



BACTERIAL CONTAMINATION RELATED TO HARBOR SEALS IN PUGET SOUND, WASHINGTON

Final report to

Jefferson County
and
Washington Department of Ecology

In cooperation with

Washington Department of Social and Health Services

By

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"whenever a man gets the idea that he is going to work out the bacteriology of the intestinal tract of any mammal, the time has come to have him quietly removed to some suitable institution."

Attributed to Jordan in Miller 1959.
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EXECUTIVE SUMMARY

High concentrations of fecal coliforms have been reported at several sites in northern Hood Canal where hundreds of harbor seals congregate to haul out and rest. A preliminary study concluded that harbor seals had the potential to be major sources of fecal coliform concentrations in these area. Even though the management options are limited for controlling contamination from a natural source, such as seals, information on their role in bacterial contamination is important because:

- 1) Effective mitigation of human and domestic animal sources of contamination cannot be conducted without information on whether these sources are indeed responsible for the problem in an area.
- 2) Public support for measures to reduce human and domestic animal sources is compromised if there is a perception that a major alternate source of contamination is not being examined.
- 3) Information is needed to determine the areas where future problems for shellfish production are likely to occur.
- 4) If fecal contamination in an area is predominantly of seal origin then the degree to which this contamination represents a health hazard needs to be determined.

In this study we evaluated harbor seal contributions to bacterial contamination and the health risk, if any, they pose. The objectives of the research were as follows:

- 1) Determine the number of harbor seals occurring at haul-out areas in northern Hood Canal and evaluate the trend in population size.
- 2) Determine fecal coliform concentrations in harbor seal feces and identify factors that may alter contamination.
- 3) Examine fecal coliform contamination in water and shellfish at a site with high concentrations of seals and negligible other sources of contamination.
- 4) Examine the bacterial contamination contributed by seals in a closed captive environment.
- 5) Evaluate the evidence that harbor seals carry diseases transmissible to humans.

Our censuses indicated harbor seal numbers have significantly increased since 1984 at study sites in northern Hood Canal, once the effect of other variables including season and tide were taken into account. Two aerial survey counts indicated a minimum of 1,400 seals in Hood Canal, with up to 403 seals in Quilcene Bay.

Fecal coliform densities in harbor seal feces varied significantly by site, with dramatically lower concentrations in feces from captive seals. Significant differences also were found among the three sites where feces of wild seals were collected. Sampling variables may have accounted for some of these observed differences. Captive seal studies provided useful information on the dissolution of feces in the water column in a closed environment and demonstrated some of the limitations in estimating the contribution of fecal coliforms from seals in the environment. The large differences in fecal coliform densities in captive seals compared to those in the wild, however, limited the comparability to the natural ecosystem.

High levels of fecal coliforms were found in water and shellfish in Still Harbor, an embayment of McNeil Island that is the largest haul-out area for harbor seals in Puget Sound. Fecal coliform concentrations in both water and shellfish were highest at stations closest to the haul-out area. Bacteria also entered the bay from several small seasonal streams entering the harbor. The fecal coliform loading of these streams was far less than that calculated for seals, and the distribution of contamination was not consistent with these streams being the major source of fecal coliforms.

Factors consistent (+), inconsistent (-), or unknown (?), with seals being responsible for bacterial contamination of three regions are as follows:

Factor	Dosewallips	Quilcene	Still Hbr.
High contribution by seals	+	+	+
Lack of other sources	+	-	+
Contamination concentrated near seal haul-out areas	+	-	+
Historical contamination patterns	-	?	?

We conclude that the bacterial contamination at Dosewallips River Delta and at Still Harbor appears to be caused primarily by harbor seals. The role of seals in the bacterial contamination at Quilcene Bay is harder to determine because a number of other sources of contamination have been

identified and there is no evidence that contamination is highest at the seal haul-out areas. Continued increase of harbor seal populations in Puget Sound will only increase the potential for conflicts involving harbor seals and shellfish operations. The human health threat posed by seal fecal contamination cannot be determined with existing data.

Three avenues of future research are required to identify further the degree to which seal-related contamination poses a problem:

- 1) Develop techniques to identify whether bacteria in marine water and shellfish are from seals or from humans or domestic animals.
- 2) Examine the distribution of bacterial contamination in water and shellfish at Quilcene Bay and other sites with large numbers of seals to evaluate the source of contamination, as was done at Dosewallips and Still Harbor.
- 3) Test seal wastes for the presence of pathogenic organisms to evaluate the human-health risks of this contamination.

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INTRODUCTION

Bacterial contamination has been found in water and shellfish from several areas in Hood Canal, including Quilcene Bay, Dosewallips River Delta, and Duckabush River Delta (Cook 1984, 1985, Welch and Banks 1987, DSHS 1987, 1988). Harbor seals (Phoca vitulina) inhabiting these waters may contribute to the bacterial contamination (Calambokidis and McLaughlin 1987, 1988, DSHS 1988). The potential role of marine mammals as a source of bacterial contamination has not been examined in other areas. Most research on sources of non-point bacterial contamination have focused on human and domestic animals. Intensive studies in other areas of Puget Sound have identified livestock, municipal discharges, and failing septic systems as responsible for bacterial contamination (Determan et al. 1985, Taylor 1984).

Information on marine mammal contributions to bacterial contamination is valuable although management options for regulating seal populations are limited. Marine mammals are currently protected under the Marine Mammal Protection Act of 1972. This legislation limits the management options available to state agencies in regulating seal numbers. An understanding of the role of marine mammals as a source of bacterial contamination, however, is important for several reasons:

- 1) Effective mitigation of human and domestic animal sources of contamination is impossible without information on whether these sources are indeed responsible for the problem in an area. The degree to which these controllable sources of contamination are responsible for contamination cannot be accurately assessed without information on all sources of contamination.
- 2) Public support for measures to reduce human and domestic animal sources is compromised if there is a perception that another major source of contamination is not being examined.
- 3) Information is needed to determine areas where future problems for shellfish production are likely to occur. Harbor seal populations in Washington State are increasing rapidly (Calambokidis et al. 1979, 1985, 1988, Beach et al. 1985). If seals are currently major contributors to contamination in some areas, this problem will increase as seal numbers increase. Information on seal distribution and abundance can be used to identify the areas where problems may occur in the future.

4) Current bacterial tests rely on fecal coliforms as an indicator of fecal pollution and associated pathogenic organisms. Organisms pathogenic to humans occur in the feces of humans and domestic animals. The degree to which fecal coliforms accurately indicate the potential health hazard to humans when the fecal contamination is from seals has not been determined. If pathogens in seal feces are not zoonotic (transmittable from animals to humans) then the use of fecal coliforms as an indicator of the threat to humans is inappropriate in areas where seals are the primary source of bacterial contamination.

5) Non-lethal management of seals is possible under the MMPA in areas where the seals can be demonstrated to pose a health risk to humans.

We conducted research on the role of seals as a source of fecal contamination under contract 88-001 to Cascadia Research from Jefferson County with funding from the Washington Department of Ecology.

GENERAL APPROACH AND OBJECTIVES

We evaluated harbor seal contributions to bacterial contamination and the potential risk, if any, they pose to human health. Because little research has been done examining the role harbor seals play in bacterial contamination, a number of topics required further examination. Our approach was to examine harbor seals at three sites in Puget Sound and in a captive environment, and quantify the variables necessary for calculating the contributions of harbor seals to fecal coliform contamination at these sites. The objectives and rationale of the research were as follows:

Determine the number of harbor seals occurring at haul-out areas in Northern Hood Canal and evaluate the trend in population size.

Estimates of seal abundance are required for determining the fecal contribution by seals and to determine if populations are increasing and thereby likely to cause increasing difficulties. Numbers of harbor seals utilizing haul-out areas is highly variable. Frequent counts are required to reliably assess the average number of seals at a site and factors, such as season, that affect seal use of a site.

Determine fecal coliform concentrations in harbor seal feces and identify factors that may alter contamination. Information on fecal coliform concentrations in feces was needed to determine the total fecal coliform load produced by seals in different areas. Previously we had quantified fecal coliform concentrations in 10 seal feces from a single site (Calambokidis and McLaughlin 1987). The high variance and small sample size made extrapolation of these results to the entire harbor seal population at a site, risky. Additional samples were needed to provide a more accurate estimate of average fecal coliform concentrations. Samples of feces at additional sites were used to determine if location or diet altered fecal coliform concentrations, therefore compounding application of data from one to site to another. Potential sources of bias in quantifying fecal coliform concentrations needed to be examined. These included the effect of collection time and submersion in water on fecal coliform concentrations in feces.

Examine fecal coliform contamination in water and shellfish at a site with high concentrations of seals and minimal other sources of contamination. At many areas with high bacterial contamination, a variety of potential sources of contamination exist. This complicates identifying the role of a single potential source. A test of whether harbor seals can be responsible for bacterial contamination can be conducted at a site with high seal numbers in an embayment that has

few alternate sources of contamination. The largest concentration of harbor seals in Puget Sound is in Still Harbor, an embayment with few other sources of contamination. If harbor seals have the potential to cause bacterial contamination then this site should show high levels of contamination.

Examine the bacterial contribution of seals in a closed captive environment. Only in a closed environment can seal impacts on bacterial contamination of marine water be precisely quantified. Seals in an enclosed tank provide a means of simultaneously examining all variables associated with bacterial contamination. The degree to which the fecal coliform levels in the feces of captive seals deviates from wild populations must also be tested.

Evaluate the evidence for harbor seals carrying diseases that might be transmitted to humans. Fecal coliforms only serve as an indicator of fecal contamination. Human health is threatened by other pathogenic micro-organisms in a concentration too low to be tested economically. Fecal coliforms are good indicators only if they are correlated with the presence of pathogenic bacteria. An examination of bacteria reported for all marine mammals provides some insight into possible bacteria present in harbor seals.

METHODS

Census Methods

Harbor seal census results at sites in northern Hood Canal and Still Harbor for 1988 were analyzed in conjunction with data gathered in 1977 (Calambokidis et al. 1978, 1979) and from 1984 to 1987 (Calambokidis et al. 1985, Calambokidis and McLaughlin 1987, 1988). This allowed an evaluation of current abundance, annual rates of change, and other factors affecting the number of seals observed at haul-out areas.

