FECAL	TOTAL	FECAL	TOTAL	FECAL
В	2	88	1		1850	6	2100	34	3300	14
8	6	88			3600	0	4400	В	1800	0
8	8	88	1600	20	3200	0	2900	30	1400	0
В	10	88	2550	300			3060	18	1800	0
8	13	88	8500	20			13000	10	2800	0
8	15	88	50000	0	50000	0	50000	0	60000	0
В	17	88	24000	0	28000	42	44000	0	3000	1
8	18	88			1600	12				
8	20	88	18000	89	14000	10	12000	0	2000	0
8	24	88	12000	0	44000	0	19000	145	68000	278
8	27	. 88	3000	0	1	0	11000	3	5000	0
9	29	88	620	0						

TOTAL COLIFORM IN MARGARET LINDLEY PARK, 1990									
	DATE		HEMLOCK BROOK	BATHING AREA	POND INLET	POND OUTLET			
6	16	90	4500	150000	8	200000			
6	18	90	1 0000	29000	11000	20000			
6	23	90	7800	10000	7700	11000			
6	25	90	10000	5000	6000	3500			
6	27	90	5200	11000	6200	19000			
6	30	90	25000	18000	40000	14000			
7	2	90	4400	2500	3600	TNTC			
7	5	90	6000	1800	5600	200			
7	. 7	90	3800	8000	800	700			
7	' 9	90	4200	0	0	0			
7	21	90	2800	1	24	0			
8	15	90	760	20	700	0			

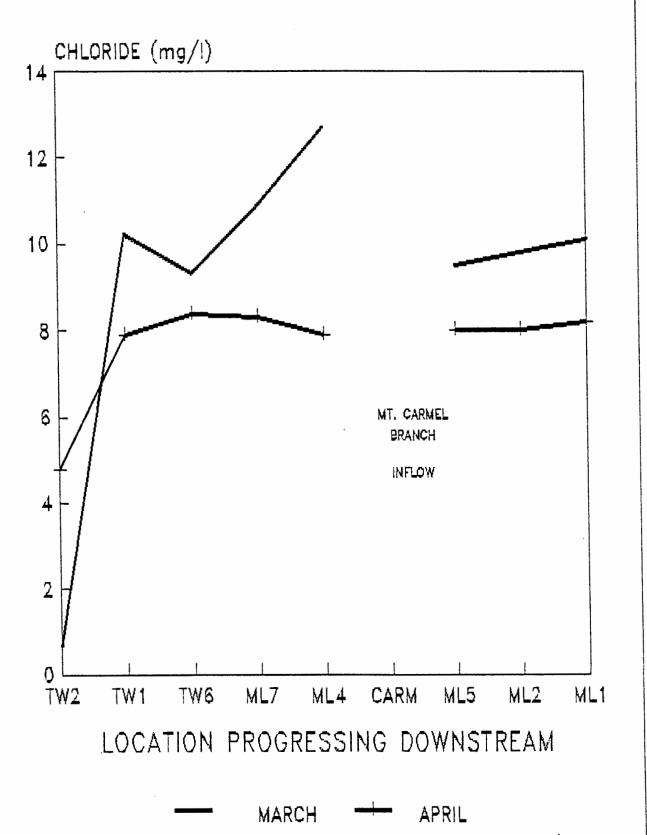
NON-CORRELATION: CHLORIDE AND COLIFORM



NON-CORRELATION OF NITRATE AND COLIFORM



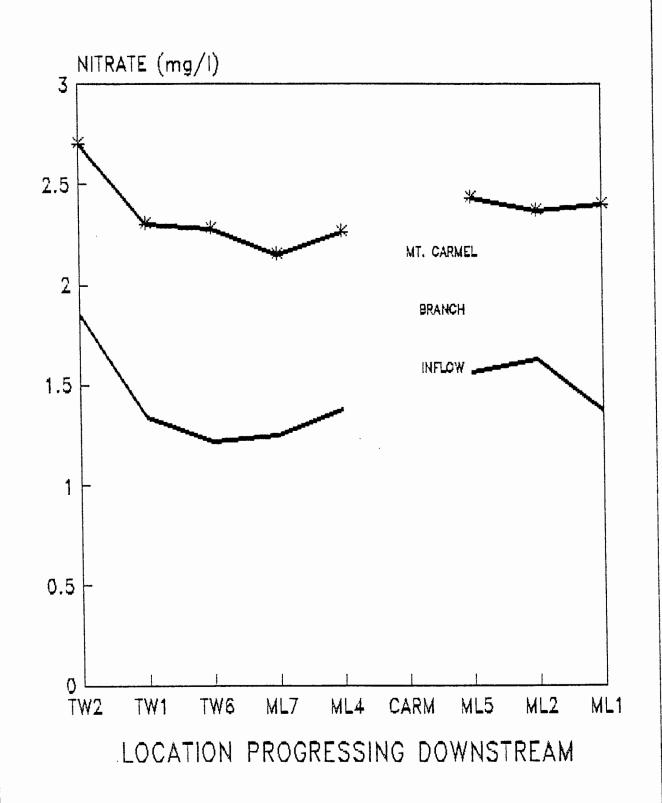
BERLIN BRANCH DOWNSTREAM PROGRESSION OF CHLORIDE



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Comprision to summer.

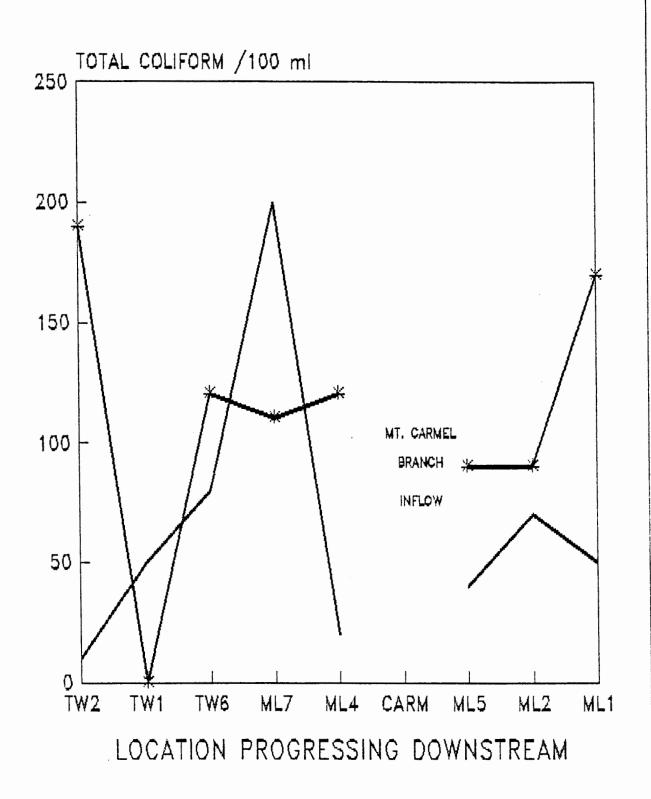
BERLIN BRANCH DOWNSTREAM PROGRESSION OF NITRATE



