# TRANSPORT AND VARIABILITY OF INDICATOR BACTERIA IN THE APALACHICOLA RIVER AND ESTUARY, FLORIDA, 1983-84 By John F. Elder

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### TRANSPORT AND VARIABILITY OF INDICATOR

#### **BACTERIA IN THE APALACHICOLA RIVER**

## AND ESTUARY, FLORIDA, 1983-84

### By John F. Elder

#### ABSTRACT

Fecal coliform bacteria in the waters of Apalachicola Bay, Florida, are monitored regularly by the State to detect conditions where pathogens may contaminate oysters harvested for human consumption. Because coliform numbers tend to increase during periods when the Apalachicola River rises above flood stage, the general practice has been to close the bay to harvesting at high river ovster Closure continues until monstages. itoring shows that the coliform numbers do not exceed the maximum allowable limit of 14 colonies per 100 milliliters. However, no direct evidence is available to show a simple relation between river stage and coliform numbers.

To assess riverine bacterial transport, numbers of fecal coliform and fecal streptococci bacteria were monitored at 12 sites on the Apalachicola River during the spring of 1983 and winter of 1983-84. The data did not show evidence of cross-sectional patterns of variation or discrepancies between the two most common methods of analysis (membrane filtration and most probable number). Coliform numbers were generally lower near the river mouth than at sites farther upstream. At most sites, fecal coliform:fecal streptococcus ratios were less than 2.0, suggesting nonhuman origin for much of the fecal bacteria. Higher ratios (up to 23) at some sites, in particular a site below a wastewater treatment facility, suggest human origins. With the exception of an outlet from a wastewater treatment facility near the river mouth, tributaries generally carried lower coliform or streptococcus concentrations than the main river channel.

The data from this study suggested that the coliform:discharge relation was modified by other hydrologic factors, particularly the flood duration, the magnitude of the current flood peak relative to previous peaks, and whether the river was rising or falling. A multiple-regression model indicated that bacteria concentrations during the rising limb of the season's initial flood peak are likely to be up to 50 percent greater than concentrations during the falling limb of a later peak, given approximately equal discharge levels of the two peaks.

In the estuary, coliform data collected at 28 sites during 1983-84 were analyzed for regional differences. Median counts from sites near the river mouth were four to five times higher than those from offshore areas in the estuary. Although there are some weaknesses in the use of coliform data to assess potential pathogen contamination of shellfish, the data were consistent with the general practice of limiting shellfish harvesting from the area near the river mouth.

#### INTRODUCTION

Water quality of large rivers has been under ever-increasing study in recent years (Daniel and others, 1979; Harned, 1980; Mattraw and Elder, 1984). Much of the need for maintaining high water quality in large rivers is derived from their impact on the estuaries into which they discharge continually. The character of the estuary is largely controlled by the river which feeds it (Naiman and Sibert. 1978; Livingston, 1983). Under normal circumstances, the estuary is highly productive and provides economically important resources for Alterations to the riverine man. water quality by disturbances in the drainage area can lower the estuarine productivity or otherwise impact the viability of the resources.

Transport of fecal bacteria into the estuary by riverine inflow is not likely to diminish estuarine productivity, but it can threaten the usefulness of any commercial seafood harvested from the estuary. Bacterial occurrence is extremely variable, but it is likely to be affected considerably by human and animal populations inhabiting the drainage basin (Geldreich and Kenner, 1969; Kress and Gifford, 1984). This study was undertaken to describe some aspects of fecal bacterial transport in a large southeastern river and to evaluate possible impacts of such transport on the distribution of fecal bacteria in the receiving estuary.

#### Description of Study Area

The Apalachicola River in northwest Florida is formed by the confluence of the Chattahoochee River and Flint River and has a 50,800-km<sup>2</sup> drainage system encompassing parts of three states (fig. 1). Approximately 12 percent of this area  $(6,200 \text{ km}^2)$ is the watershed of the Apalachicola and Chipola Rivers. With an average discharge of 870 m<sup>3</sup>/s at Chattahoochee, the Apalachicola is the largest river in Florida. The river falls 12 m in its 171-km course from Lake Seminole, Florida-Georgia at the State line, to Apalachicola Bay, in the Gulf of Mexico. Each winter and spring, rainstorms in Georgia produce increases in discharge from low-flow levels near  $300 \text{ m}^3/\text{s}$  to spring flood levels as high as  $4,000 \text{ m}^3/\text{s}$ . The flood plain, which is generally inundated for 3 to 5 months each year, occupies 450 km<sup>2</sup> and broadens downstream from 2 km wide just below Lake Seminole to more than 10 km wide near the mouth. At its mouth, the river empties into Apalachicola Bay, one of the most productive shellfish areas in the United States.

Upstream from Lake Seminole, the Chattahoochee River receives runoff and discharge from urban, industrial, and agricultural development. The city of Atlanta and several smaller cities are situated along the river. Apalachicola-Chipola However, the basin in Florida is relatively undeveloped. The major population centers within the basin--Chattahoochee, Bristol, Wewahitchka, Blountstown, Sumatra, and Apalachi-Marianna, cola--all have fewer than 8,000 in-

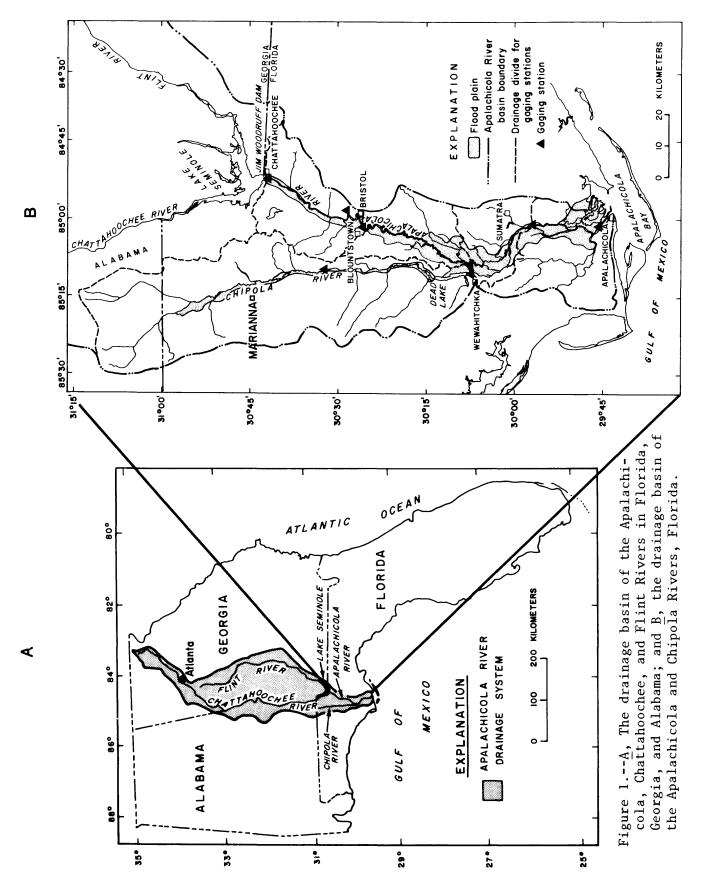


Table 1Principal	wastewater treatmer	t plants	and their daily
effluent volumes	in the Apalachicola	-Chipola	River basin

[From Gerald	Herting, Flor:	da Department of Environmental	
	Regulation, or	al commun., 1982]	

Site	Treated effluent volume (in cubic meters per day)	Site	Treated effluent volume (in cubic meters per day)
Marianna	5,500	Blountstown	1,600
(Chipola River)		Wewahitchka	270
Chattahoochee	2,400	Apalachicola	1,700
Florida State Hosp (Chattahoochee)	pital 2,300	-	

habitants. Municipal wastewater treatment plants at some of these communities are listed in table 1. Principal industries and agriculture are listed in table 2.