In 1988, land-based censuses at seal haul-out areas in Hood Canal and Still Harbor were conducted using methods similar to those described in Calambokidis et al. (1985) and Calambokidis and McLaughlin (1987). Two aerial surveys were conducted of the entire Hood Canal in September 1988 to provide near simultaneous counts of harbor seals within the region during the pupping season. Aerial surveys were conducted from single-engine high-wing aircraft with three observers. Both visual and photographic counts were used to determine the number of seals and pups present at each site during aerial surveys.

Land counts were conducted at or near high tide in Hood Canal and at or near low tide in Still Harbor, and corresponded to the anticipated times of maximum numbers of seals at these haul-out sites (Calambokidis et al. 1978, 1985). Census effort at sites in northern Hood Canal since 1984 is summarized in Table 1. During each visit, seals were counted every 30 minutes. Information recorded on data sheets included numbers of seals hauled and in the water, number of pups, and weather conditions. Counts were made using binoculars and a 15-60X spotting scope. Specific locations and procedures used for land counts at each site are provided below.

Quilcene Bay: Land-based counts of harbor seals on log booms in Quilcene Bay (Figure 1) were made from East Quilcene Bay Road just above the logbooms. This site provided a close view of seals and good elevation. Counts of seals hauled on oyster rafts in the southeast part of Quilcene Bay were generally made from the Quilcene yacht basin (Boat Haven). Censuses of both areas were included in all censuses after 11 September 1986. The Boat Haven site was a relatively poor one due to the distance from the seals and lack of elevation.

Dosewallips River Delta: Harbor seals at the Dosewallips River Delta were counted from a clearcut just south of the delta. Despite the distance from the areas used by the seals, the elevation of this site allowed more accurate counts than closer locations. This site was used primarily after

Table 1. Census effort at sites in northern Hood Canal.
 Number of censuses indicates separate visits to
 site usually at or near high tide.

Site	Number of censuses		Hours of effort	
	1988	1984-88	1988	1984-88
Quilcene	32	97	39	95
Dosewallips	23	131	12	155
Duckabush	24	97	13	100
TOTAL	79	325	64	350

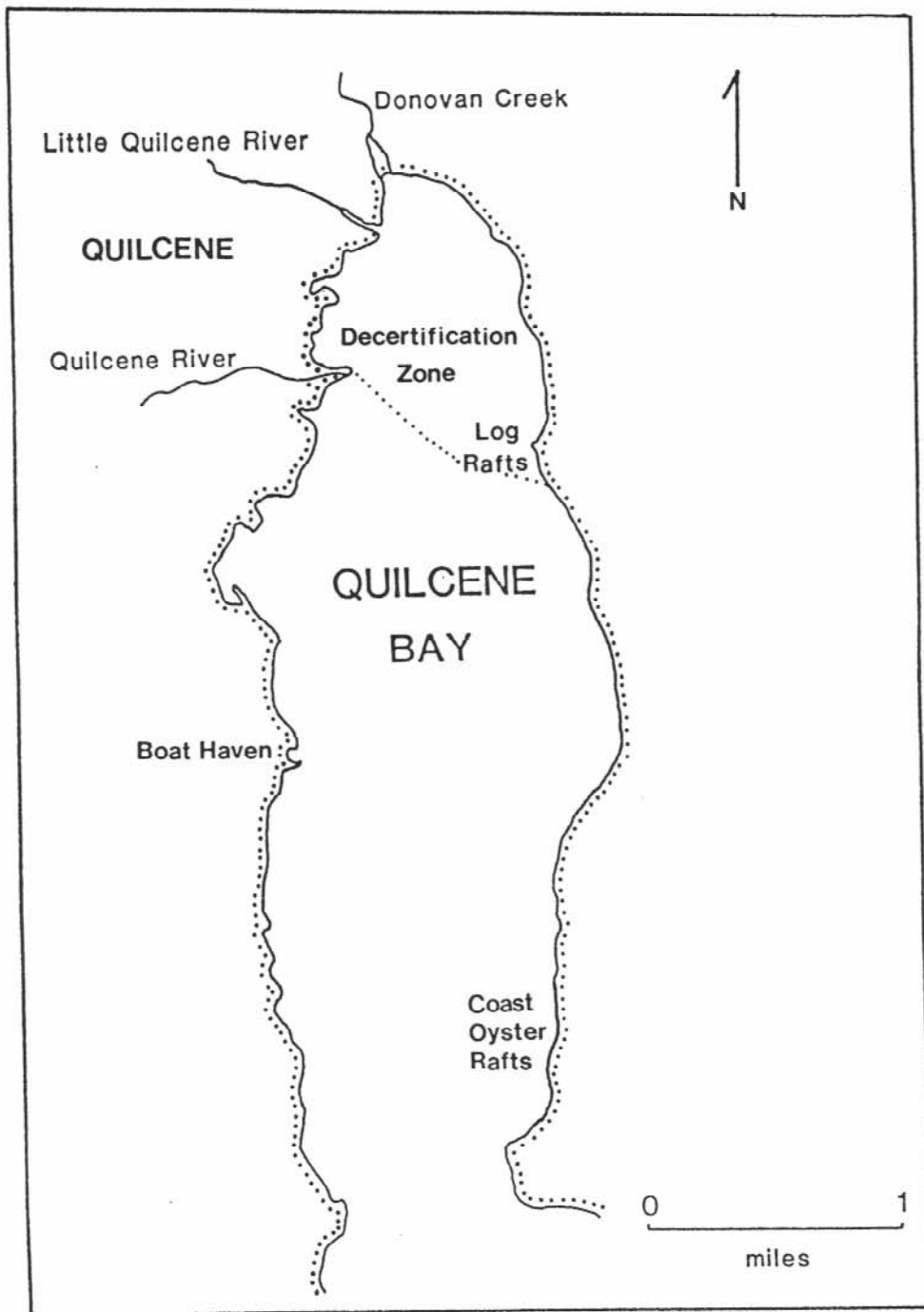


Figure 1. Quilcene Bay showing the locations of log booms and oyster rafts used by seals to haul out. Dotted line indicates the boundary of an area decertified for commercial shellfish growing.

30 October 1985. Before this, counts were made from multiple locations to allow coverage of the entire delta.

Duckabush River Delta: Land counts of harbor seals at the Duckabush River Delta were conducted from three different locations along Highway 101, including occasional counts from atop the bridge over the Duckabush River.

Still Harbor: In 1988, land counts of harbor seals on Gertrude Island were made from Still Harbor, McNeil Island (Figure 2). Because seals generally occupied both the east and west sides of the spit at the south end of Gertrude Island, censuses were generally conducted from two locations, the end of the Still Harbor dock and from a blind located on the southeast shore of Still Harbor. Counts were made by different observers communicating by radio. Radio contact allowed resolution of any confusion about counts of seals visible from both locations.

Feces collection

Seal feces were collected at four locations to determine fecal coliform density in feces, examine food habits of seals, and conduct dissolving and other experiments (Table 2). Feces were collected for fecal coliform determination only when their location or appearance indicated they were deposited recently. Scats were collected at the Dosewallips Delta in the early morning soon after a high tide to minimize the time between defecation and collection and to insure that scats were as cold as possible. Similarly, scats were collected immediately after seals were hauled out on floats in Quilcene Bay and at Gertrude Island in Still Harbor. Feces were also collected from tanks with captive seals at an aquarium. Scats were weighed, then transported in cooled sterile containers to the Department of Social and Health Services (DSHS) Public Health Laboratory in Seattle. Time from collection to analysis never exceeded 24 hours.

Two experiments were conducted to examine how sampling strategies may affect fecal coliform densities. We tested whether short variations in the time between defecation and collection or the submersion of feces in saltwater altered fecal coliform densities. Two feces on rafts in Quilcene Bay were subsampled on three consecutive days (the first collection was made immediately after defecation). Samples were marked and remained undisturbed by placing a small wooden frame placed around them. The frame did not alter the feces exposure to precipitation or sunlight. Subsamples of three feces were also submerged in saltwater for 36 to 48 hours to determine if occasional submersion in saltwater, which likely occurred with some of our samples, altered fecal coliform densities.

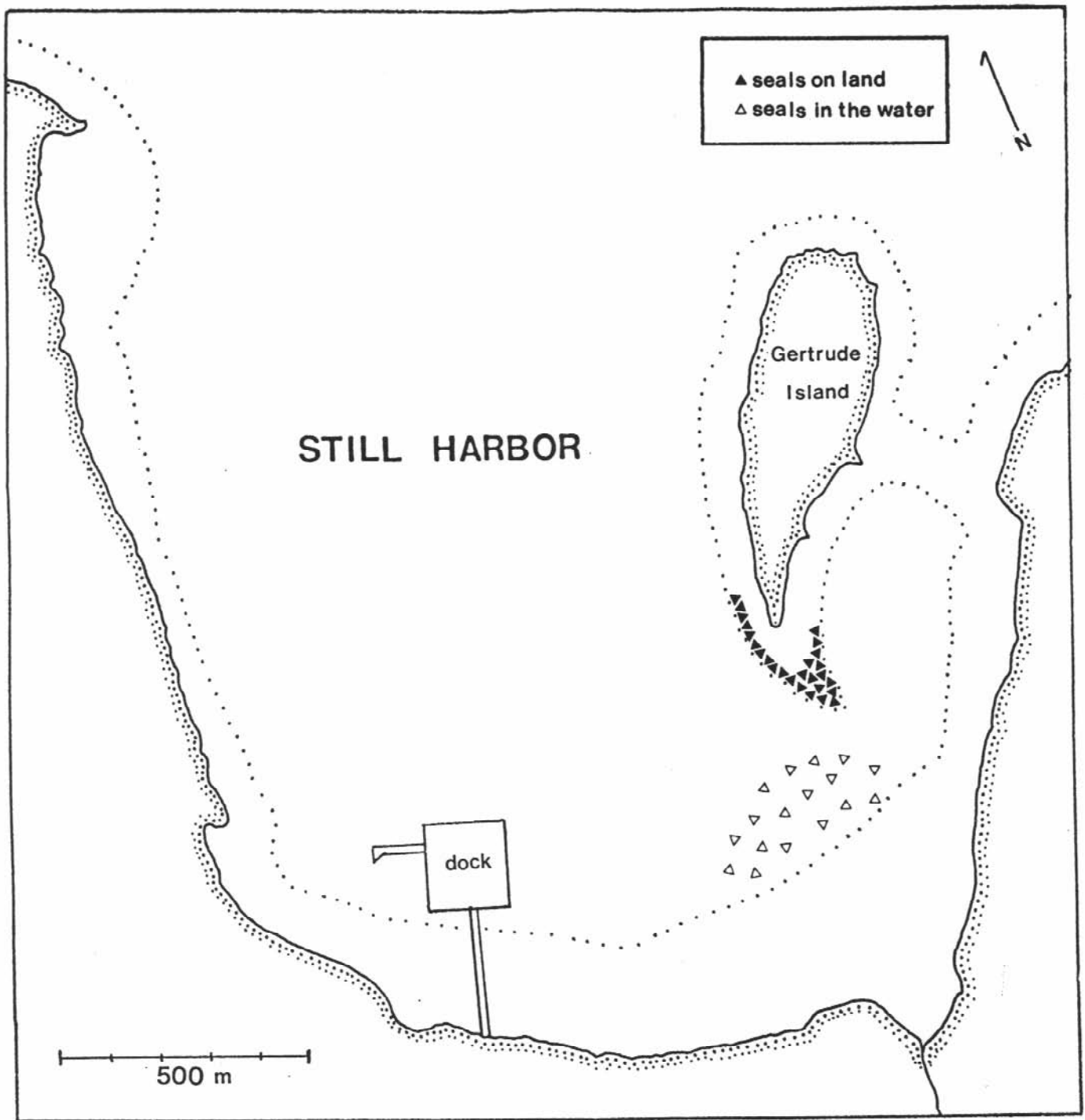


Figure 2. Still Harbor and Gertrude Island showing typical locations where seals haul out and congregate in the water. Dotted line indicates approximate Mean Lower Low Water.