MARCH ** APRIL

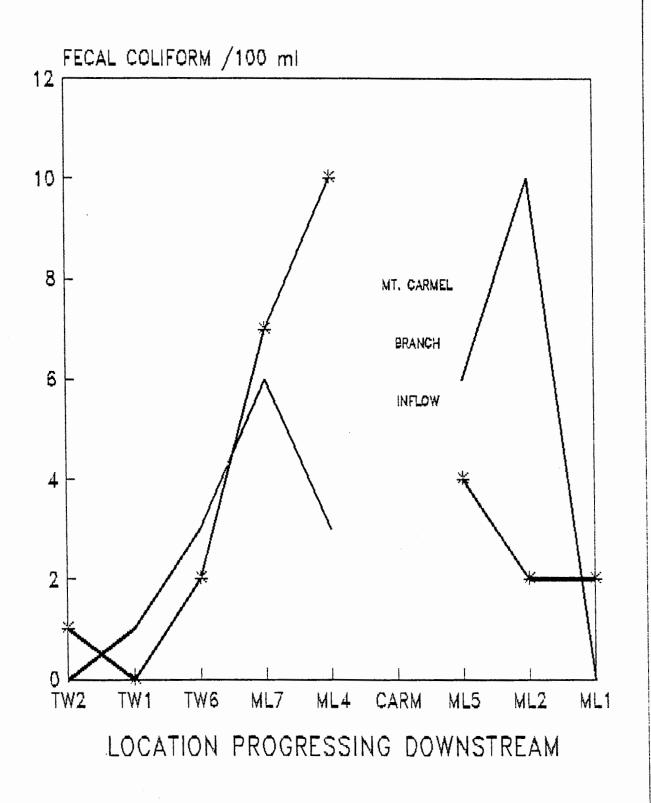
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BERLIN BRANCH DOWNSTREAM PROGRESSION OF TOTAL COLIFORM



── MARCH -** APRIL

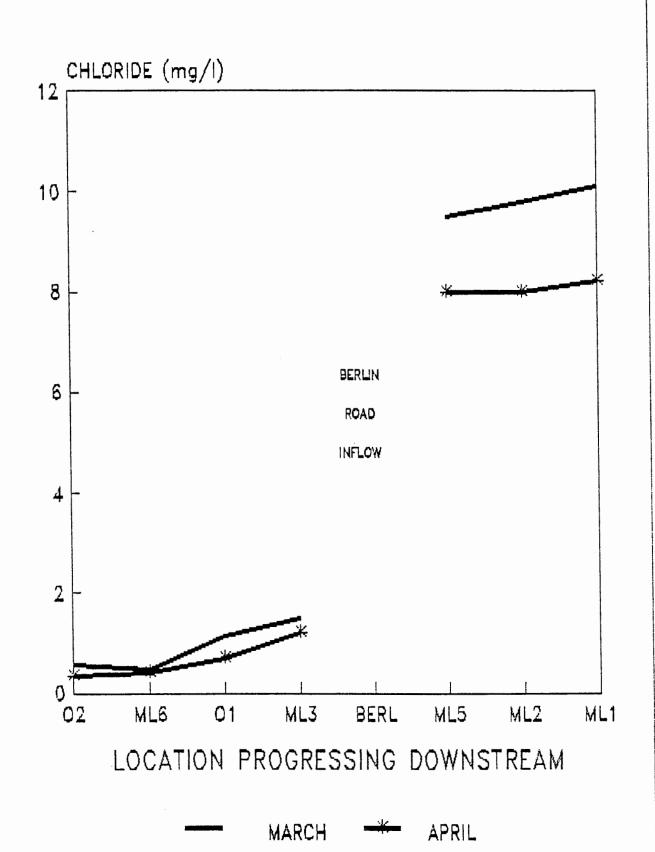
BERLIN BRANCH DOWNSTREAM PROGRESSION OF FECAL COLIFORM