#### **Background Information**

Earlier studies conducted by scientists at Florida State University (Livingston and others, 1974) and the U.S. Geological Survey (Mattraw and Elder, 1984) have documented the close dependence of the oyster and other shellfish populations on flow from the river. The hydrologic behavior of the river, in particular the annual flooding, is critical to support the dense bottom land-hardwood forest which occupies the flood plain (Leitman and others, 1983). Leaf and woody litter from the forest partially decomposes on the forest floor and provides a vast source of detritus which can be transported by the floodwaters to the bay (Elder and Cairns, 1982). This material supports the highly productive detritivore community in the bay (Livingston, 1983). In this and other ways, the relation between the river system and the bay contributes directly to the economic welfare of the city of Apalachicola and vicinity. **Oysters** (Crassostrea virginica Gmelin) depend on an adequate nutrient supply to sustain their planktonic food source. Periodic pulses of freshwater provide these nutrients and also discourage predation by oyster drills (Menzel and others, 1966). Oyster harvesting is the major industry of the county. Shrimp (Penaeus sp.) and blue crab (Callinectes sapidus), both detritivores, are the second and third most important seafood species.

Although the river flooding is beneficial to the Apalachicola estuarine community, it also may create certain problems by transporting increased loads of pathogenic organisms (Thompson and others, 1984) and toxic substances (Elder and Mattraw, 1984). High concentrations of coliform bacteria in the water indicate the possibility of pathogen ingestion by shellfish species, thereby creating a potential health hazard when those species are harvested. Such bacterial increases occur guite commonly and make it necessary to continually monitor the bay waters for presence of coliform. The Florida Department of Natural Resources (FDNR), Division Marine Resources, maintains a of weekly monitoring program covering 28 widely distributed sampling sites in the bay (fig. 2). Sampling is more frequent during flood stage.

Standards of maximum coliform bacteria contamination have been set by the Interstate Shellfish Sanitation Program (ISSP), an association of the

4

### Table 2.--Principal industries and agriculture in the Apalachicola-Chipola River basin

			Cou	nty		
Jac	kson	Gadsden	Calhoun	Liberty	Gulf	Franklin

# A.--Industries in cities of the Apalachicola-Chipola River basin, by category (from Industries Guide, Inc., 1982).

Seafood						16
Other food and dairy	7					
Beverages	2					1
Animal feed, grains	3		1			
Lumber and building	5	1	5			2
supplies, wood products						
Rock, concrete, concrete	3		1			
products, asphalt						
Machinery, metal products	3		2			
Chemicals	5			1		
Paper, pulp, chips			4			
Clothing, cloth products	2		1			1
Publishing, printing	1	1			1	2

# B.--Farmland in the six counties of the Apalachicola-Chipola River basin, 1982 census (from U.S. Department of Commerce, 1984).

Number of farms Areas: <sup>1</sup>	960	404	169	81	39	5
Cropland and pasture	70.3	15.6	15.6	2.1	6.8	N <sup>2</sup>
Woodland	22.5	12.5	4.0	4.1	9.0	N
Other	16.2	3.4	3.0	.5	4.4	.1
Total farmland	109.0	31.5	22.6	6.7	20.2	N
Percent of land area	44.7	23.5	15.4	3.1	14.0	N
occupied by farmland						

<sup>1</sup> Areas in thousands of hectares.

<sup>2</sup> N = data not available.

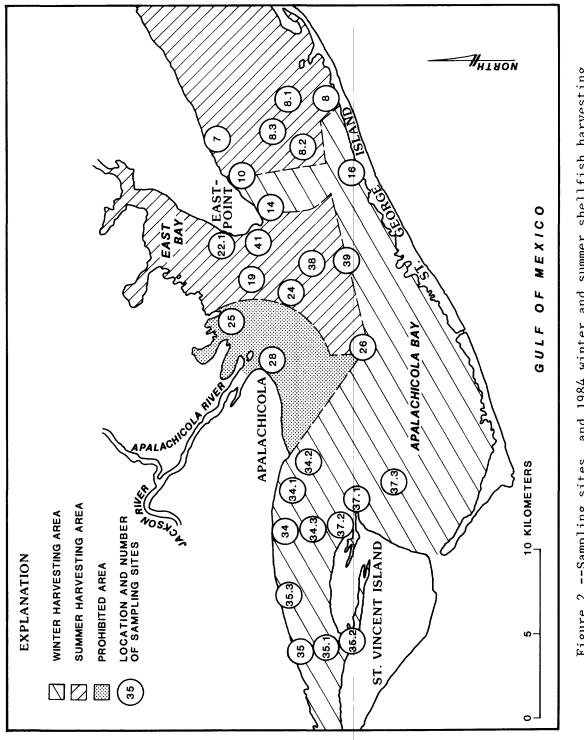


Figure 2.--Sampling sites, and 1984 winter and summer shellfish harvesting areas and prohibited areas, Apalachicola Bay. State of Florida, the U.S. Food and Drug Administration, and the shellfish industry. Areas of the estuary which show median fecal coliform MPN (most probable number) values greater than 14 MPN per 100 mL (milliliter) and which exceed 43 MPN per 100 mL more than 10 percent of the time exceed the standards and may be declared prohibited for shellfish harvesting (Thompson and others, 1984). Although coliform abundance is not a direct measure of pathogenic organisms in the water or oyster tissue, the setting of standards is important to protect both public health and the seafood industr**v**. Further understanding of occurrence, distribution, mortality rates of bacteria, and correlations between bacterial occurrence in the water and pathogenic content in tissues of shellfish organisms may assist in refining management strategies for the estuary.

Data from the 28 sites in Apalachicola Bay show that during the 5year period 1979-84 only 2 sites, both near the mouth of the river, exceeded both of the ISSP standards (Thompson and others, 1984). However, at 4 other sites the winter medians exceeded 14 MPN per 100 mL, and at 15 other sites the winter coliform counts exceeded 43 MPN per 100 mL more than 10 percent of the time. Summer counts were lower, and except for the two river-mouth sites, rarely exceeded either standard.