Table 2. Harbor seal feces collected for food habits and fecal coliform analyses.

Date collected	Sample numbers	Total collected	Number analyzed	
			food habits	FC levels
<u>Dosewallips</u>				
2 May 88	DO 1-3	3	3	3
16 May 88	DO 4-11	8	7	8
19 Jun 88	DO 12	1	1	1
20 Jun 88	DO 13-17	5	2	5
24 Jul 88	DO 18-21	4	2	4
Total		21	15	21
<u>Quilcene</u>				
28 Nov 88	QU 1-6	6	6	-
4 Dec 88	QU 7-20	14	14	5
5 Dec 88	QU 21-22	2	2	1
6 Dec 88	QU 23-24	2	1	1
Total		24	23	7
<u>Gertrude</u>				
8 May 88	GI 1-11	10	9	9
22 May 88	GI 12-16	5	5	4
10 Jul 88	GI 22-24	3	2	2
2 Aug 88	GI 27-28	2	2	2
Total		20	18	17
<u>Aquarium</u>				
2 May 88	AQ 1-2	2	-	2
9 May 88	AQ 3-4	2	-	2
16 May 88	AQ 5-6	2	-	2
6 Nov 88	AQ 7A	1	-	1
8 Nov 88	AQ 7B-9	2	-	2
Total		9	-	9
All sites		74	56	54

Harbor seal food habits

Seal scats were also examined to determine the food consumed by seals. Scats used for fecal coliform analysis as well as other scats were screened through nested 2.0 to 0.5 mm screens. Otoliths (ear bones in bony fishes that are often not digested completely) recovered from scats were compared to a reference collection at Cascadia Research to identify fish species. The length of otoliths was measured with calipers and the degree of wear or damage scored. Any other identifiable prey remains (eg. cephalopod beaks) also were used to determine food habits.

Dissolving experiment

Experiments were conducted with 10 seal feces samples to examine how readily they dissolved in saltwater. Procedures were as follows:

- 12-16 liters of seawater were poured into a clean 5 gallon bucket
- a water sample was collected from seawater in the bucket
- approximately 10-20 g of feces were placed into the bucket without mixing
- water and feces were agitated for 30 seconds with gentle circular stirring with a rod
- 30 seconds after end of stirring the second water sample was taken
- water was allowed to rest undisturbed for 1 hour in cool shady spot
- the third water sample was taken
- water was agitated for 30 seconds as above
- after 30 seconds the fourth water sample was taken
- water and feces samples were kept on ice until delivered to the lab

Water and shellfish sampling

Seawater and shellfish samples were collected from Still Harbor, McNeil Island, an area where large numbers of harbor seals congregate. Water samples were collected from 9 sampling stations, 8 inside the harbor and one control station outside the harbor (Figure 3). The shoreline of the harbor was searched on each visit and samples taken at each input of water into the harbor. These freshwater sources were sampled at the culverts where they drained into the harbor. Flows were measured by the time taken to fill a 2 liter container. Sampling was conducted on 4 days, two sets of samples were taken each day from the 9 sampling stations, typically the first near low ebb tide and the other at flood tide.

Ten samples of littleneck clams (Protothaca staminea) were taken from the Still Harbor area for fecal coliform analysis. Each sample consisted

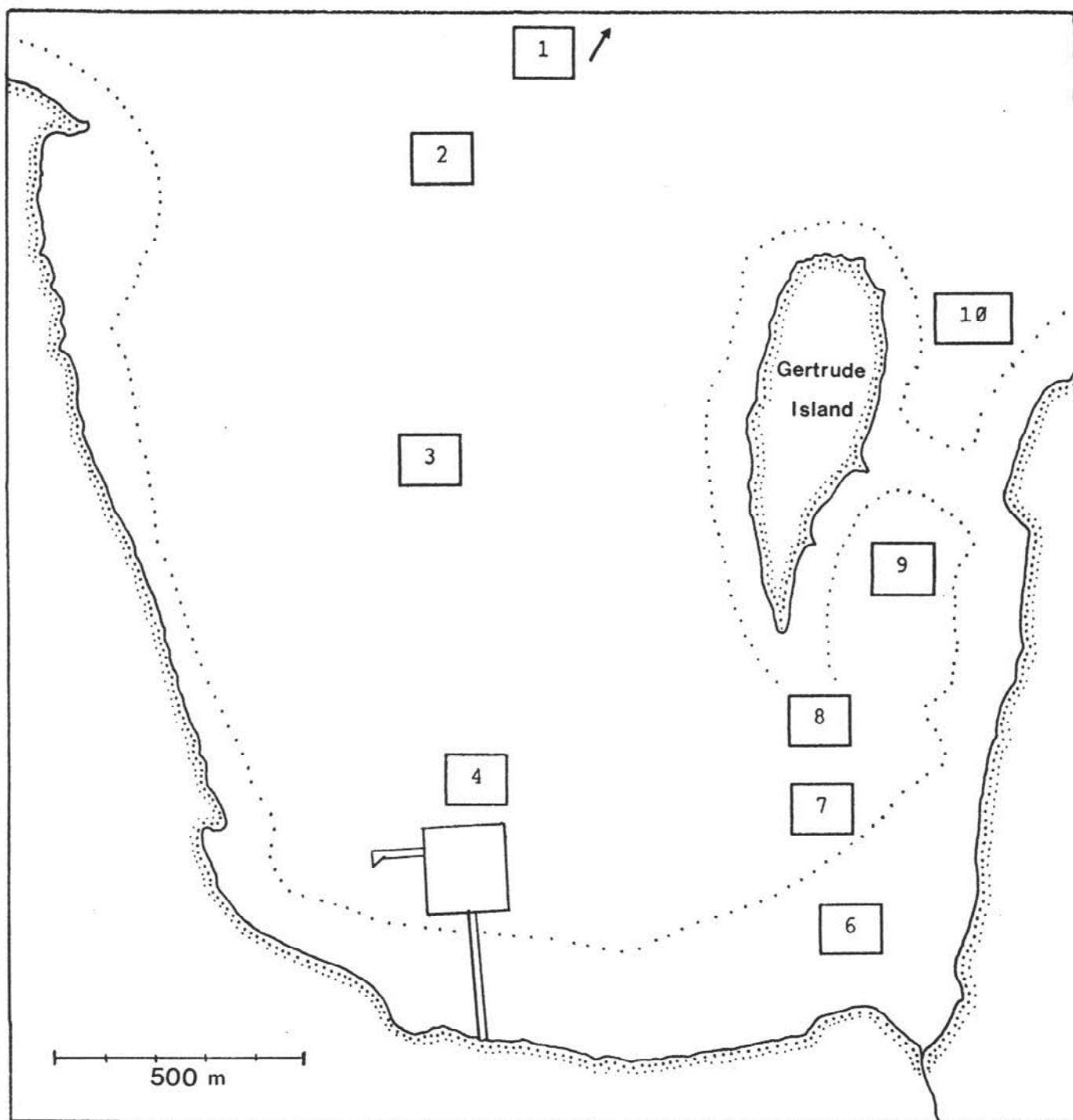


Figure 3. Water sampling stations in Still Harbor. Station 1 (control) was located off the map, one kilometer in the direction indicated.

of approximately 20 clams. Collections were made on three days at locations shown in Figure 4.

Captive experiment

We conducted supplemental research on bacterial densities of seals in a captive environment to provide an independent estimate of bacterial contamination by seals in a controlled environment and to compliment current efforts to estimate separate variables used to define bacterial contamination. The following specifications were used in conducting the captive study:

An enclosed tank containing two harbor seals and a known volume of water.

Seals in tank were not currently being fed antibiotics or other drugs known to alter their intestinal bacteria.

Seawater in the tanks was not chemically treated and was at a temperature similar to Puget Sound.

Water flow was shut off for a 46-hour period.

Sampling of water was conducted at 2 hour intervals throughout the 46-hour experimental period.

Fecal samples were collected from the seals in the tank both before and after the experiment.

Laboratory methods

Samples were analyzed by the Washington State Public Health Laboratory in Seattle, WA. Samples were kept cool until analysis within 24 hours of sample collection. Analyses of feces were conducted using approximately 4 g subsamples. Total coliforms and fecal coliforms were determined for all fecal, shellfish, and water samples using the Most Probable Number (MPN) technique. Decimal dilutions with marine and freshwater samples allowed detection limits of 1.8 to 2,400 organisms per 100 ml. Shellfish samples were generally tested to allow detection of 1.8 to 2,400 organisms per 100 grams, although several samples from an area where previous samples exceeded the upper limit were tested for higher concentrations. Feces were tested with a lower limit of detection of 2×10^2 to 2×10^4 and an upper limit of detection of 2×10^9 . A range of detection limits was used with samples of marine water from the dissolving experiments.