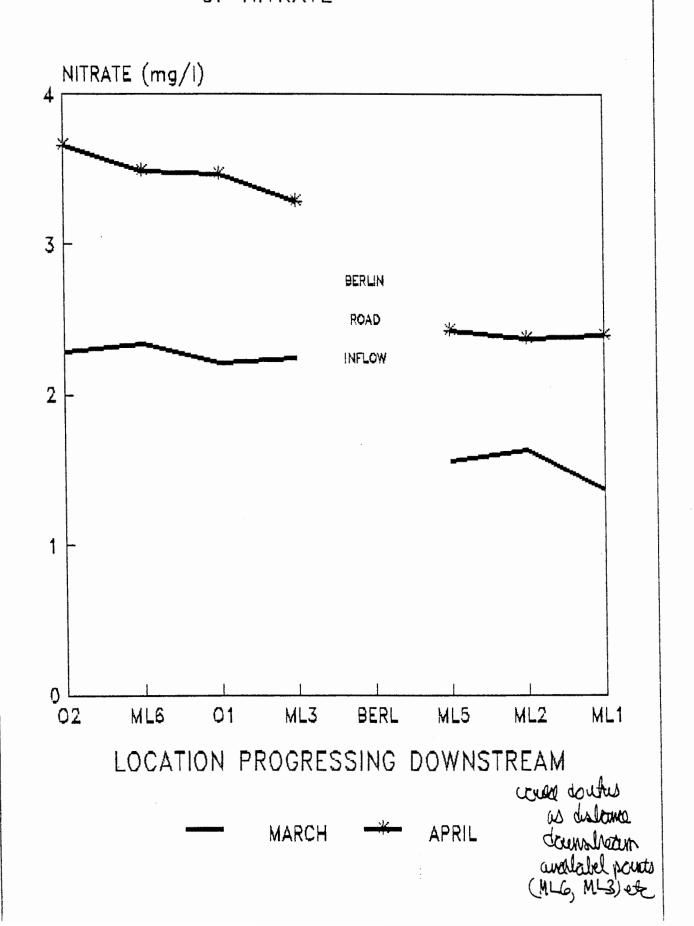
MARCH

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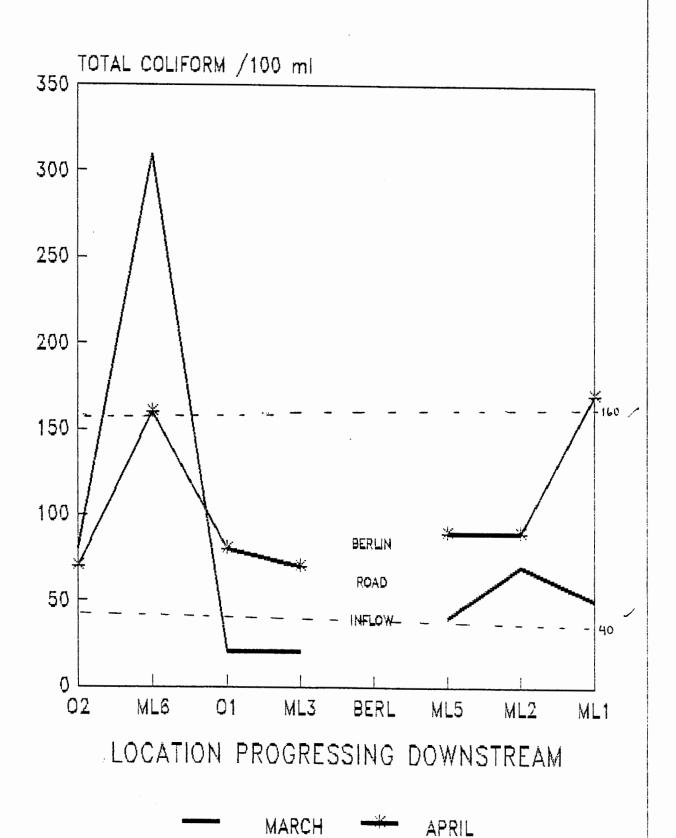
CARMEL BRANCH DOWNSTREAM PROGRESSION OF CHLORIDE



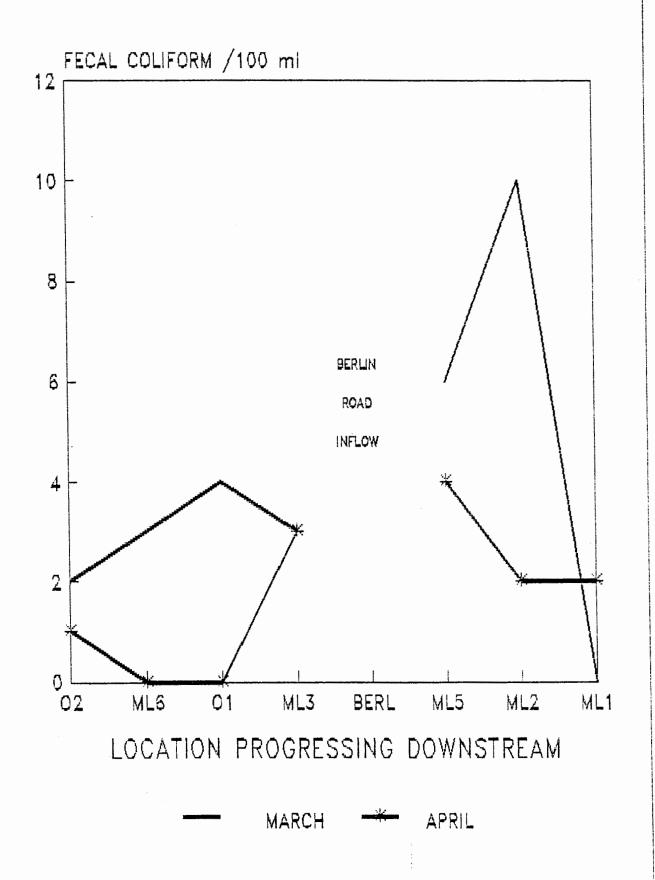
CARMEL BRANCH DOWNSTREAM PROGRESSION OF NITRATE