Numerous uncertainties exist with respect to sources of coliform bacteria found in the bay. Historical data from estuarine bacterial counts seem to show a correlation between high bacterial counts and river flooding, suggesting that the bacteria are transported from sources in the flood plain. However, there have been no parallel data from points along the river to show specific areas of bacterial input. Lacking this information, but given the apparent relation to flooding, the continually monitored river stage at Blountstown is used as an index for estimating the danger of bacterial contamination of the bay. A stage of 4.9 m, corresponding to a flow of  $1,200 \text{ m}^3/\text{s}$ , calls for closure of commercial oystering until actual measurements in the bay show that coliform abundance does not exceed ISSP standards.

Prior to this study, there have been no data to identify point sources for coliforms along the Apalachicola River. Septic tanks buried below flood stage are quite common in the Wewahitchka and Apalachicola vicinities, and are commonly suspected of accounting for flood surges of bacteria. Fecal material from animals in the flood plain may also be an important source, in which case the fecal streptococcus level should be relatively high. Sewage treatment plants, which discharge effluent aftreatment, ter secondary can be another source.

#### Purpose and Scope

The purpose of this investigation and report is to provide information about transport of indicator bacteria and variability of bacterial concentrations in the Apalachicola River. Such information is valuable for further understanding of the impacts of river hydrology on the shellfish populations in the estuary. Specific objectives of the study are:

- To determine the relations, if any, among river discharge and rainfall in the basin and bacterial densities in the estuary;
- 2. To determine sources, numbers, and types of fecal indicator bacteria transported by the river; and
- To describe implications of the study results with respect to estuarine bacterial abundance and distribution and the usefulness of the fecal bacteria as contamination indicators.

#### Units of Measure

The units of measure used in this report are metric in order to facilitate comparability with most other literature dealing with similar research. However, river sampling sites were numbered according to their locations with respect to river navigation miles. As standard reference points, which are familiar to local users and managers of the system, the river-mile designations will continue to be used throughout the report.

#### Acknowledgments

The author expresses appreciation to the Florida Department of Natural Resources, Division of Marine Resources, for cooperation on this study. Division personnel who made substantial contributions included Edwin A. Joyce, John Schneider, Charles Futch, John Taylor, Jr., and Woodard W. Miley, II.

The study was conducted over the entire course of the Apalachicola River, from Chattahoochee to Apala-Samples were collected in chicola. 1983 and 1984, primarily during flood periods, and analyzed for fecal indicator bacteria. Field measurements of pH, temperature, and specific conductance were made at the time of sample collection. Concurrently with the river sample collections, regular monitoring of the bay by FDNR provided data which could be used to address the first objective. There are also data on river flow and rainfall for the period of the study and for many previous years.

#### METHODS

Twelve sites on the river were sampled for indicator bacteria. The 12 sites are listed in table 3, and their locations are illustrated in figure 3. Samples were collected at these sites nine times during the rise and recession of the 1983 spring flood, and again in December 1983 and March 1984. Sampling at the downstream sites (0-42), was done with at least a 1-day lag after the upstream sites to allow for time of travel.

At each site, grab samples were taken just below the surface in sterile plastic bags from a central position in the main stream. The samples were; accompanied by on-site measurements of temperature, pH, and specific conductance. Within 8 hours after collection, samples were analyzed for total coliform, fecal coliform, and fecal streptococci bacteria by the FDNR Shellfish Sanitation Laboratory in Apalachicola. Bacterial densities, as MPN, were determined by the multiple-tube fermentation technique (Greenberg and others, 1980).

Prior to the regular sampling of the 12 sites during the 1983 spring flood, 4 cross sections were sampled and analyzed by U.S. Geological Survey personnel using the membranefiltration (MF) technique (Greeson and  $\phi$  thers, 1977). The areas of this preliminary sampling were at river miles 106, 76, 26, and 21. The purpose of this sampling was to determine the extent of cross-sectional variation in bacterial counts. Tf the variation was high, and more importantly, if it conformed to some regular pattern (such as higher concentrations near the banks), it would suggest that sampling should be done as **cr**oss-section composites rather than center-channel grabs. There was also some analysis of variation with depth, and an initial analysis of downstream changes in total bacterial loads and fecal coliform:fecal streptococci (FC:FS) ratios.

Personnel of FDNR sampled the 28 sites in Apalachicola Bay for total coliform 60 times between January 1, 1983, and April 30, 1984. Most of the sampling was concentrated during the spring flooding periods of 1983 and 1984, sometimes being conducted at daily intervals. Sampling during low-flow periods was approximately monthly. The samples from the bay were analyzed for total coliform by

Site No.	Location	Ríver mile
106	Chattahoochee, below Highway-90 bridge	105.7
94	Ocheesee landing	94.0
86	Jollis landing, near Little Sweetwater Creek	85.8
80	Bristol landing	80.3
76	Blountstown, below landing	76.0
42	Glenn landing, near Wewahitchka	41.8
28	* Chipola River, mouth	27.8
12	* Brothers River, mouth	12.0
10	** St. Marks River, headwaters	10.3
6	* Jackson River, mouth	6.0
1	* Outlet of Scipio Creek	.5
0	Mouth of Apalachicola River, below bridge	0

Table 3.--Location of sampling sites on the Apalachicola River and its branches

[Tributary sites marked by asterisk; distributary sites marked by double asterisk]

the multiple-tube fermentation technique.

River flow was monitored continuously at river miles 106, 77, 42, and 21. Descriptions of those sites and the type of information available from them are given in table 4. Rating tables based on streamflow measurements (Leitman and others, 1983) were developed for each of the sites to permit conversion of river stage to discharge.

Rainfall data were obtained from monthly publications of National Weather Service rainfall records (National Oceanic and Atmospheric Administration, 1983-84). These data were obtained from rain gages located at Apalachicola and Wewahitchka, Fla., and Columbus and Bainbridge, Ga.

#### RIVERINE COLIFORMS AND STREPTOCOCCI Sampling and Analytical Variability

The results from sampling at four river cross sections in February 1983 were used to determine variability over spatial gradients in a transverse direction (river cross section), vertical direction (river depth), and longitudinal direction (river length). The two upstream cross sections at river miles 106 and 76 (1 km and 49 km downstream of the dam) were sampled February 18. The lower two cross sections at river miles 26 and 21 (130 km and 138 km downstream of the dam) were sampled 5 days later.

Table 5 shows the range, mean, and standard deviation of fecal coliform and fecal streptococcus counts taken at the surface at each cross section. The cross-sectional variability indicated by these data is no greater random variability. natural than This was confirmed by an analysis of variance (ANOVA) test of differences among sections with respect to fecal coliform (FC) and fecal streptococcus The test showed no sig-(FS) values. differences (probability nificant level = 0.10) for either variable.