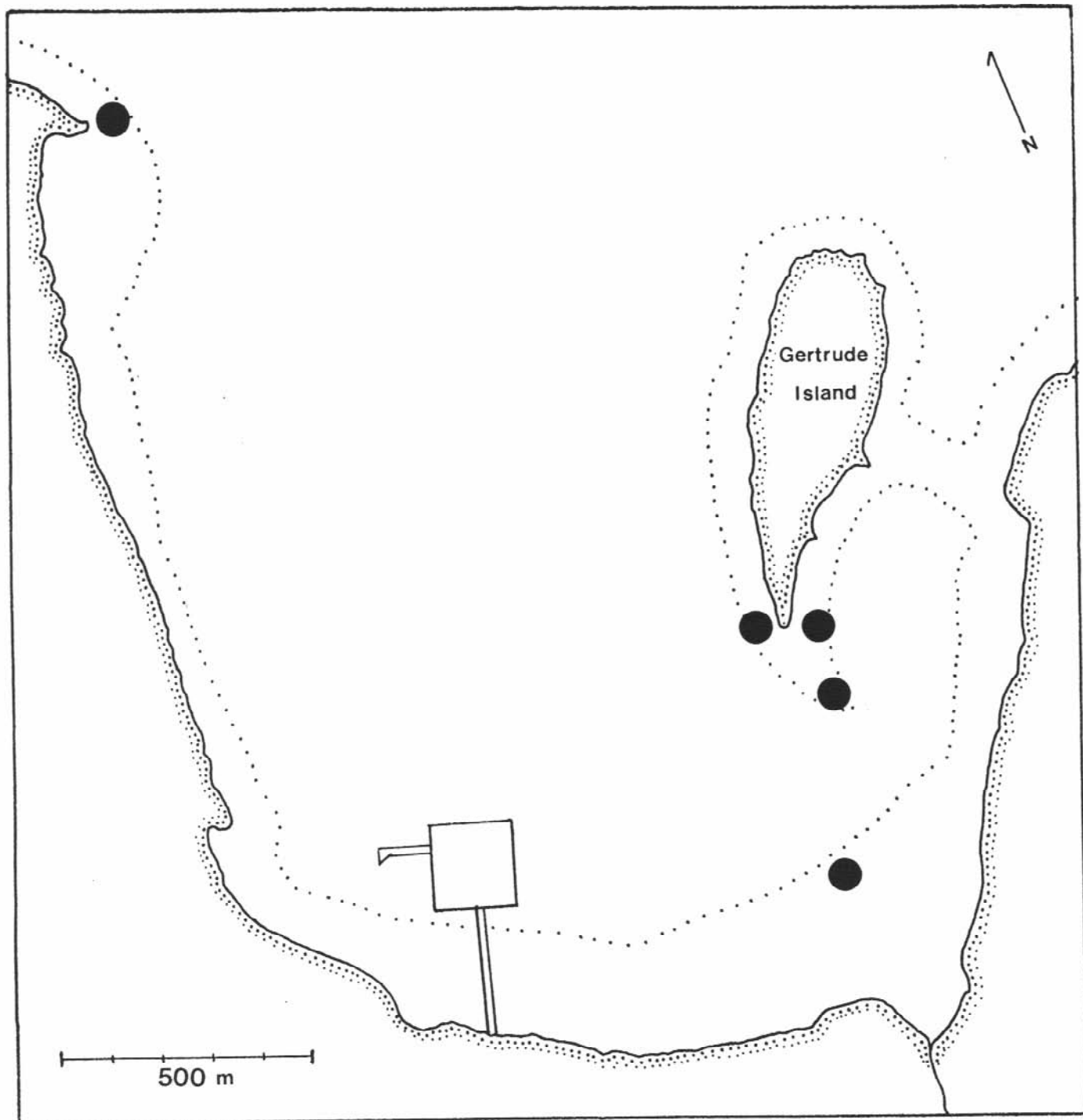


Figure 4. Location of little-neck clam sampling areas (indicated by dots).

Data and statistical analysis

Census data and fecal coliform concentrations were compiled and analyzed on a PC computer using DBASE III, LOTUS 123, SYSTAT, and SYGRAPH software programs. Census data were analyzed by step-wise linear regression, multiple linear regression, and analysis of variance (ANOVA) using SYSTAT (Wilkinson 1988). Factors evaluated for influence on seal numbers at a site included: year, month, time of day, time of high tide, height of high tide, duration of observation, visibility, precipitation, temperature, sky cover, and wind speed.

Fecal and total coliform concentrations were log-transformed before statistical analyses. Geometric means were calculated (from the log value). A standard deviation interval was calculated by adding or subtracting the standard deviation from the mean of the log values before retransforming back to non-log values. Levels of fecal coliform in water that fell below detection limits were assigned the lower detection limit value for analysis. The only exceptions were for fecal samples where some additional analysis was conducted excluding these values; these are noted in the text and tables.

RESULTS AND DISCUSSION

Seal populations

Numbers of harbor seals at sites in the northern Hood Canal are summarized in Table 3. Dosewallips River Delta tended to have the largest number of harbor seals during 1988, an average of 178 seals were seen during each visit. Duckabush River Delta and Quilcene Bay had averages of 139 and 91 harbor seals, respectively, per visit in 1988. Numbers of harbor seals at each site were highly variable and ranged from 0 to over 300 seals at each site.

Seal numbers counted at Gertrude Island during days water and shellfish sampling were conducted are shown in Table 4. On three of four days a maximum of 375 to 483 harbor seals were counted. On the fourth day, no seals were hauled out when we arrived and only 70 were counted in the water. Tracks and fresh fecal remains on the beach, however, indicated a larger number of seals likely had been hauled out before our arrival. Peak counts of seals at Gertrude Island in 1984 averaged 324 ($n=59$, $s.d.=116$) with a maximum of 483 counted (Calambokidis et al. 1985). Beginning in 1985, maximum numbers of over 500 were observed. The numbers of seals counted during our sampling trips were consistent with these previous counts and the general increasing number of seals at this site.

Seal numbers varied seasonally at the three major haul-out sites in northern Hood Canal (Figure 5). At all three sites average counts of seals varied significantly by month (ANOVA, $p<0.001$, all three cases); although the seasonal pattern was slightly different at each site. At Quilcene the highest average counts were made from September to December with the highest single count of seals made in December. Lowest seal numbers were generally from February through April. Seals at Dosewallips had a similar trend with generally highest numbers from September through January and lowest March through July. Seasonal data from Duckabush River Delta are not as complete but indicate a much more dramatic variation in seal numbers by season. More seals were generally seen from July to October with a dramatic decline in numbers by November. The decline in seal numbers in the fall at Duckabush corresponds to an increase in seal numbers at Quilcene and Dosewallips and suggests a shift in the distribution of seals in northern Hood Canal.

The average number of harbor seals seen on any day was generally between 25 and 50% of the maximum number seen at that site. This reflects seasonal shifts in use of some sites and the proportion of seals that do not haul-out on any given day. The proportion of days that individual

Table 3. Census results at sites in northern Hood Canal. Number, mean, and standard deviation are shown for the highest count of seals made during each visit to the site.

Site	1988				1984-88			
	No.	Mean	S.D.	High	No.	Mean	S.D.	High
Quilcene	32	91	95	403	97	94	79	403
Dosewallips	23	178	99	372	131	188	116	484
Duckabush	24	139	91	322	97	125	91	322

Table 4. Censuses of harbor seals at Gertrude Island in Still Harbor during days that water and shellfish were collected.

Date	Time			Number of seals		Comments
	Start	End	Peak	Low	High	
8 May 1988	1400	1830	1530	50	451	Disturbance at 1533
22 May 1988	1415	2000	1500	50	375	Disturbance at 1635
10 Jul 1988	0720	1330	0720	40	70	Seals had already entered the water
2 Aug 1988	1300	1830	1400	30	483	Disturbance at 1515

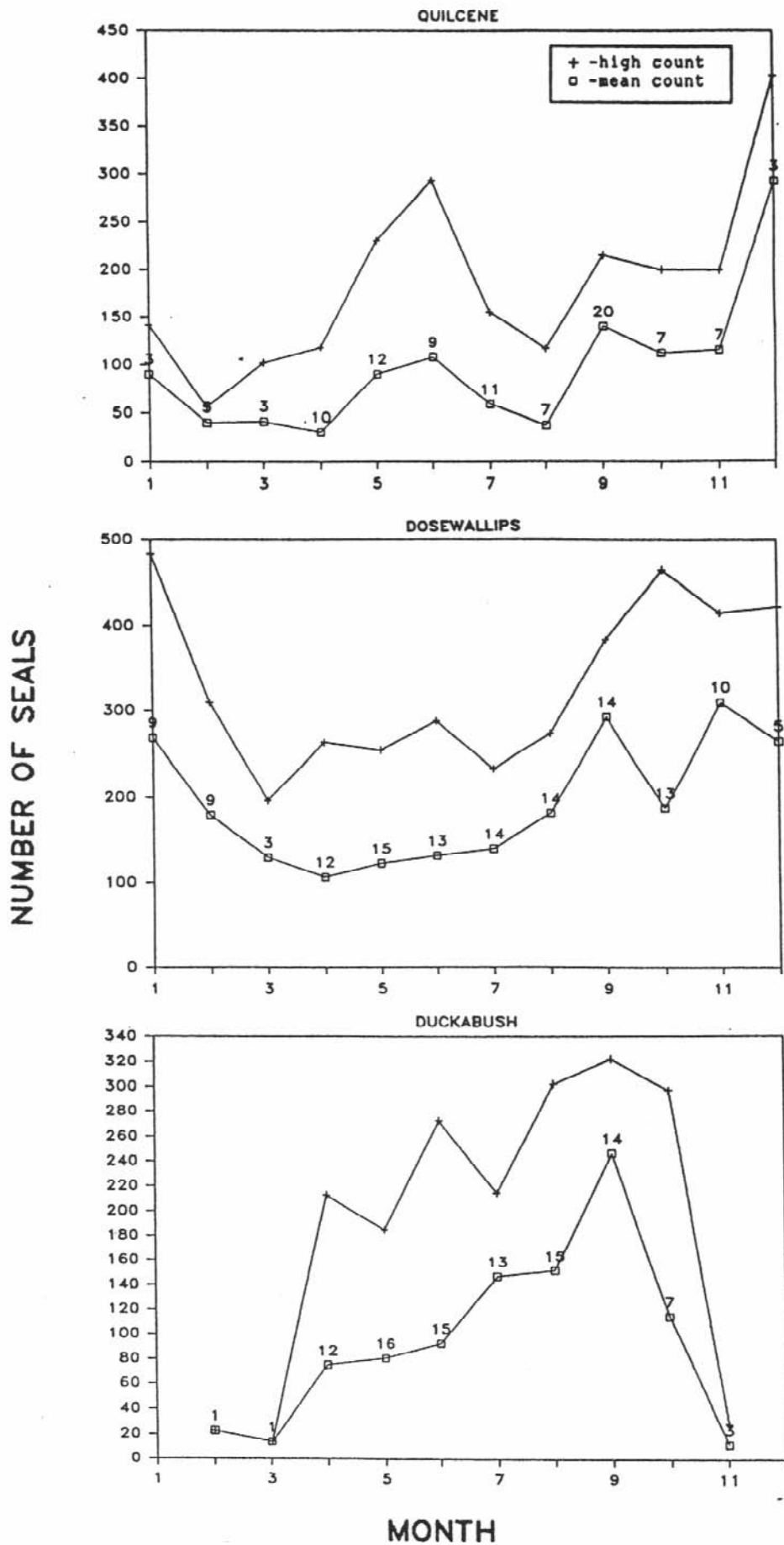


Figure 5. Highest counts and average number of harbor seals in Quilcene Bay and at Dosewallips and Duckabush River Deltas by month. Number of days counts were made are shown.

seals haul out has been reported as 41-65% (Stewart and Yochem 1983), 41-50% (Pitcher and McAllister 1981), and 44% (Sullivan 1979).