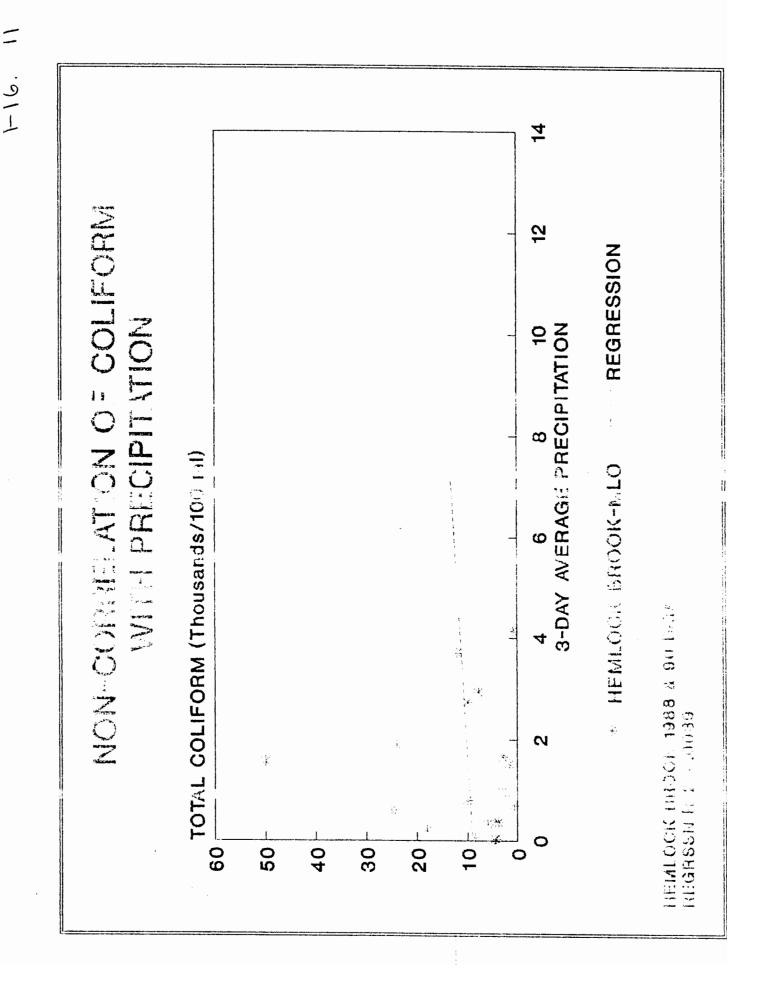


CARMEL BRANCH DOWNSTREAM PROGRESSION OF TOTAL COLIFORM

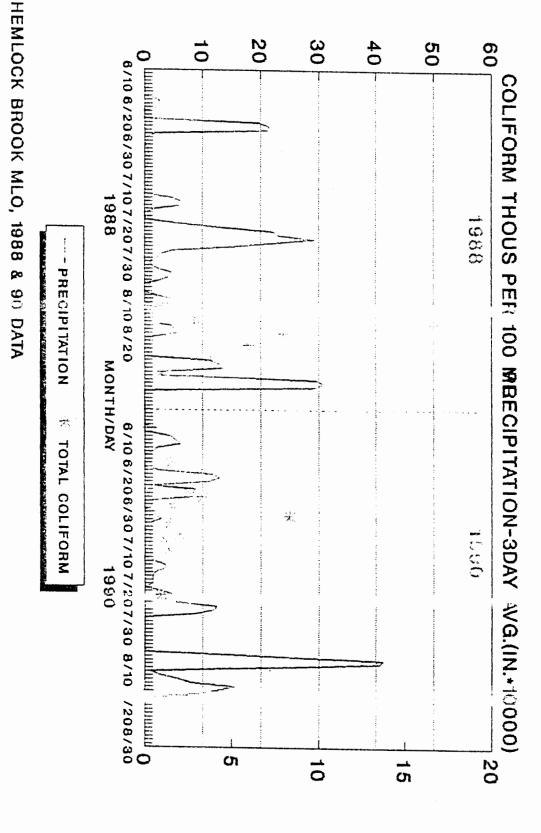


CARMEL BRANCH DOWNSTREAM PROGRESSION OF FECAL COLIFORM



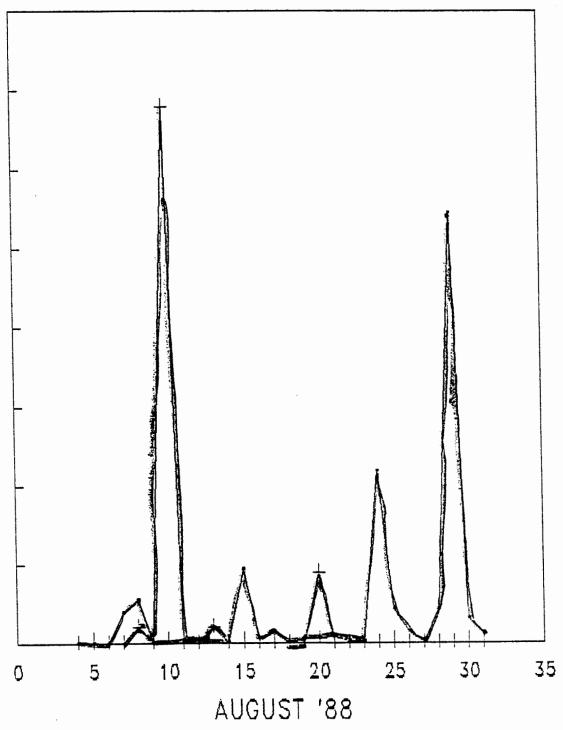






PRECIPITATION AND FECAL COLIFORM

FECAL COLIFORM AND PRECIP.



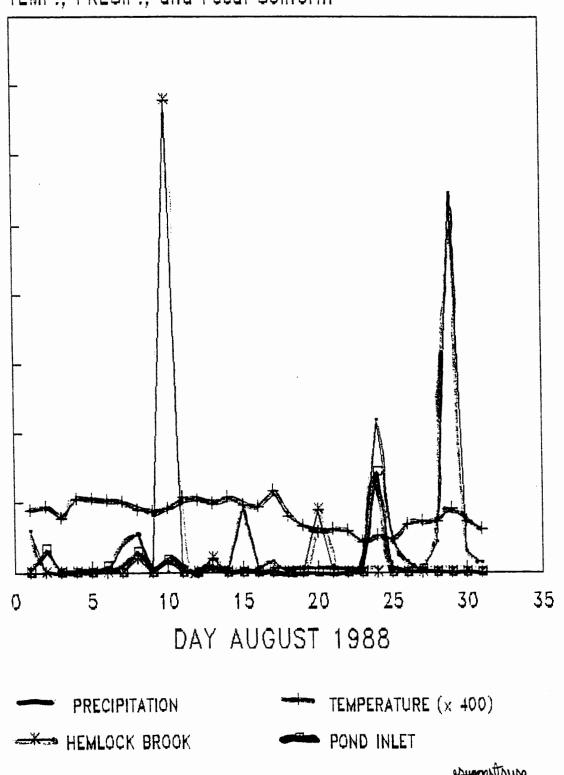
PRECIPITATION

FECAL COLIFORM
IN HEMLOCK BROOK V

Scales adjusted for display (wats?.)

Fecal Coliform in Brook and Inlet Compared to Precip. and Temp.

TEMP., PRECIP., and Fecal Coliform



Relative scales are altered

SOURCES OF COLIFORM BACTERIA IN HEMLOCK BROOK

Heather Stoll Williams College August, 1991 entering the stream on upper Berlin Road is the product of human and animal waste.

Because "natural" coliform input accounts for a significant percentage of coliform pollution in Hemlock Brook, and pollution from all sources is dramatically increased by precipitation events, this report recommends that the Recreation Committee continue chlorinating the pond in Margaret Lindley Park.

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INTRODUCTION

Counts of coliform bacteria in the pond at Williamstown's Margaret Lindley Park have exceeded the state bathing water standard (1000 colonies per 100ml) frequently over the course of the last three summers (Fig. 1). As a result, the pond, Williamstown's only public outdoor swimming area, was closed for extended periods during the summers of 1988 and '89, and was extensively chlorinated during the summers of 1990 and '91.

Levels of coliform in lower Hemlock Brook, which is used to feed the pond, have also been high in recent years. (M. Muzyka, Town Health Records, '88-'90). In this investigation, one hundred twenty nine samples were collected throughout the Hemlock Brook watershed between May and August to test for coliform contamination and ions associated with water contamination to investigate the source of these elevated coliform levels.

The effectiveness of the coliform test in tracking coliform entry lies in its ability to detect all living coliform organisms in the stream. However, recent research conducted by M.W. LeChevallier and G.A. McFetters (1985) demonstrated that coliform organisms stressed by chlorine used in municipal water treatment could go undetected by conventional analyses because they do not undergo cell division and cannot produce the colonies necessary for detection. A reduction of this stress permitted the organisms to resuscitate within a short time.

Since coliform bacteria derived from human and animal sources are acclimated to a warm body temperature of 37 degrees Celsius, stream temperatures of 16 degrees Celsius represent a significant cold stress. To assure that this cold stress was not interrupting coliform detection in the same manner as the chlorine stress, with injured organisms resuscitating in the warmer temperatures of the warming pool and pond in Margaret Lindley Park, a controlled test of coliform survival in stream and pond temperatures was also conducted.

Setting

The Hemlock Brook watershed above Margaret Lindley Park is located in the Berlin and Williamstown quadrangles in northwestern Massachusetts and adjacent New York. Three major branches - Treadwell Hollow Brook, Berlin Mountain Brook, and a southern branch flowing past the Mt. Carmel Novitiate (referred to here as the Mt. Carmel Branch) - drain the west side of the Taconic Crest between the Berlin Mountain Summit and Route 2, an area of 3.75 square miles (Fig. 1). The annual discharge of the brook has been approximated as 200,000,000 cubic feet (assuming that Hemlock Brook has the same unit area discharge as Birch Brook).

The Taconic Trail State Park occupies most of this land.