FC data from samples which were taken at depths of 1, 2, or 4 m and at midchannel all fell within the ranges shown in table 5. This conformance was also true for FS data with the exception of a value of 850

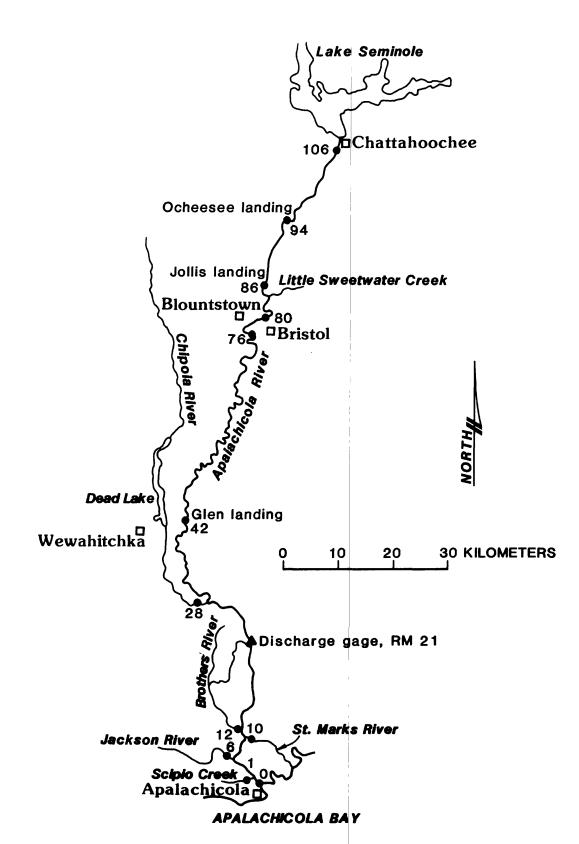


Figure 3.--Sampling sites on the Apalachicola River and its tributaries.

area
Surface-water gaging stations in the investigation a
the
in
stations
gaging
Table 4Surface-water
Table

Station name and No. <sup>1</sup>	River mile No.	o. Location	Period of record	Type of record
Apalachicola River Chattahoochee, 02358000.		106 On downstream side of right main pier on U.S. Highway 90, 1.0 km downstream from Jim Wood- ruff Dam, and 1.6 km west of Chattahoochee.	1928-84	Mean daily discharge.
Apalachicola River near Blountstown, 02358700.	17	On the right bank 152 m upstream from Neal Lumber Company landing, 2.4 km southeast. of Blounstown.	1920-57	Once daily stage and occasional discharge.
Do.		do.	1958-84	Mean daily discharge.
Apalachícola Ríver near Wewahitchka, 02358754.	42	On the right bank just above the Chipola Cutoff, 5.5 km east of Wewahitchka.	1965-84	Mean daíly stage and occasíonal díscharge.
Apalachicola River near Sumatra, 02359170.	21	On left bank at Brickyard landing, 3.9 km west of Fort Gadsden and 8.5 km southwest of Sumatra.	1950-59	Mean daily stage and occasional discharge.
Do.		do.	9/ 9/77- 9/30/84	Mean daily discharge.

<sup>1</sup> Station identification number.

River mile No.	Fecal coliform			Fecal streptococci		
	Range	Mean	Standard deviation	Range	Mean	Standard deviation
106	310-620	488	147	320-670	548	166
76	380-570	478	84	550-740	664	70
26	76-100	88	9	44-60	52	6
21	74-130	102	22	30-93	58	22

Table 5Cross-sectional	variation in fecal bacteria
at four river cross	sections, February 1983

[Data are from five surface samples per site, taken in approximately equidistant segments from left to right bank; all data are in number of colonies per 100 mL]

at 1-m depth, site 76, and a value of 70 at 2-m depth, site 26. These deviations were not statistically significant (probability level = 0.10).

The lack of significant crosssectional or depth variation in bacteria levels in the river implies that the normal sampling procedure of sampling near the surface at midchannel is adequate to represent conditions for that cross section of the river. This assumes that there are no point sources at the sampling site or just upstream from it. Ponded water very near the shoreline may not have the same characteristics as water in the main channel, but the through-flowing water, which is of primary interest with respect to downstream effects, is relatively uniform. Dissolved oxygen, specific conductance, and temperature data collected at several depths, and sections with a submersible water-quality monitor also showed insignificant variability, indicating thorough mixing in the river channel at various river stages.

One other test of methodology that was conducted with the February 1983 samples was to compare results of two analytical methods. Duplicate samples from sites 26 and 21 were provided to the FDNR Shellfish Sanita-

tion Laboratory where they were analyzed by the multiple-tube fermentaprocedure tion to determine MPN Corresponding samples were counts. analyzed by the membrane filtration technique by U.S. Geological Survey personnel. The results are shown in table 6. Analysis of variance results, also shown, indicate no significant differences in data obtained by the two treatments.

# Downstream Variability

The February 1983 data shown in table 5 suggest an appreciable difference between upstream and downstream sections of the river. ANOVA confirmed a significant difference between (probability level 0.01)counts at the two upper river sections versus the two lower river sec-Whether this difference was tions. due to an actual downstream decrease in bacteria counts or was simply attributable to the fact that the two lower river sections were sampled on a different day than the upper river sections is not clear from these data.

Further investigation of downstream variability of coliform bacteria was conducted using the samples taken during the March-April flood in 1983 and subsequent sampling in May

		rom analyses of	
e samples fro	m Apalachicola )	River sites 26	
February 23,	1983		
	Fecal	Fecal	
Section	coliform	streptococci	
	MF MPN	MF MPN	
	February 23,	Section <u>coliform</u>	

84

86

94

76

100

130

110

110

74

88

33

130

70

79

170

110

350

240

94

130

1

2

3

4

5

1

2

3

4

5

Table 6Comparison of membrane filtration (MF) and
most probable number (MPN) results from analyses of
duplicate samples from Apalachicola River sites 26
and 21, February 23, 1983

F = 2.29F = 0.04Analysis of variance  $(NS^1)$  $(NS^1)$ between methods

<sup>1</sup> Not significant at probability level = 0.10.

1983, December 1983, and March 1984. This included 10 samples from each of the 5 upstream sites (76-106) and 11 from each of the 7 downstream sites Means and standard devia-(0-42).tions of fecal coliform and total coliform data from each site are presented in table 7.

26

21

There is no evidence of any difference in coliform levels in any of the main channel sites from the headwaters downstream to site 42. Any differences that do appear are minor in comparison to natural and seasonal variability as indicated by standard deviations. However, below site 42, at sites 10 and O, а distinct decrease in FC levels was observed. This decrease was found to be highly significant (probability level 0.05) by an ANOVA test of combined data from sites 106-42 versus combined data from sites 10 and 0. The decrease in total coliform at the same sites was not significant at probability level = 0.10.

Coliform numbers in three of the four tributaries were not higher than the values observed in the main chan-The tributary at site 1 is nel. Scipio Creek, which receives effluent from the wastewater treatment facility at the city of Apalachicola. A1though its coliform levels were high, discharge in the Apalachicola its River did not reverse the tendency for coliform abundance to decrease in a downstream direction. It is possible, however, that Scipio Creek water does not thoroughly mix with river water before its outflow to the estuary.