Evaluation of the annual change in seal numbers is complicated by seasonal and other factors that also are responsible for variations in seal numbers seen at haul-out sites. Between 1977 and 1984 harbor seal numbers increased 8% per year at Dosewallips Delta and 7% per year at Duckabush Delta (Calambokidis et al. 1985). Differences among years from 1984 to 1988, without considering other factors, were only significant at Dosewallips (ANOVA, $p < 0.001$). Average and maximum counts by year for 1984 to 1988 at all three sites in northern Hood Canal are shown in Figure 6. The most dramatic increase in maximal numbers of seals was seen in Quilcene Bay, where over 400 seals were counted for the first time in December of 1988. This was dramatically higher than any previous counts and may have represented animals entering this area in response to the presence of spawning squid. The recovery of squid remains is discussed in the food habits section.

Results of two aerial surveys provided minimum estimates of the number of seals in the entire Hood Canal (Table 5). These are the highest counts for the entire region made to date and indicated a minimum of 1,419 seals in the Hood Canal from Quilcene south. Four aerial surveys of the entire Hood Canal in summer and fall 1984 all yielded less than 1,000 seals, far lower than the 1988 counts.

Step-wise and multiple linear regressions allowed simultaneous and more reliable determination of the factors (including annual changes) that influenced seal numbers at a site. These models, however, tested for the significance of changes that followed a linear pattern. The following factors were found to be significant at one or more sites and were included in the multiple linear regressions for each site as noted:

Year - Annual changes were included in the model for both Quilcene and Dosewallips. Yearly changes made a contribution to the model at Quilcene that fell short of statistical significance ($p = 0.06$), whereas yearly changes were highly significant at Dosewallips ($p < 0.001$). The slope of the annual component of the regressions predicted annual increases of 20 seals at Dosewallips and 11 seals at Quilcene. This corresponds to annual increases of just over 10% of average numbers for these two sites and is consistent with the 7-8% increase reported by Calambokidis et al. (1985) for 1977 to 1984. No significant yearly differences were seen at Duckabush.

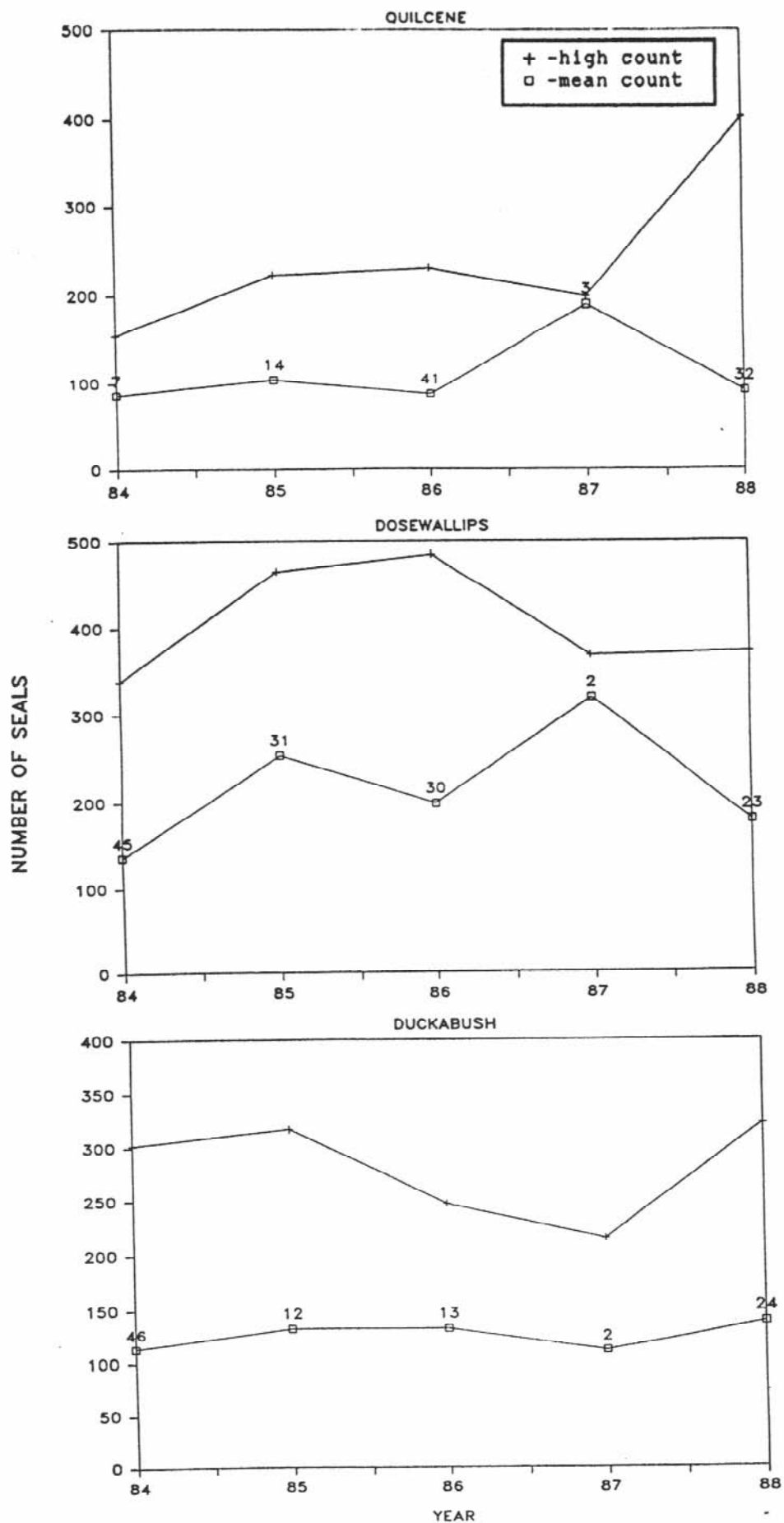


Figure 6. Changes in harbor seal numbers in Quilcene Bay and at Dosewallips and Duckabush River Deltas between 1984 and 1988. Number of days counts are made are shown. Trends are not adjusted for the number or seasonal timing of counts. Significant yearly increases were found at Dosewallips, for example, when these factors are considered (see text).

Table 5. Numbers of seals seen during aerial surveys of Hood Canal.

Site	13 Sept. 1988			14 Sept. 1988		
	hailed	water	total	hailed	water	total
Skokomish	186	10	196	330	4	334
Hamma Hamma	213	15	228	232	-	232
Duckabush	312	-	312	322	-	322
Dosewallips	345	8	353	372	-	372
Quilcene - log boom	73	6	81	55	3	58
Quilcene - oyster rafts	105	7	112	85	5	90
Other	-	7	7	7	4	11
TOTAL	1,234	53	1,287	1,403	16	1,419

Month - Monthly changes were significant at all three sites (simple linear regression and ANOVA). For the multiple linear regression model, the chronological order of calendar months by time of year was retained but months were numbered through the year starting with the season with lowest counts (e.g. the first month was March and the last month February). This invalidates the significance of the monthly component of the multiple regression model but allowed more precise evaluation of the other non-seasonal factors influencing seal numbers.

Tide - Seal numbers were significantly associated with three factors related to tide. These included: 1) the height of the high tide at Duckabush ($p < 0.007$) and Dosewallips ($p = 0.002$), but not at Quilcene ($p > 0.05$), seal numbers were greatest when the tide was highest; 2) the time of the high tide at all three sites ($p < 0.001$ at Dosewallips and Duckabush and $p = 0.02$ at Quilcene), with early morning high tides having higher numbers than high tides later in the day; and 3) length of time from the maximum count to high tide at Dosewallips ($p < 0.001$), with the counts made closest to high tide being greatest. The relationship between high tide and seal numbers at sites in the Hood Canal has been previously documented; counts were made primarily at or near the time of high tide at all sites. The smaller effect of tide on seals at Quilcene is not surprising because, unlike Dosewallips and Duckabush Deltas where seals primarily haul out on marshes only accessible during high tides, seals at Quilcene haul out on logs and floats that are accessible at all but lower low tides.

Duration of counts - The number of seals counted was significantly related to the duration of counts at Dosewallips ($p < 0.001$). This would be expected because longer observation periods increase the probability of obtaining a higher count; it is somewhat surprising the same relationship was not also found at other sites.

Environmental factors - There was some evidence that temperature, wind speed, and sky cover all had some influence on seal numbers. Because each of these factors was not significant at more than one site and were intercorrelated with the seasonal differences in seal numbers, they were not included in the model.

Food habits

Prey of seals was different among the study sites (Table 6). Feces collected at Dosewallips River Delta had the least variety of prey remains. All nine feces with identifiable remains (otoliths or cephalopod beaks) contained remains of Pacific hake (also called Pacific whiting, Merluccius

Table 6. Summary of prey found in seal scats in 1988. Identifiable remains indicate presence of otoliths or cephalopod beaks.