Only 264 acres in the drainage area is "developed" and actively used for residences and farmland. The remaining 2400 acres are

Table 1. Williamstown Precipitation and Temperature '88-'91

	June				July				
Year	88	89	90	91	88	89	90	91	
Daily avg. temp. (C)	15.6	19.8	17.8	18.6	21.0	21.4	20.4	20.1	
Precipitation (in.)	2.97	6.08	3.61	3.13	5.85	3.97	2.45	1.64	

fatalities had reduced the coliform levels in all cases by the next downstream residence. Since the high levels after these sources did not remain in the stream as far as the park, the high summer levels in the pond could not conclusively be attributed to these residences. In the spring of 1989, the town sampled several locations upstream, but found low levels at all sites which gave no indication of the source of the high summer levels.

On March 18 and April 11 of 1991, I sampled fifteen locations throughout the Hemlock Brook watershed above Margaret Lindley Park, and tested the water for total and fecal coliform and chloride and nitrate. Levels at all locations were low (less than 350 colonies/100 ml), making locational trends difficult to identify. Chloride and nitrate levels showed no correlation to either type of coliform.

In the spring of 1991, I also analyzed all of the 1988 1990 town test results for Margaret Lindley Park, and compared
them with temperature and precipitation data for those years. I
found that over this time period, levels of fecal coliform in
the brook rose following precipitation events. In addition, I
noted that levels of fecal coliform in the pond inlet rose only
during times of high precipitation, many times without preceding
highs in brook coliform levels. This suggested that a resident
coliform population may have existed in the calm warming pool
where the bacteria could settle, adhering to rocks and sediment
during drier weather. The bacteria would be flushed into the

significantly in the absence of a health hazard, 4) the indicator is more resistant to physiological stress within aquatic environments and is more resistant to disinfectants commonly used, and 5) the indicator reaction or test data is unique and characteristic of that microorganism, and the methodology is of minimal complexity and expense. (McFeters, Schillinger, and Stuart, 1978).

Aerobacter, Escherichia, Citrobacter, Klebsiella, Enterobacter and other species are included in the count for total coliform bacteria, while Escherichia is selectively cultured by temperature stress in the fecal coliform procedure, since it can survive a higher incubation temperature than the other strains. Consequently, the fecal coliform count is the most direct indicator of recent waste pollution.

The Enterobacter species and "Lancefield's Group D Streptococcus" comprise the fecal streptococcus group, which originates primarily from the intestines of warm-blooded animals. Hence, the fecal streptococcus test is a useful indicator of fecal pollution; however, fecal streptococci have a very limited survival time outside of the host animal, so counts are only useful in evaluating recent pollution.

Fecal coliform bacteria in human waste consist primarily of the Escherichia coli strain, while fecal coliform bacteria in animal waste is primarily comprised of streptococcus varieties. These differences can be used to determine whether bacterial pollution is derived from human or nonhuman sources, as long as recent pollution is sampled. Common ratios of fecal coliform to

Field

Twice in May, twice in June, and three times in July, I collected water samples successively at twelve to twenty locations along the Hemlock Brook system over a period of 3 hours. Sample localities are shown in Fig. 1. At each site, I collected 500 ml of water from the main current in a sterile, deionized plastic bottle to be tested for coliform and anions.

In late July, I collected 150 ml of water in deionized plastic bottles at ten locations progressing downstream in three areas of suspected contamination, to be tested for levels of anions. At these same target areas, I also collected 500 ml of water in sterile, deionized bottles at eight sites to be tested for fecal coliform and fecal streptococci bacteria.

Each month, I measured stream and pond temperatures.

Stream discharge was measured by instruments on Birch Brook as part of Williams College's Center for Environmental Studies' continuous monitoring in Hopkins Memorial Research Forest.

Weather data were also obtained from the Hopkins Forest weather station.

Laboratory

Within six hours, samples were cultured for total and fecal coliform according to standard E.P.A. approved methods (Federal Pollution Control Act, Section 304) Approximately 125 ml of

Table 2. COLIFORM SURVIVAL AND COLD SENSITIVITY

TOTAL COLIFORM

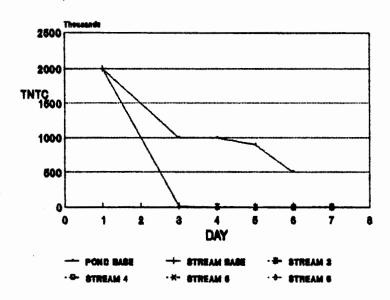
	DAY 1	DAY 3	DAY 4	DAY 5	DAY 6	DAY 7
POND BASE	TNTC*	TNTC*	TNTC*	900000	500000	********
STREAM BASE	TNTC*	11700	1400	••••••	0	0
STREAM 3**			4200	1100	2400	4300
STREAM 4					40	35
STREAM 5					10	10
STREAM 6						2

Note: On day 2, both the pond and stream bases were diluted 1:2 with sterile deionized distilled water.

A series of periods represents cultures which were not countable due to high background growth and other interferences.

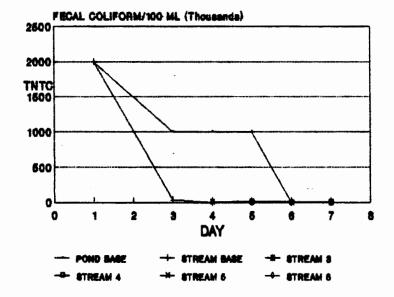
^{*} A 1/100 dilution was used.

^{**} STREAM 3 refers to the stream sample moved to pond temperature on the third day, STREAM 4 to the stream sample moved on day 4, etc.



STREAM 3 refere to the etrope complethal was seved to pend temperature on on the third day, etc.

Fig. 2. Total Coliform Survival



STREAM 3 refers to the atream sample moved to pond temperature on the third day, etc.