49

79

33

26

79

70

46

79

49

23

60

48

44

53

54

54

54

93

30

60

FS counts were done only on the samples collected in February 1983, December 1983, and March 1984. The February data (table 5), again, gave strong indication of a decrease from upper to lower river cross sections. remaining data available are The shown in table 8. For the two dates shown (December 1983 and March 1984),

<u></u>	Fecal	coliform		Tota	l coliform
Site	Mean	Standard deviation		Mean	Standard deviation
		AMain chan	nel sítes	<u>•</u>	
106	327	508		727	499
94	250	382		1,380	1,010
86	282	522		1,140	518
80	278	508		1,620	1,280
76	253	302		696	<sup>´</sup> 567
42	118	114		877	465
10	83	68		614	664
0	48	37		464	476
		<u>BTributar</u>	y sites.		
28	88	73		585	435
12	83	96		472	431
6	44	36		332	302
1	488	651		3,050	3,930
	* *	· * * *	* *	* *	

# Table 7.--Fecal colifor and total coliform at Apalachicola Riversampling sites, March 8, 1983 through March 6, 1984

[Means and standard deviations are from 10-11 samples]

Table 8Fecal streptococcus	
December 19, 1983,	and March 5-6, 1984

Site	December 19, 1983	March 5 <b>-</b> 6, 1984	Site	December 19, 1983	March 5-6, 1984
<u>A</u>	Main channel sit	es.	<u>B</u> .	Tributary site	es.
106	9	17	28	93	240
94	43	27	12	240	33
86	43	23	6	93	79
80	43		1	93	140
76	20	13			
42	21	34			
10	240	110			
0	36	33			

[Values given in MPN per 100 milliliters]

there is no evidence of the downstream trend suggested by the February results. On the contrary, site 10 stands out as a point where FS levels were considerably higher than at other sampling points in the river. Scipio Creek (tributary site 1) did not contain FS counts higher than other tributaries, as it did for coliforms. Because the source of site 1 bacteria is predominantly human fecal material from the sewage treatment plant, they tend to be predominantly coliform (Geldreich and Kenner, 1969). At sites 10, 12, and 28, streptococci counts were as high or higher than coliform counts, suggesting greater importance of nonhuman sources in the drainage to these channels.

#### Fecal Coliform:Fecal Streptococcus Ratio

The FC:FS ratio is considered an indicator of the sources of the fecal bacteria, whether human or animal. Human fecal material contains a much higher FC:FS ratio than fecal material from other mammals and birds (Geldreich and Kenner, 1969; Wheater and others, 1979). Therefore, as general rule, the higher the FC:FS ratio, the greater the probability of human origin. Although theoretically sound, this rule is subject to frequent exceptions because of natural variability in both coliform and streptococcus numbers. Furthermore. survival rates are different for the two types of bacteria (Oragui and Mara, 1983), making it difficult to accurately monitor the true FC:FS ratio.

Despite the relatively high coliform counts in February 1983 in the Apalachicola River, the FC:FS ratio was less than 1.0 at the two upper river cross sections. It increased slightly at the downstream sites, averaging 1.7 at river mile 26, and slightly more than 2.0 at river mile 21. These ratios suggest animal origin for much of the fecal bacteria in the river water.

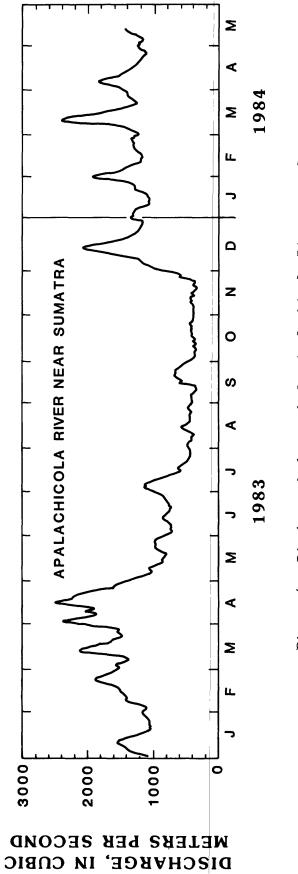
In the December 1983 and March 1984 sampling, the FC:FS ratio was much more variable and lacked any clear downstream trend. On December 19, the ratio ranged between 0.4 at site 10 to 23 at site 76. On March 5-6, the FC:FS range was narrower--1.25 to 8.5--but the minimum and maximum again occurred at sites 10 and 76, respectively. The high FC:FS ratio at site 76 is presumably attributable to its location less than 4 river miles downstream from the communities of Bristol and Blountstown and the wastewater treatment plant near Blountstown. It appears that the ratio tends to decrease from that point downstream, with minimum values at site 10. FC:FS ratios at all tributaries except Scipio Creek were generally less than 1.0.

The city of Chattahoochee at the northern end of the river also appeared to have an effect on FC:FS ratios of samples taken in December and March. The ratio was 10.3 on December 19 and 6.5 on March 5, strongly indicating bacteria of human origin. Two wastewater treatment plants in the area, one for the city of Chattahoochee and one for the Florida State Hospital, probably are the primary contributors to this source.

#### Relation to River Discharge

Because river discharge is monitored continuously at several points along the river, it is a convenient variable to use as an indicator of potential bacterial contamination. Furthermore, it has been documented (McDonald and Kay, 1981; Thompson and others, 1984) that higher discharge may bring with it higher bacterial loads. Therefore, an important part of this study was to investigate the relation between discharge and concentration of riverine fecal bacteria.

The discharge hydrograph of the Apalachicola River near Sumatra (figure 1) for the period of study, January 1983 through May 1984, is shown in figure 4. The river exhibited the characteristic pattern of multipeaked floods during March and April, preceded by some slightly smaller peaks in December through February. Low flows prevailed in late spring, summer, and fall, interrupted only by





minor peaks resulting from local heavy rainstorms. The range of discharge during the period shown was  $340 \text{ m}^3/\text{s}$  to  $2,500 \text{ m}^3/\text{s}$ .

Linear regression analyses applied to the river data from March 1983 to March 1984 indicated no significant correlations between discharge and bacterial concentrations at any of the main river sites except site 0, the river mouth. A plot of logtransformed FC data versus discharge is shown in figure 5 for site 0. The least-squares linear regression line and its correlation coefficient indicate a relatively strong relation between discharge and log-transformed Only one data point deviates FC. substantially from the model, showing a very low FC value at a discharge of over  $1,400 \text{ m}^3/\text{s}$ . This data point represented the sample of March 24, 1983, corresponding to the descending limb of the first flood peak.

Similar data for site 86 are plotted in figure 6. The higher degree of scatter in this data characterized all of the upper river sites and illustrates why the linear regression correlation coefficient was not significant. Visual inspection suggests, however, that a relation between FC and discharge exists and there were two deviant data points, both having low FC values. The two points, which are circled on the graph, represented data collected March 22 and March 28, 1983. As previously observed at site 0, the deviant values correspond to the descending limb of the first flood peak.

A possible interpretation of the site-specific FC versus discharge data is that there is a relation between the two variables, but superimposed upon it is a moderating effect of flood duration. As the flood progresses, FC levels are likely to decrease even though the stage remains high. The principal impact of the flood, therefore, would be expected during the first few days and on its rising limb.

This interpretation is supported by a time-sequence plot of FC versus discharge, illustrated in figure 7. The data points represent medians, within the ranges shown, for all of the five upper river sites (sites 76-106). Points representing the 1983 spring flood (March-May) are connected in chronological order of The first five samples sampling. (March 8, 9, 10, 14, and 22) showed a marked decline in FC (more than two orders of magnitude) whereas discharge actually increased from March 8 to March 9 and was still above flood stage at the end of this 2-week period.