Site	Number of feces										
	Exam ined	w/fish remains	Ident- tifiable	Hake	Tom- cod	Squid	Her- ring	Scul- pin	Perch	Mid- shipman	Other
Gertrude	19	17	8	4	4	0	0	0	1	2	Sole
Dosewal.	15	15	9	9	0	0	1	0	0	0	
Quilcene	23	23	22	18	0	14	3	2	4	1	Pollock

productus). The only other species identified was Pacific herring (Clupea pallasii) from a single sample. The almost exclusive consumption of hake, indicated by these recent samples, is consistent with previous findings at Dosewallips (Calambokidis et al. 1978, Calambokidis and McLaughlin 1987). Food habits of harbor seals in the Puget Sound region has been shown to vary by location reflecting the opportunistic nature of harbor seal prey selection (Calambokidis et al. 1978, Everitt et al. 1981).

Prey eaten by harbor seals at Quilcene Bay and Gertrude Island were more variable than at Dosewallips. Hake was again one of the most frequently recovered prey items at these sites, however, many other species were found consistently. At Quilcene, 14 of 23 scats contained remains of opalescent squid (Loligo opalescens), with one scat containing 18 beaks. In addition to squid, six species of fish were also identified in remains from Quilcene. Because of the difficulty in collecting feces from Quilcene in previous years, these are the first food habits results from this site.

The high number of seals in Quilcene Bay in December may be in response to the availability of squid. Squid have not been recovered from feces of Puget Sound harbor seals (Calambokidis et al. 1978, Calambokidis and McLaughlin 1987). Everitt et al. (1981) reported recovery of just four squid beaks from 129 seal scats collected at Protection Island in the Strait of Juan de Fuca. The recovery of squid beaks in seal feces in Quilcene Bay coincided with reports of squid aggregations in Quilcene Bay. Opalescent squid congregate on spawning grounds in sheltered bays with sandy or muddy bottoms less than 20 fathoms deep (Macfarlane and Yamamoto 1974), making Quilcene a prime candidate. Post-spawning squid reportedly die or are eaten by predators. The feces were collected in December during a time period when we found the highest numbers of seals in the bay (>400). Scats from Dosewallips in January showed no signs of squid.

Hake are the dominant food of harbor seals in Hood Canal, but relatively little is known about hake in this area. Hake form the basis of a commercial fishery in Saratoga Passage and Port Susan (Pederson 1985). The Puget Sound stock are genetically distinct from the hake occurring in the eastern Pacific. Hake are the dominant prey item of harbor seals throughout the Hood Canal (Calambokidis et al. 1978). Fishermen in Quilcene Bay were not aware of hake occurring inside the bay and most predation on hake likely occurs in deeper waters of Hood Canal.

Fecal coliform densities

Fecal coliform densities found in seal feces from various sites in 1988 is summarized in Table 7 and in Figure 7 along with results for

Table 7. Fecal coliform densities in feces of harbor seals collected from various sites. Because five samples (two Dosewallips, two captive, and one Gertrude) had outlying values falling below detection limits (2×10^2 to 2×10^4), results are summarized with and without the detection limit values.

Site	Year	N	Arith. mean	Std. dev.	Geom. mean	Std. dev. interval
<u>Including values below detection limits at the detection limit</u>						
Dosewal. ¹	1986	10	1.7×10^8	3.0×10^8	3.1×10^7	$4.9 \times 10^6 - 1.9 \times 10^8$
Dosewal.	1988	21	2.0×10^8	2.3×10^8	6.4×10^7	$6.3 \times 10^6 - 6.6 \times 10^8$
Quilcene	1988	7	1.3×10^7	1.2×10^7	5.8×10^6	$1.3 \times 10^6 - 2.4 \times 10^7$
Gertrude	1988	17	6.3×10^7	9.5×10^7	6.2×10^6	$1.4 \times 10^5 - 2.6 \times 10^8$
Captive	1988	9	5.4×10^4	1.0×10^5	5.6×10^3	$4.3 \times 10^2 - 7.3 \times 10^4$
<u>Using only values above detection limits</u>						
Dosewal. ¹	1986	10	1.7×10^8	3.0×10^8	3.1×10^7	$4.9 \times 10^6 - 1.9 \times 10^8$
Dosewal.	1988	20	2.1×10^8	2.4×10^8	9.6×10^7	$2.1 \times 10^7 - 4.3 \times 10^8$
Quilcene	1988	7	1.3×10^7	1.2×10^7	5.8×10^6	$1.3 \times 10^6 - 2.4 \times 10^7$
Gertrude	1988	15	7.2×10^7	9.8×10^7	1.8×10^7	$1.5 \times 10^6 - 2.1 \times 10^8$
Captive	1988	7	6.9×10^4	1.1×10^5	1.4×10^4	$1.8 \times 10^3 - 1.2 \times 10^5$

¹ from Calambokidis and McLaughlin (1987)

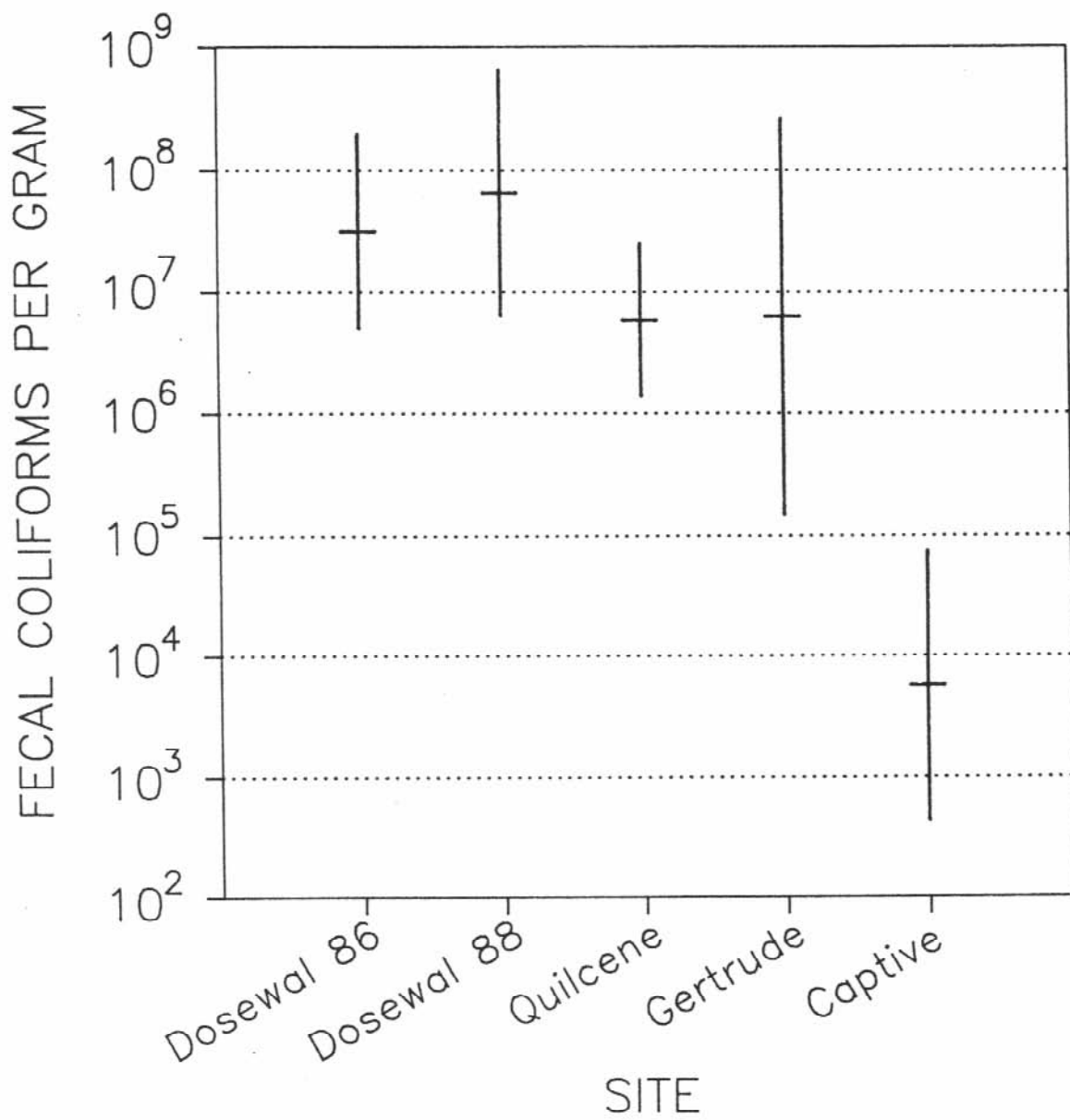


Figure 7. Geometric mean fecal coliform densities in seal feces from different sites. Bars show 95% confidence intervals.

Dosewallips Delta in 1986 previously reported by Calambokidis and McLaughlin (1987). Samples from Dosewallips from 1986 and 1988 were not significantly different (t-test, $p > 0.05$) although the analyses were conducted 2 years apart by different laboratories. For statistical analyses both sets of data for Dosewallips were pooled.

The distribution of the sample results showed some unusual patterns. Three samples from wild harbor seals had values that fell below detection limits (2×10^3 to 2×10^4) as did two samples from captive animals (analyzed with a detection limit of 2×10^2). Because the detection limits fell far below the typical fecal coliform densities found in feces, these values represented outlying values sharply separated from the majority of the samples. There was nothing unusual in how these samples were collected, condition of the samples, or prey remains recovered compared to other samples. For statistical analyses, duplicate runs were conducted; 1) including these samples with their value set at the detection limit, and 2) excluding these outlying values from the analysis.

Differences by site

There were significant differences in fecal coliform densities among sites (ANOVA, $p < 0.001$). Most of these differences were the result of dramatically lower fecal coliform concentrations in the 9 feces from captive seals. Densities in feces from captive animals were all less than 3.3×10^5 per gram; of the 55 feces from wild animals examined only 4 samples were equivalent or lower than this value.

The reason why captive seals had such dramatically lower bacterial densities is not clear. Samples from captive animals were collected from several individual seals on five different days over a 6 month period and therefore were not the result of short-term factors. There was some evidence of an increase in concentrations over the 6 month sample period, though the sample size was too small to test this trend. The captive seals were not currently being given antibiotics or other medication known to kill bacteria. The diet of the captive seals was herring and squid that had been frozen.

Though there were differences in how captive and wild seal feces were sampled, this factor does not appear to be responsible for the differences between these populations. Fecal samples from captive animals were taken from the enclosure tank that was filled with marine water, unlike the sampling of wild seals where samples were collected off the substrate of the haul-out area. These differences resulted in captive samples potentially soaking in the marine water for a prolonged period prior to

collection. This factor does not appear to be responsible for the lower concentrations, however, because soaking in marine water did not dramatically alter densities in three experiments we conducted (see later in this section). The lower fecal coliform densities in captive seals also was apparent in results of an experiment examining fecal coliform densities in the water of captive seals discussed later in this report.