Fig. 3. Fecal Coliform Survival

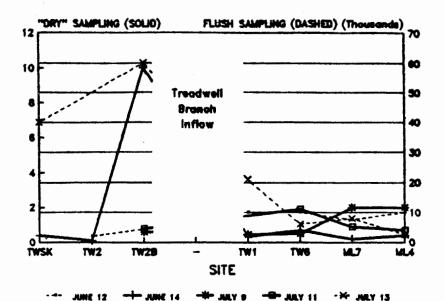


Fig. 5 — Total Coliform, June & July 1991 Berlin Branch

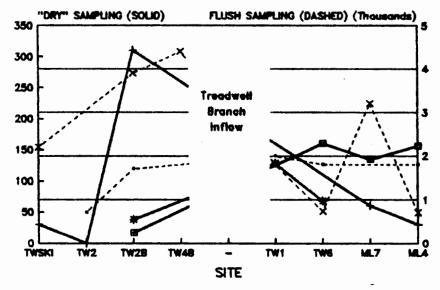


Fig 6 — Fecal Coliform, June & July 1991

Berlin Branch

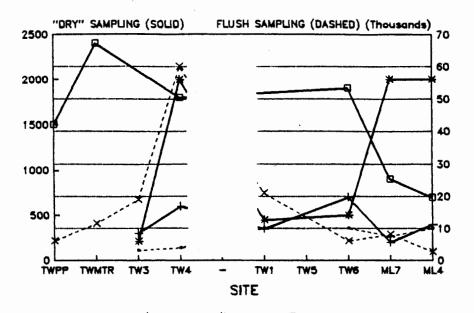


Fig. 9 — Total Coliform, June & July 1991
Treadwell Branch

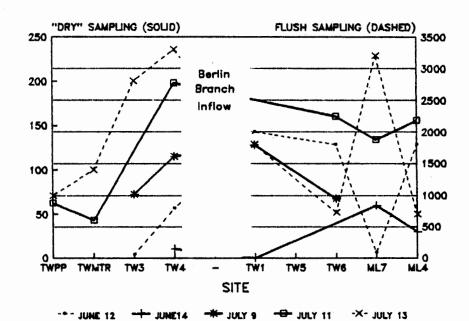


Fig. 10 —— Fecal Coliform, June & July 1991 Treadwell Branch

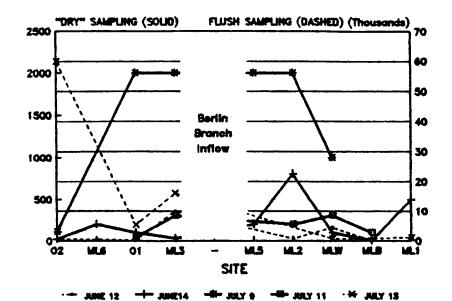


Fig. 13 -- Coliform, June & July 1991 Mt. Carmel Branch

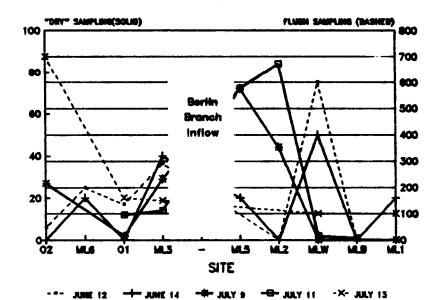


Fig. 14 -- Fecal Coliform, June & July 1991
Mt. Carmel Branch

DISCUSSION

The data suggest that in an experimental setup, the reduction of cold stress does not promote resuscitation of injured coliform organisms. Data from the stream sampling suggest that significant levels of coliform are entering the brook from "natural" sources above homes and pastures, and that coliform is entering regularly in three areas below homes and pastures. Weather appears to affect levels entering from both sources.

Coliform Survival and Cold Sensitivity

Numbers of both total and fecal coliform bacteria decreased over the course of the study, suggesting that coliform bacteria do not grow in early summer stream and pond conditions. (Fig.2 & 3) Consequently, after initial pollution, the coliform bacteria population in the stream is determined by conditions which inhibit or promote the survival of the organisms.

Water temperature appears to have a significant effect on survival rates. The 5 degree water temperature difference between the pond (21 C) and the stream (16 C) gave the pond samples a significant advantage. After two days, the total coliform bacteria population in the stream sample was only 1% of the pond sample population for that day. Concurrently, increasing the temperature of the stream samples increased

flow between the two sample sites. This permits the interpretation of recurring coliform increases over certain intervals, visible on Figs. 5 through 16, as areas where bacteria are entering the stream.

Influence of Weather

The average coliform levels increased significantly between the spring and summer of 1991, and notable increases occurred between the May and mid-summer samplings. As Table 4 shows, there was an appreciable increase in stream temperature between March and July. Figure 17 depicts the relationship between stream temperature and total and fecal coliform in non-precipitation flush conditions. As the cold sensitivity research predicted, a direct relationship exists between stream temperature and coliform survival. With the first increase in stream temperature, fecal coliform levels began to rise. Total coliform, on the other hand, appears to have a threshold temperature around 15 degrees: before the stream reached that temperature, levels remained relatively low; above 15 C, even small temperature increases enabled significant increases in survival.

Precipitation increased coliform levels in the stream significantly. Levels of coliform during the stream surges following the June 12 and July 13 rainstorms were, on average, 10 times higher than levels during dry periods. Because heavy rainfalls naturally increase the volume of water filling the stream channel, pollutants which were previously higher than the

streamflow are washed in. Heavy rainfalls which raise the water table could also be expected to contribute to overflow in faulty septic systems. In addition, higher stream discharges and higher stream velocities would reduce travel time from sources to test sites, possibly increasing coliform survival, would reduce the number of calm pools where bacteria could settle out of suspension, and would overcome adhesion of bacteria to banks, rocks, log dams, etc., leaving more in the current.

Natural Sources of Coliform

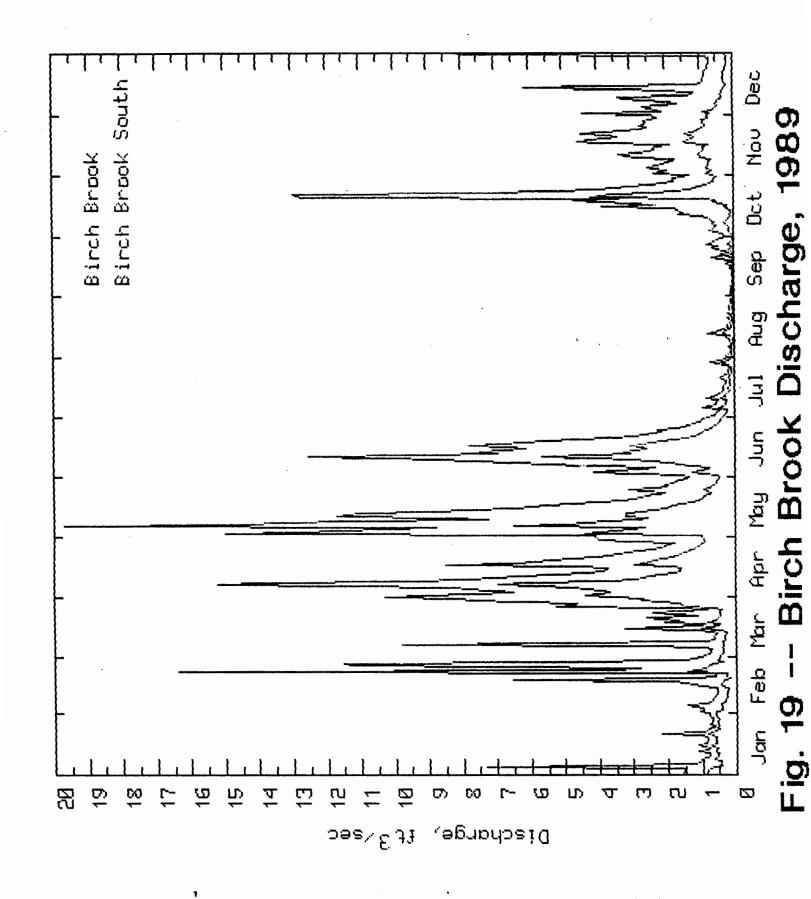
Estimates of the minimum levels of natural coliform entering the stream from non-domestic sources on the sample dates were obtained from the coliform levels at the sites which are above homes and pastures on each branch: TWSKI or TW2 on the Berlin Branch, the sum of TWPP and MTR, or TW3 on the Treadwell Branch, and O2 on the Mt. Carmel Branch. To determine the impact of natural pollution on the overall coliform pollution in the stream, these numbers were compared with the levels of coliform at the end of each branch. The percentage of the final, end-of-branch coliform count which the natural level comprised was calculated. The results appear in Table 5.