When the second peak of the 1983 spring flood occurred in late March, discharge again exceeded 2,000 m<sup>3</sup>/s. At four of the five sites, FC also increased by approximately one order of magnitude over the previous sample. This was still far short of the abundance found on March 8 and 9, the first 2 days of the flood. Finally in May, both discharge and FC reached low levels characteristic of low-flow conditions.

The overall result of the combined effects of river discharge and flood duration is a spiraling descent of coliform numbers. As the river level fluctuates above flood stages during a single flood, the impact of each succeeding peak in transporting fecal bacteria is decreased. Similar discharges occurring at different phases of the flood are likely to transport progressively smaller amounts of fecal bacteria.

Further evidence of the increased flushing potential of initial flood peaks appears in the two data points from December 1983 and March 1984 (fig. 7). Those both represent low flood discharges (1,200-1,400 m<sup>3</sup>) which were independent of earlier

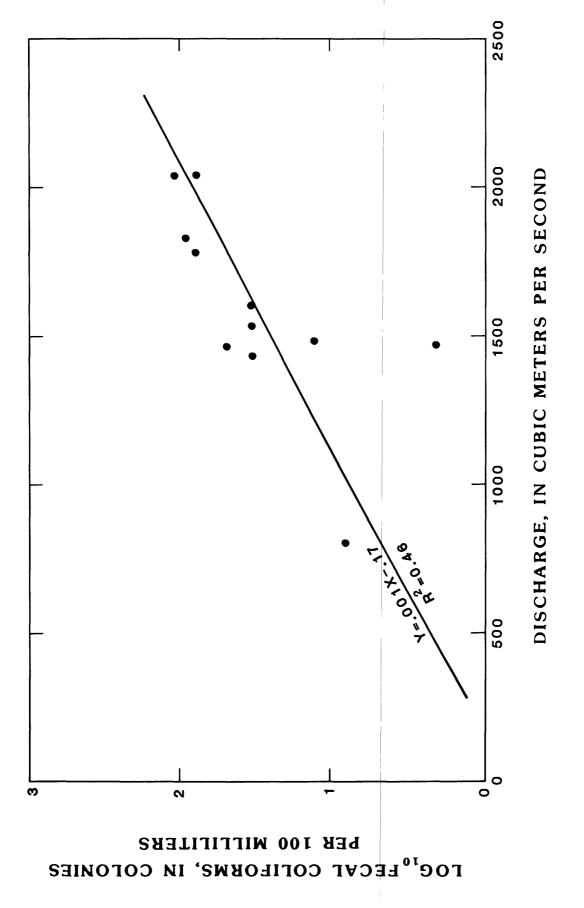
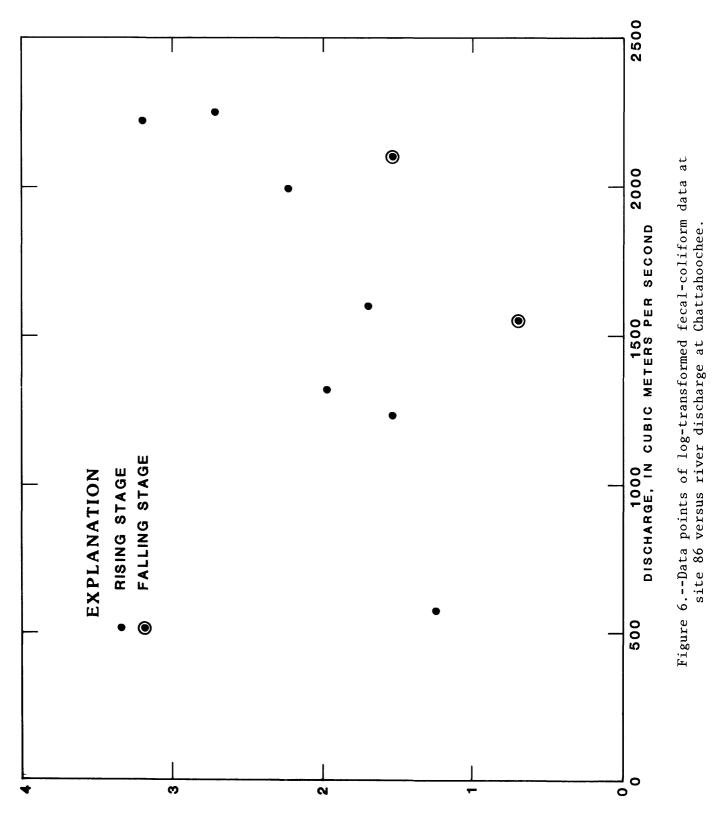


Figure 5.--Linear regression of log-transformed fecal-coliform data at river mouth (site 0) versus river discharge at Sumatra gage.



**DEK 100 MILLILIERS** LOG<sup>10</sup> FECAL COLIFORMS, IN COLONIES

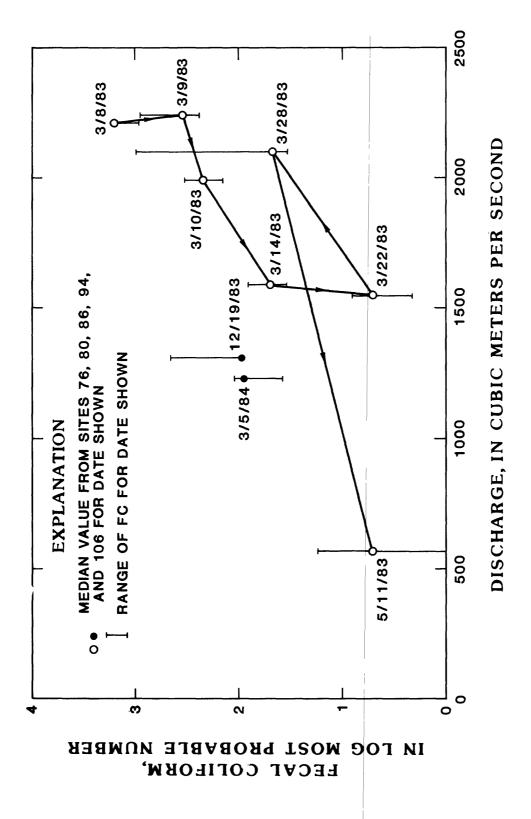


Figure 7.--Time-sequence plot of log-transformed median fecal coliform values from five upper river sites (sites 76, 80, 86, 94, 106) versus river discharge at Chattahoochee. floods. At these moderate discharges, FC counts were again quite high. The median counts were higher, in fact, than those of March 18, 1983, when discharge was greater by some  $850 \text{ m}^3/\text{s}$ . Because these samples represented independent hydrologic events, the FC transport was again more characteristic of initial phases of floods.

The presumed explanation of the downward spiral effect is that the first peak flushes the constituents that have been accumulating for some time before the flood while the river stage was low. After the initial flood peak, accumulation between peaks is relatively minor; thus, there is reduced flushing potential with each succeeding peak.