In addition to the values found for captive seals, differences also existed among the sites where wild seals were sampled. Fecal coliform densities were significantly different among the three sites sampled (ANOVA, $p < 0.05$ including values below detection limits; $p < 0.01$ excluding values below detection limits). Samples from Dosewallips River Delta had the highest fecal coliform densities with a geometric mean more than five times higher than at Gertrude Island or Quilcene Bay. Fecal coliform densities in samples from Quilcene Bay and Gertrude Island were similar.

Differences in prey among sites does not appear to be responsible for the differences in bacterial densities. Harbor seals are opportunistic feeders and differences in prey of harbor seals among sites in Puget Sound has been noted (Calambokidis et al. 1978). Similarly, examination of fecal samples during this study revealed differences in prey consumption among sites we sampled. We found no association, however, between prey species and fecal coliform concentrations in those samples where we had both bacterial and food habits information.

Large variations in the intestinal microflora of other animals also have been noted (Geldreich 1976, Mara and Oragui 1981). Identifying the sources of these variations, even in humans, has proven difficult. Gorbach et al. (1967) examined the effect of diet and age on intestinal bacteria and reviewed other factors studied. Although diet and age both have been reported to affect intestinal bacterial densities, they found no significant effects. Individuals, however, tended to have remarkably consistent bacterial densities over time.

Sampling factors altering fecal coliform densities in feces

There were some differences in sampling strategies that potentially could have affected the observed densities in our samples. Feces were collected at Dosewallips from the marsh haul-out areas used by seals at high tide. Feces from Quilcene were collected from rafts used by large numbers of seals immediately before collection, whereas at Gertrude feces were collected from an intertidal area used by harbor seals at low tide. At all sites only feces that were judged to be recently deposited (<12 hours) were sampled. This was a subjective determination, however, and

there was a greater potential for erroneously sampling older feces at Dosewallips.

Changes in fecal coliform counts between time of defecation and collection was examined by repeated sampling of two feces left at the haul-out area in Quilcene Bay (Figure 8). The samples, both having identical initial densities, increased to the same value after one day, and then by the second day, decreased more dramatically to levels slightly lower than initial values. The results suggest that the time between defecation and sampling could alter fecal coliform densities. Despite the consistency between the two samples, these results should be interpreted with caution because of the small sample size. The differences in bacterial densities observed are smaller than those noted between the different locations where feces were collected.

A similar increase in fecal coliforms with time has been noted in cattle feces by Thelin and Gifford (1983). They found that releases of fecal coliforms from cattle feces during a simulated rain were highest two days after defecation. Only after 5 days did fecal coliform counts decline below levels for fresh feces. They concluded that fecal coliform continue to grow and multiply for several days after defecation. Feces more than 2 days old showed a logarithmic decline in the release of fecal coliforms (Thelin and Gifford 1983, Kress and Gifford 1984).

Immersion of two seal feces in marine water for a 36 hour period did not dramatically alter fecal coliform densities, considering typical variability inherent with fecal coliform concentrations. Of the two feces from wild seals immersed for 36 hours, one showed no change and the other a slight decline (Figure 8). A third feces from a captive animal showed a similar small decline from 1.7×10^4 to 7.8×10^3 per gram, after immersion for 48 hours.

The viability of fecal coliforms after excretion has been the subject of considerable research. A wide variety of factors influence fecal coliform die-off or regrowth in soils, freshwater, and marine waters. Examples of some of the factors identified include:

Factor	Medium	Reference
Sunlight and salinity	Fresh and sea water	Chojnowski et al. 1979
Marine microflora conc.	Sea water	Mitchell and Morris 1969
Season	Soil	Chandler et al. 1981
Season, moisture, and Ph	Soil	Van Donsel et al. 1967
Salinity, sunlight	Fresh and sea water	Determan et al. 1985

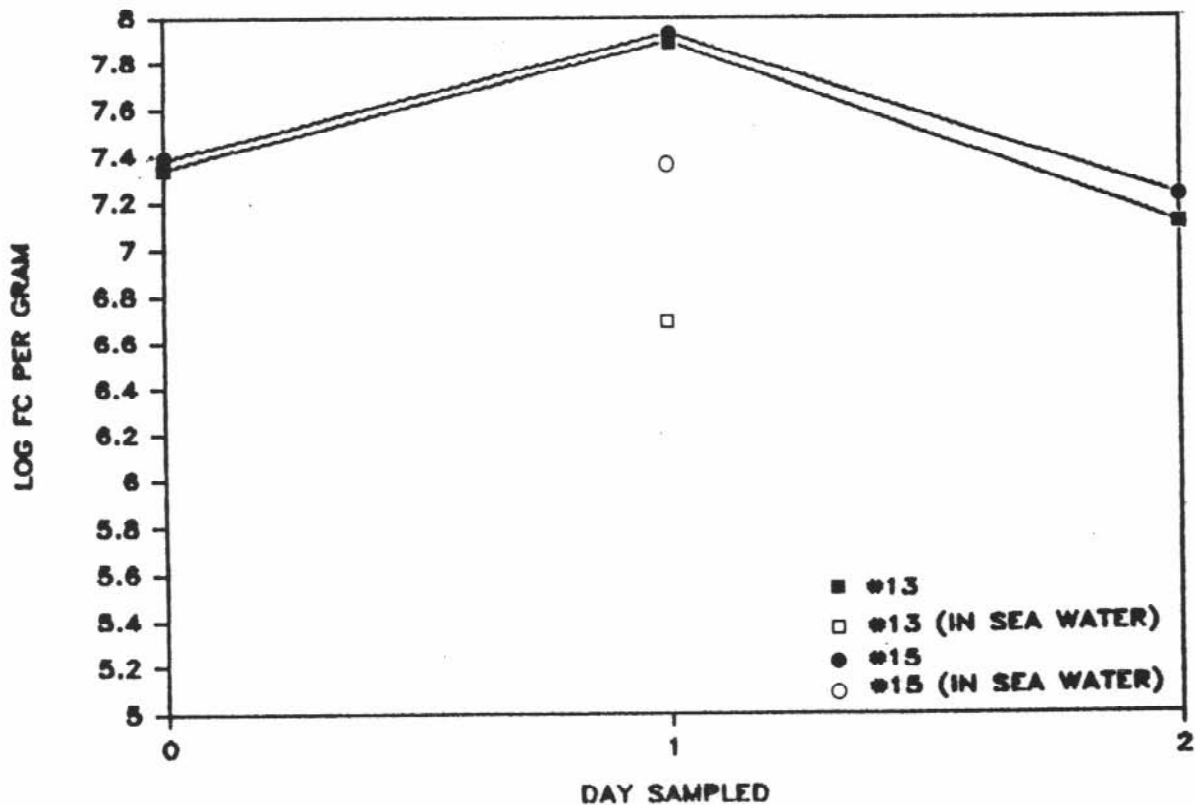


Figure 8. The log values of fecal coliform densities in two feces from harbor seals that were left at the haul-out area and sub-sampled after 24 and 48 hours. Densities of these feces were also examined when submerged in sea water for 36 hours and are shown as open symbols.

Comparison to other species

The densities of fecal coliform we found are high compared to most other species (Table 8). Compared with median values for humans and a variety of domestic and wild animals summarized by Geldreich (1976), fecal coliform levels in scats from Dosewallips harbor seals were the highest for any species, but wild seals at other sites were in the same range as humans and a few other domestic and wild animals. Compared to geometric mean values reported by Mara and Oragui (1981), harbor seal concentrations were still relatively high, but several species including humans, dogs, cats, and turkeys had higher mean concentrations than seals at any of our study sites.

Defecation rates in conjunction with fecal coliform densities allow comparison of fecal coliforms produced per individual of different species (Table 9). The per animal fecal coliform production of harbor seals was higher than humans and domestic animals using the Dosewallips fecal coliform concentrations. Using the lower values we found for seals at Gertrude Island, seal fecal coliform production per animal was lower than cows or sheep but similar to humans.

Dissolving of feces in marine water

Experiments mixing feces in marine water provided information on how readily seal feces dissolve into the water column and whether they remain in suspension. Results of these experiments are summarized in Table 10 and indicated that the behavior of feces in marine water was highly variable, with some samples readily dissolving into the water column and others not dissolving at all. From 0.0% to >32% of the fecal coliforms dissolved in the water column after 30 seconds of gentle agitation. Allowing samples to settle for 1 hour did not result in consistent changes in the proportion of the feces dissolving in water. A second gentle agitation of the water after this one hour period, however, did result in consistently higher concentrations in the water column. These final concentrations ranged from 0.0% to 55% of the fecal coliforms suspended in the water column.

The physical consistency of the feces was responsible for how readily they dissolved in marine water. One sample that was of a solid consistency (GI-28) and a second sample that was extremely oily (DO-21) both showed very low dissolving rates. Conversely, the three feces showing the highest dissolving rates (DO-18, DO-19, and GI-27) were all runny with few solid pieces. Seals usually defecate in water rather than on land, where we collected the feces used in these experiments. The feces we used, although recently deposited, may have dried out slightly thereby reducing how easily

Table 8. Comparison of fecal coliform concentrations per gram of feces in humans and other animal species.