Significant percentages of both total and fecal coliform in the stream can thus be attributed to natural sources. Seventeen measurements of total coliform levels above human development and eleven measurements of fecal coliform levels above human development were compared with coliform levels at the end of the

respective branches. In 41% of the cases, levels of total coliform at the initial natural sample sites were as high as, or higher than the coliform levels at the end of the branch. In 27% of the cases, fecal coliform above homes and pastures was as high as or higher than the levels at the end of the branch. In 65% of the cases, total coliform levels above homes and pastures were more than half of total coliform levels at the end of the branch: this was true in 55% of the cases for fecal coliform.

The percent of pollution from initial natural sources may have varied because conditions which affect coliform survival or entry into the stream, like weather, may affect natural and human/domestic sources differently. In the May samplings, for example, natural total coliform pollution accounted for more than half of the final branch pollution in all cases, while in June and July only 45 percent. The discharge of the brook in May is generally higher than in June or July (Figs. 7 and 8 - No '91 data available), which may have washed coliform more continuously from natural sources and diluted human/domestic sources.

These percent figures of the role of natural sources provide only minimum estimates of how much of the stream coliform is from natural sources, because the final branch figures do not take into account coliform which may have been added from natural sources between the highest sample site (above human developments) and the last branch site. However, no data are available to demonstrate that the entire number of bacteria at the top of the branch traveled all the way to the



chloride and/or nitrate levels; these two anions are often indicators of human or animal waste pollution.

Significant recurring coliform trends occurred between sites in three areas in the Hemlock Brook Watershed surveys.

Levels of coliform consistently increased between TW2 (or TWSKI) and TW2B, between TW3 and TW4, and between O1 and ML3.

On every sampling, May through July, fecal coliform increased between TW2 (or TWSKI) and TW2B (See Table 6). Total coliform also increased between these sites in June and July. Nitrate levels also rose slightly between these sites in August and July, as did chloride in all of the targeted progression sampling in July, suggesting that the coliform increases were probably associated with pollution from human or animal waste (Figs. 7 & 8)

Levels of total and fecal coliform increased between TW3 and TW4 at every sampling in June and July. (Table 7) The frequent decrease in chloride and nitrate levels over this interval may be a consequence of uptake by plants and algae in Stinson's pond which the stream flows through between these sites, or it may reflect dilution from increased discharge. (Figs. 11 & 12)

Although the increases in total and fecal coliform between O1 and ML3 were not as consistent as those on the Berlin and Treadwell Hollow Branches, there is sufficient recurrence to warrant closer examination. (Table 8) Nitrate levels showed no repeating pattern, whereas chloride levels in June and July

Table 7. COLIFORM INCREASES: TREADWELL BRANCH TW3 -- TW4
TOTAL COLIFORM

DATE	<u>EWT</u>	<u>TW4</u>	% INCR.
8/12	3000	3800	26.87%
6/14	300	600	100.00%
7/9	210	2001	852.86%
7/13	19000	60001	215.79%

FECAL COLIFORM

DATE	TW3	<u>TW4</u>	% INCR.		
8/12	;	54	800	1381.48%	
8/14	NDA		10		
7/9	;	73	115	57.53%	
7/13	280	00	3300	17.88%	

always increased between these sites, indicating a possibility of pollution from human or animal waste. (Figs. 15 & 16)

Fecal coliform/fecal streptococcus ratios for these intervals show that human pollution is probably a factor only in the Berlin interval. Animal pollution is responsible for the fecal coliform along Treadwell Hollow and below Oblong Road, although no distinction can be made as to whether the animals are domestic horses, cows, sheep, etc., or wild animals like deer, raccoon, and coyote.

A survey of the stream beginning above the last house on Berlin Road and continuing to the junction of the Berlin and Treadwell Branches found no evidence of direct discharge into the stream from pipes or trenches.

CONCLUSIONS

"Natural" coliform bacteria originating from sources upstream from human development may be responsible for a significant percentage of the coliform pollution in Hemlock Brook. During precipitation flushes which occurred on June 12 and July 13, levels of coliform bacteria in the stream exceeded the bathing water standard at two of the three sites above human development.

Three areas of significant recurring increases in both total and fecal coliform have been identified; on upper Berlin Road, on Treadwell Hollow Road, and on the Mt. Carmel Branch below Oblong Road. The coliform on Treadwell Hollow Road and

I would recommend that the Recreation Committee look into other means of chlorine addition which might yield the same results at less expense, although continued daily "drag" chlorination of the beach area should continue regardless.

I would also recommend that the Town Health Department further investigate the possibility of human waste pollution entering along upper Berlin Road with more site-specific testing during the summer months when increased coliform survival maximizes pollution detection.

To test the accuracy of the coliform test method, ten samples were collected successively from location TW2B. Each sample was split and tested for both total and fecal coliform. The results appear on Table and are graphed in Fig. .

In a totally precise procedure, the points would be expected to lie along the x = y line since the bacteria population count for split 2 would be the same as the count for split 1. This is not the case for either graph.

The statistical accuracy of the method was analyzed by a hypothesis test (paired difference experiment). Since the splits of each bottle were initially the same sample, the difference between the counts for each pair of bottles should be zero. To determine whether the true mean of the population of differences (Mp) could equal zero, we formulate two hypotheses. The hypotheses are:

Ho:
$$M_D = 0$$
 ($M_1 - M_2 = 0$) (Method is accurate)

$$H_A: M_D /= 0 (M_1 - M_2 /= 0) (Method is not accurate)$$

Either the true mean of the population of differences is equal to zero, or it is not. To prove one of the hypotheses false, we determine whether the true mean of differences could be zero by seeing if zero falls within the 95 % confidence limits for the true mean. The formula for finding these differences using the mean difference xp, the critical value of t for the appropriate

METHOD CHECK RESULTS: TOTAL AND FECAL COLIFORM

TOTAL COLIFORM

BOTTLE	SPLIT 1	SPLIT 2	DIFFERENCE			
A	150	80	70			
В	20	360	-340			
C	50	50	0			
D	70	50	20			
E	110	90	20			
F	80	100	-20			
G	90	100	-10			
Н	90	110	-20			
1	60	110	-50			
J	90	110	-20			
		AVG. D	-35			
		118				
		UP. LIM.				
•		LOW. LIM.				