Bacteria data from the river mouth were aggregated with river discharge and stage data at the recording gage site near Sumatra to develop a model which would take into account the combined effects of river discharge, flood duration, and other hydrologic characteristics. The definitions of variables used in the model were based on the assumption that, for any particular flood, the fecal bacteria transport potential changes as the flood progresses. A single flood is considered to be the period of time that the river is continually above flood stage, regardless of the fluctuations that occur above that stage.

The hydrologic variables, other than discharge, which were used for development of the model are defined as follows (see table 9 for more detail):

- <u>i</u> = difference between the current stage and the highest previous stage in the current flood.
- m = limb designation.
- $\underline{d}$  = duration of the current flood, in days.
- P = imd, or the product of the pre-

vious three variables. This is a single factor which characterizes the combined effect of peak height relative to previous peaks, rising or falling river stage, and duration of the flood.

It was found that the logarithm of P was inversely related to FC numbers at the river mouth. A forward-stepping multiple regression analysis with discharge (Q) and log P as independent variables produced the model described by the equation:

 $FC = 0.06Q - 12.8 \log P + 11$ 

The correlation coefficient  $(r^2)$ of this model is 0.658, significant at a probability level of 0.01. In terms of standard partial regression coefficients (beta coefficients), the discharge (Q) accounts for 53 percent of the total variance in FC, whereas log P inversely accounts for 32 percent.

Table 9 gives the predicted FC numbers at the river mouth for selected hydrologic conditions at the Sumatra gage, as predicted by this model. Only two discharge values are shown; yet for each of these values, there is a wide range of predicted FC abundance, depending on the other variables, i, m, and d. As one or more of these variables increases, FC is expected to decrease. Based on this model, the influence of  $\underline{i}$ ,  $\underline{m}$ , and d is not as large as the discharge effect, but it is important enough to appreciably alter the simple FC-discharge relation, especially at lower discharge levels.

#### ESTUARINE COLIFORMS Abundance and Distribution

Periodic extensive monitoring of 28 sites in Apalachicola Bay is conducted by FDNR to provide a basis for management of the shellfish industry. Data from 235 sampling excursions (from January 10, 1979, to March 1, 1984) were compiled and analyzed in a report by Thompson and others (1984). Various statistical approaches were used to examine the relation between fecal bacteria at the estuarine sites and various hydrologic variables, including rainfall, river discharges, river stage, and ambient salinity and temperature. A multiple regression technique established that the two most important variables were river stage and 3-day rainfall (total rainfall occurring in the vicinity of the estuary during the previous 3 days). An equation describing this multiple correlation was used to create a

table of river stage and 3-day rainfall combinations which would constitute conditions for closure to harvesting.

Because riverine bacteria levels are at least partially dependent on river-basin hydrologic conditions, fecal bacteria populations in the estuary are also likely to be partially dependent on riverine hydrology. This relation is not apparent in analyses of the existing data, however, and cannot be demonstrated by statistical analyses without an extremely extensive data base. The association is partially masked by

- Table 9.--Predicted fecal coliform (FC) abundance at site 0 (river mouth) under selected hydrologic conditions at the Sumatra gage
  - [i = difference, in cubic meters per second, between the current stage and the highest stage to have previously occurred in the current flood.
  - m = limb designation. If the river is rising (stage is higher than previous day) m = 1. If the river is falling, m = 2.
  - $\underline{d}$  = duration of the flood, or the number of days since discharge at Sumatra surpassed 1,200 m<sup>3</sup>/s on the rising limb of the current flood.

Discharge (m <sup>3</sup> /s)	$(m^{3}\frac{i}{7}s)$	(1 or 2)	(days)	FC (MPN per 100 milliliters)
1,400 1,400 1,400 1,400 1,400	1 10 10 10 30	1 1 1 1 1	1 10 30 30	90 78 65 59 53
1,400	30	2	30	49
2,500 2,500 2,500 2,500 2,500 2,500 2,500	1 10 10 30 30	1 1 1 1 2	1 10 30 30 30	153 140 127 121 115 111

P = imd, or the product of the previous three variables]

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variations in moderating factors such as time of travel, survival time, and tidal effects.

Other patterns of bacterial distribution in the bay are detectable from the FDNR data. The data collected between January 3, 1983, and April 13, 1984, (60 sampling trips at the 28 bay sites) were analyzed for areal differences and dependence on other factors. Since the effects of temperature and salinity on fecal bacterial abundance were already described by Thompson and others (1984), that aspect will not be examined further in this report. The primary focus of the following discussion is the variation of bacterial abundance with location in the bay.

Apalachicola Bay is separated into winter and summer shellfish harvesting areas, and prohibited areas (fig. 2). These areas were established by law, according to measured bacteriological water-quality data relative to ISSP guidelines. The map also shows the location of the sampling sites within the seasonal harvesting areas. Some sites (10, 24, 26, and 39) are on boundaries between two or three areas.

Data analyses for this study were done by groups of sites to test differences between areas. There was also some subgrouping within sites to test for possible smaller scale regional differences.

The winter harvesting area includes 11 sites in the western part of the bay near St. Vincent Island. It also contains a section offshore from Eastpoint which bisects the summer harvesting area. Two sites (14 and 16) are located in this eastern section of the winter area. Site 10, on the winter-summer boundary was also included in the winter area for the purpose of data analysis.

The prohibited area near the river mouth contains only two sites (25 and 28). Boundary sites 26 and 39 were included with five other sites (19, 22.1, 24, 38, and 41) to characterize the western segment of the summer harvesting area.

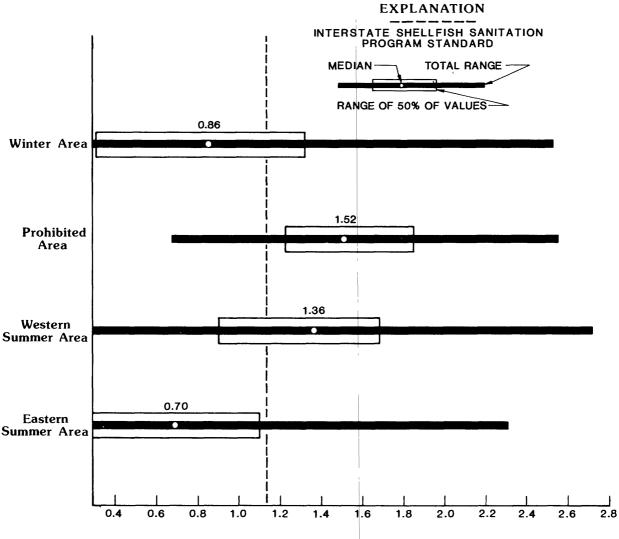
The comparisons of fecal bacterial abundance in the different areas are illustrated by box plots in figure 8. The logarithms of FC numbers are used, reflecting a range over more than 2 factors of 10. Around the median points, whose values are given, the body of the boxes illustrate the range of data quartiles. Half of the data points are within that range. The remaining data are scattered with the range shown by the lines extending beyond the quartile limits.