FECAL COLIFORM

BOTTLE	SPLIT 1	SPLIT 2	DIFFERENCE		
Α	1	5 1	14 1		
В	2	2 3	35 -13	1	
C	3	6 3	32 4		
D	3	8 3	32 6		
E	. 3	7	45 -8	•	
F	1	6 3	31 -15		
G		3 2	≥1 -18	,	
		AVG. D	е	;	
		11			
		UP. LIM.			
		LOW. LIN	11 6	į	

APPENDIX B

Coliform and Ion Analysis Results

Explanation:

These sheets list the coliform and ion information obtained for each Hemlock Brook sample between May and July 1991. The time of collection, precipitation in previous 24 hours, and daily average temperature is also provided. In cases of significant precipitation, the conditions of increased stream discharge on sampling are approximated with two percentages: 1) how high the stream was at the sampling time compared to the highest discharge level of the precipitation event, listed as "Percent of discharge peak," and 2) how much the stream discharge had increased at that time compared to the average discharge for the 12 hours preceding the precipitation event, listed as "Discharge percent increase." Blank spaces were left where data was not obtained. For the fecal coliform/fecal streptococcus ratios, interpretation on the pollution source based on standard E.P.A. values is given.

COLIFORM AND ION ANALYSIS

5/31/91

Time of Collection:

9:30 - 11:30 AM

Precipitation in previous 24 hours:

0

Temperature (Daily Average):

23.3 C

	COLIFORM				
	colonies/10		mg/l		
SITE	TOTAL	<u>FECAL</u>	NITRATE	CHLORIDE	TEMP. C
T W 1	140	4	1.15	8.1	14
TW2	180	**	1.27	0.4	14
TW2B	40	57	1.22	1.63	14
EWT	190	**	1.04	20.06	14
TW4	70	52	1.01	17.5	14
TWBEN	150	52	1.76		14
TW5	50	26	0.38	1.45	14
TWB	70	30	1,07	10	16
01	40	0	1.62		17
05	230	116	1.76	0.48	16
ML1	50	1	1.09		18
ML2	110	24	1.04		16
ML3	70	0	1.47		16
ML4	30	45	1.07	12.9	16
ML5	40	58	1.07		17
ML6	30	0	1.67		16
ML7	110	55	1	9.8	16
MLB	0	2	0.56		26
MLWAN	30	0	1.01		17
ML5B P	0	80			
AVG.	82	28	1.17	7.56	

^{**} Suspicious results: Recent examination suggests that the bottom layer in the fecal colliform incubator bath may be hotter than the prescribed temperature, adversely affecting (burning out) colliform growth. The zero counts originally obtained for these cultures do not appear to be consistent with counts upstream from that particular site, or with counts for that site on other days.

COLIFORM AND ION ANALYSIS

6/14/91

Time of Collection:

9:30 - 11:30 AM

Precipitation in previous 24 hours:

0

Temperature (Dally Average):

15.28 C

	COLIF			NIONS
SITE	COlonies/10	FECAL	mg/l NITRATE	CHLORIDE
TW1	350	**	1.21	18.1
TW2	100	0	1.21	0.45
TW2B	10001*	310	1.37	0.6
EWT	300	**	1.31	35.2
TW4	600	10	1.28	22.4
TWB	700	**	1.16	10.4
O 1	100	0	1.47	0.62
02	20	0	1.83	0.47
ML1	500	20		17.4
ML2	800	**	1,14	29.1
ML3	30	40	1.38	1.96
ML4	400	30	1.18	26.2
ML5	200	20	1.2	14.7
ML6	200	20	1.61	0.43
ML7	200	60	1.05	8.8
MLW	100	50	1.11	11.3
MLB	0	0	0.93	
TWSKI	400	30	1.06	0.49
AVG.	833	33	1.19	11.68

^{*} Large values with several zero digits ending in a 1 represent a best approximation of cultures which were too numerous to count. This approximation is based on the dilution and the highest countable number in that day's series. This approximation was used to compute the average.

^{**} Suspicious results: Recent examination suggests that the bottom layer in the fecal collform incubator bath may be hotter than the prescribed temperature, adversely affecting (burning out) colliform growth. The zero counts originally obtained for these cultures do not appear to be consistent with counts upstream from that particular site, or with counts for that site on other days.

COLIFORM AND ION ANALYSIS

7/11/91

Time of Collection:

10:00 - 12:00 AM

Precipitation in previous 24 hours:

0

Temperature (Daily Average):

15.87 C

COLIFORM colonies/100mi

	CO1011185/100m1				
SITE	TOTAL	FECAL			
TW2B	800	17			
TW4	1800	198			
TW6	1900	160			
O 1	50	12			
ML2	220	84			
ML3	300	14			
ML4	700	156			
ML5	2001	72			
ML7	900	134			
MLW	300	2			
MLB	100	1			
AVG.	825	77			

CHLORIDE AND NITRATE: TARGETED PROGRESSIONS

	7/18/91	•	7/21/91		7/23/91		7/13/91	
SITE	CHLORIDE NITE	ATE (CHLORIDE N	ITRATE	CHLORIDE N	ITRATE	CHLORIDE	NITRATE
TWSKI			0.57	1.69	0.56	2.11	0.61	1.54
A-395	0.41	2.4	0.07	1.05	0.56	2.11	0.61	1.54
A-331	0.35	2.58						I
TW2B	0.45	2.61	0.67	2.32	0.49	2.66	0.51	2.45
A-191	0.8	2.61			_			
HSKM	0. 98	2.66	1.29	2.84	0.81	2.68	2.1	2.6
		1						
TW3	19.6	2.6	23.8	3.45	22.2	2.91		
TW3B	21.6	2.66	30.8	2.94	17	2.95		İ
TW4	20.6	2.05	22.8	2.49	20.2	2.39		
01		1			0.57	0.24	0.70	1.05
O1 ML3					0.57 1. 38	2.31 2.35	0.79	1.85 1.27
MILO		j			1,38	2.30	1.41	1.21
24 hr precipi	tation:	o			o	45 ln		0.65 ln
Time of coile	ction: 4:0	D PM		4:00 PM	9:	MA OE		5:00 PM
% discharge	peak: -		1	00%	5.	7%	·	100%
			•		_	•	1	
% discharge	base: -		. 5	0%	91	B 9%		2379%
Temperature	: 24.7	С	2	3.9 C	2	4,4 C		18.3 C

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