The prohibited area and the western summer harvesting area are shown by this analysis to be similar to each other and relatively high in FC The winter harvesting area counts. and eastern summer harvesting area are similar to each other and relatively low in FC abundance. Both have median coliform numbers less than one-quarter of the prohibitedarea median. Because of a large number of samples showing undetectable counts in these areas (less than 2 MPN per 100 mL), their distributions are sharply skewed toward low values.

Factors which are associated with the distribution of coliforms in the estuary were discussed by Thompson and others (1984). In addition to rainfall and river discharge, important influencing factors included temperature and salinity. water These factors were found to be closely correlated to FC densities at all sites in the winter harvesting area (fig. 2) and some sites in the summer harvesting area.

Some subgroup analyses of data from the winter area were done to determine if there were any differences in coliform counts between nearshore and offshore sites, or be-

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LOG<sub>10</sub> FECAL COLIFORM COUNTS PER 100 ML

Figure 8.--Range, median, and variability of fecal coliform data, January 1983-April 1984, by harvesting areas in Apalachicola Bay. (Data from Florida Department of Natural Resources.)

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tween western sites (near St. Vincent Island) and eastern sites (10, 14, and 16). No evidence of any such intra-area variation was found.

### Coliforms as Contamination Indicators

Because of the economic importance of the Apalachicola Bay shellfish industry, the transport and survival of pathogenic organisms which may find their way to the tissues of major marketable shellfish species are of major concern. As filter feeders, shellfish nonselectively ingest particles suspended in the water, including bacteria. Wood (1979) pointed out that bacteria are generally not killed in the digestive tract and they do not harm the shellfish; hence, they tend to accumulate in the gut without causing any ill effects in the health of the host. On the other hand, the shellfish species generally depurate rapidly when water quality improves; bioaccumulation of pathogens may be expected to be only a temporary condition reflecting high pathogen occurrence in the ambient water.

Although these principles of bioaccumulation would suggest a close correlation between bacteriological water quality and pathogenic contamination of shellfish, coliform counts in water samples are generally found to be a rather poor index of shellfish contamination. There are several reasons for this:

- □ Bacterial incorporation from water to biological tissues is highly variable, affected by numerous environmental factors (pH, temperature, salinity, and others), feeding rates, food organisms available, physical disturbances, concentrations of suspended matter, and most of all, individual and species differences (Wood, 1979).
- Coliform bacteria can reproduce rapidly in enriched waters where-

as populations of the true pathogens may be stable or even diminishing (Dutka, 1979). Thus, high total coliform counts may give a false indication of a hazardous condition.

- $\Box$  Another type of false alarm may be caused by the existence of Klebspecies which are not siella associated with fecal pathogens, but are hard to distinguish from total coliforms by the standard counting procedures (Dutka, 1979). Klebsiella species generally originate from industrial effluents. Improvements in the selectivity and sensitivity of analytical procedures are clearly needed to enhance the reliability of coliform as an index organism.
- □ Batik and others (1983) monitored drinking waters. coliforms in some of which had been found to produce disease outbreaks. No statistical difference in coliform counts was found between waters which caused disease outbreaks and those which did not. If coliform counts are not a good index of water quality, they would seem highly questionable as a good index of bioaccumulation in organisms inhabiting the aquatic system.
- □ Another analytical problem described by Xu and others (1983) which may produce underestimates of potential hazards is that coliform cells may exist in samples in a nonrecoverable but still viable form. Thus the MPN or MF techniques may detect little or no bacteria, even when viable organisms are present.
- Different species have very different survival potential (Oragui and Mara, 1983). The indicator bacteria Escherichia coli and various fecal streptococcal species have much shorter survival times than some other species.

This factor might produce underestimates of potential hazards.

Many pathogens are not of human fecal origin. Their abundance is thus independent of the abundance of FC (Blake and Rodrick, 1983). Examples are viruses and several species of <u>Vibrio</u>, which can cause gastrointestinal and wound infections.

Studies done in other surfacewater systems (Al-Mossawi and others, 1983; Volterra and others, 1984) have suggested that the use of a bivalve species as a biomonitor is a more accurate index of pathogenic contamination than is direct testing of However, such a prowater samples. cedure done on a routine schedule would require more elaborate sample preparation prior to analysis, making it considerably more time consuming and expensive. Additional variability would result from the introduction of another biological medium into the analytical procedure.

#### Consideration of Hydrologic Variables

Assuming that FC counts will continue to be the primary criterion for managing shellfish harvesting, relation between coliform abundance and river-basin hydrology is critical information. The Apalachicola Bay study by Thompson and others (1984) provided insight into the complexity of the relation by consideration of multiple hydrologic variables which could impact coliform abundance. The resulting multiple-regression equation was used to provide a management plan which takes into account the most important observed variables -river stage, 3-day rainfall, and season. One additional step in understanding the coliform-hydrology relation is to consider the effects of flood duration. Such consideration may be useful for continued refinement of management strategy.

the observations of this From study, it appears that the relation between estuarine coliform abundance and river stage changes during any particular flood. Thus, a hazardous condition which forces closure of shellfish harvesting may be triggered by a rising flood, especially if it follows a prolonged dry period, but the hazardous condition may subside a few days later, even if the flood continues. Winter floods on the Apalachicola are often of several weeks, or even months, duration. Closure of the bay solely on the basis of high river stages could result in unnecessarily long interruptions of utilization of the shellfish resource.

By means of the model described in this report, the effects of various hydrologic variables can be described. This provides some refinement over the limited capability of predicting FC abundance solely on the basis of As more data and hydrodischarge. logic records are collected, additional predictive capability will be possible. The benefits of developing a comprehensive and usable model will be manifested in improved efficiency and practicality of management policies.

#### SUMMARY

Coliform and streptococcal bacteria in the Apalachicola River and estuary waters show considerable relation to river stage, but the relation is modified by various hydrologic and other environmental factors. It is difficult to predict with any reasonable accuracy the coliform or streptococcus transport based purely on river stage. Even if such predictions were possible, the reliability of coliform counts as an index of pathogen contamination of shellfish meats is uncertain.

A large data base from coliform monitoring at 28 sites in the estuary makes it relatively easy to examine patterns of coliform abundance in different areas of the estuary. This type of analysis indicates that estuarine waters near the river mouth are higher than other areas in coliform abundance.

Samples collected from multiple vertical sections across four river cross sections showed only random variations in coliform or streptococcus abundance. There were differences among different cross sections, however. Coliform numbers were generally lower near the river mouth than farther upstream. FC:FS ratios were generally less than 2.0, indicating a nonhuman predominance of origin. Impacts of wastewater treatment plants on the river were shown in higher FC:FS ratios (up to 23).

Riverine transport of coliform and streptococcal bacteria is dependent

on discharge as well as other hydrologic characteristics of the flood. Bacteria may be flushed out of the system by the initial flood wave. Hence, bacterial transport tends to be highest on the rising limb of a flood peak, on peaks which are of the greatest amplitude in the flood, and on the early peaks.

The regression model which is defined in this report takes into account some of the important flood characteristics that modify the coliform-discharge relation. It is subject to further refinement as more information about fecal bacteria in the riverine and estuarine system becomes available. Development and continued refinement of a comprehensive model should serve to facilitate management decisions.

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