Fecal source	# samples	Median	# samples	Geometric mean	Range	
					Minimum	Maximum
Reported:	Geldreich (1976)		Mara and Oragui (1981)			
Human	43	1.3×10^7	18	6.3×10^7	1.3×10^5	9.0×10^9
Cattle	11	2.3×10^5	8	1.0×10^6	1.5×10^5	6.5×10^6
Sheep	10	1.6×10^7	7	2.7×10^6	1.8×10^5	5.6×10^7
Pig	11	3.3×10^6	8	3.9×10^7	4.9×10^6	6.0×10^8
Horse	-	1.3×10^4	5	1.0×10^3	6.3×10^2	3.4×10^3
Duck	8	3.3×10^7	5	2.0×10^7	8.8×10^6	4.9×10^7
Chicken	10	1.3×10^6	9	6.3×10^6	3.7×10^6	1.5×10^7
Turkey	10	2.9×10^5	6	7.9×10^8	2.6×10^8	2.0×10^9
Goose	-	-	3	6.3×10^3	9.7×10^2	6.6×10^4
Cat	19	7.9×10^6	5	6.3×10^7	8.9×10^4	2.6×10^9
Dog	24	2.3×10^7	5	5.0×10^8	4.1×10^6	4.3×10^9
Rabbit	14	2.0×10^1	4	1.0×10^4	2.8×10^3	4.9×10^4
Mouse	7	3.3×10^5	4	6.3×10^6	4.7×10^6	1.0×10^7
Rat	2	1.8×10^5	4	1.6×10^5	5.6×10^4	6.3×10^5
Gull	-	-	6	6.3×10^3	1.7×10^2	2.7×10^5
Chipmunk	3	1.5×10^5	-	-	-	-
Elk	32	5.1×10^3	-	-	-	-
Robin	-	2.5×10^4	-	-	-	-
English sparrow	-	2.5×10^4	-	-	-	-
Starling	-	1.0×10^4	-	-	-	-
Red-winged blackbird	-	9.0×10^3	-	-	-	-
Pigeon	-	1.0×10^4	-	-	-	-
Harbor seals (this study)						
Dosewallips	31	7.3×10^7	31	5.1×10^7	$<2.0 \times 10^4$	9.2×10^8
Quilcene	7	4.9×10^6	7	5.8×10^6	4.9×10^5	3.3×10^7
Gertrude	17	2.3×10^7	17	6.2×10^6	$<2.0 \times 10^3$	3.5×10^8
Captive	9	7.9×10^3	9	5.6×10^3	$<2.0 \times 10^2$	3.3×10^5

Table 9. Daily fecal coliform (FC) production for different animals calculated per individual. Median fecal coliform densities were used for consistency with values reported for humans and domestic animals (Geldreich 1978). Feces production is for an adult, hence seal fecal production rate is higher than for an 'average' seal, as used in loading calculations.

Animal	Feces production ^a (g per day)	Median fecal coliform density (10 ⁶ FC per g)	Fecal coliform production rate (10 ⁹ FC per day)
Human	150 ^c	13. ^c	2.0
Cow	23,000 ^a	0.23 ^c	5.3
Horse	16,000 ^a	0.013 ^c	0.20
Sheep	900 ^a	16. ^c	14.
Pig	3,200 ^a	3.3 ^c	1.1
Dog	340 ^b	23. ^c	0.78
Harbor seal (adult)			
Dosewallips	750	73.	55.
Gertrude Is.	750	2.3	1.7

^a Calculated using fecal production rates and upper weight classes given in the Livestock Waste Facilities Manual (1979).

^b From Taylor (1984), source reported as Food and Drug Administration.

^c Geldreich (1978)

Table 10. Results of dissolving feces in 12 liters of marine water. Values are given for samples immediately after gentle agitation for 30 seconds, after one hour of settling, and immediately after agitation a second time. The large variations in how well feces dissolved in seawater resulted in some of the fecal coliform values falling above or below the range that could be quantified. These values are expressed as minimums or maximums as appropriate.

Sam. num.	Wt. added (grams)	FC in scat per gram	Fecal coliforms per 100 ml			Percent dissolving		
			After mixing	After settling	After 2nd mixing	After mixing	After settling	After 2nd mixing
DO-18	13.2	2.2×10^8	9.2×10^5	1.6×10^6	$>2.4 \times 10^6$	3.8%	6.6%	$>9.9\%$
DO-19	12.8	7.0×10^7	$>2.4 \times 10^6$	$>2.4 \times 10^6$	$>2.4 \times 10^6$	$>32.1\%$	$>32.1\%$	$>32.1\%$
DO-20	12.0	2.2×10^8	7.9×10^4	4.9×10^4	1.7×10^5	0.4%	0.2%	0.8%
DO-21	10.5	5.4×10^8	2.0×10^0	2.4×10^2	2.2×10^1	.0%	.0%	.0%
GI-22	18.0	3.3×10^7	$<2.4 \times 10^5$	$<2.4 \times 10^5$	$<2.4 \times 10^5$	$<4.8\%$	$<4.8\%$	$<4.8\%$
GI-27	10.0	3.5×10^8	$>2.4 \times 10^6$	2.4×10^6	1.6×10^7	$>8.2\%$	8.2%	54.9%
GI-28	18.0	1.3×10^8	7.6×10^4	7.9×10^4	3.5×10^5	0.4%	0.4%	1.8%
QU-12	6.7	1.7×10^6	$>2.4 \times 10^3$	$>2.4 \times 10^3$	$>2.4 \times 10^3$	$>2.5\%$	$>2.5\%$	$>2.5\%$
QU-14	14.5	4.9×10^5	$>2.4 \times 10^3$	$>2.4 \times 10^3$	$>2.4 \times 10^3$	$>4.1\%$	$>4.1\%$	$>4.1\%$

they dissolved in the water column. The dissolving rates we observed may therefore underestimate what occurs when animals defecate in water. In most cases the undissolved portion of feces settled slowly to the bottom but were only slightly negatively buoyant.

From these experiments we anticipate that fecal coliforms associated with seal feces would both dissolve in the water column as well as accumulate in the sediments where animals tend to defecate. Observations of captive animals and the fairly limited recovery of feces at haul-out areas indicate that seals more commonly defecate just after entering the water at the haul-out area and not on land during haul out. Seal fecal contamination, in the water column and in the sediment, would be expected to be highest surrounding the haul-out site.

Captive experiment

Monitoring of bacterial contamination in a tank with two captive seals provided information on bacterial contamination and dynamics in a 'closed' environment. Concentrations of fecal coliforms increased significantly over the 46 hour sampling period ($n=27$, $r=.55$, $p=0.003$). Fecal coliform levels in the tank and the fitted regression line are given in Figure 9. Concentrations were somewhat variable but there was also some clear fluctuations in addition to the general increase throughout the sampling period (Figure 10). The primary peak in the data occurred after 30 hours when one of the seals defecated. We suspect the other peaks in the data are also a result of defecation before samples being taken.

Somewhat surprising is the decrease in contaminant concentrations after periods of elevated levels. These appear to be the result of bacteria die-off. The settling of fecal material was observed in the dissolving experiment, however, in those experiments the water was agitated only for a short period. In the seal tank, the animals were active throughout the experiment and their activities resulted in constant mixing and circulation in the tank. Fecal material would be expected to continue to dissolve into the water column rather than settle out under these conditions. The increasing water turbidity in the tank indicated that increasing amounts of fecal material were dissolving in the water. Die-off of bacteria is increased by exposure to salt-water and sunlight. Most of the studies of die-off have used seed solutions of bacteria; the relevance of these mortality figures in cases where most of the bacteria is still encased or attached to particulate fecal matter is not known. The enclosed tank also may have resulted in some other changes in water conditions that could have affected bacteria die-off.

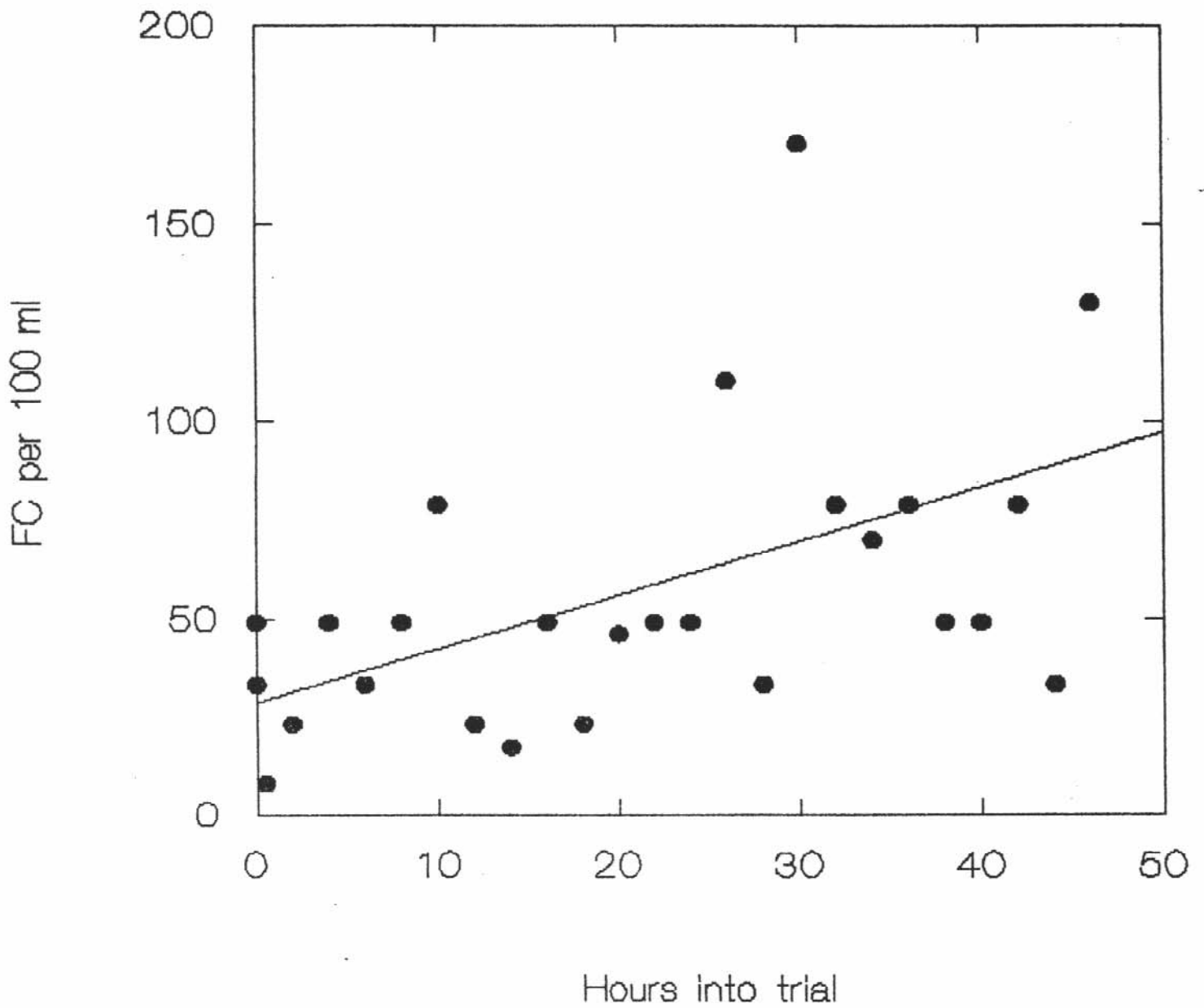


Figure 9. Fecal coliform levels in the captive seal tank over time. The fitted regression line is shown ($n=27$, $r=0.55$, $p=0.003$).

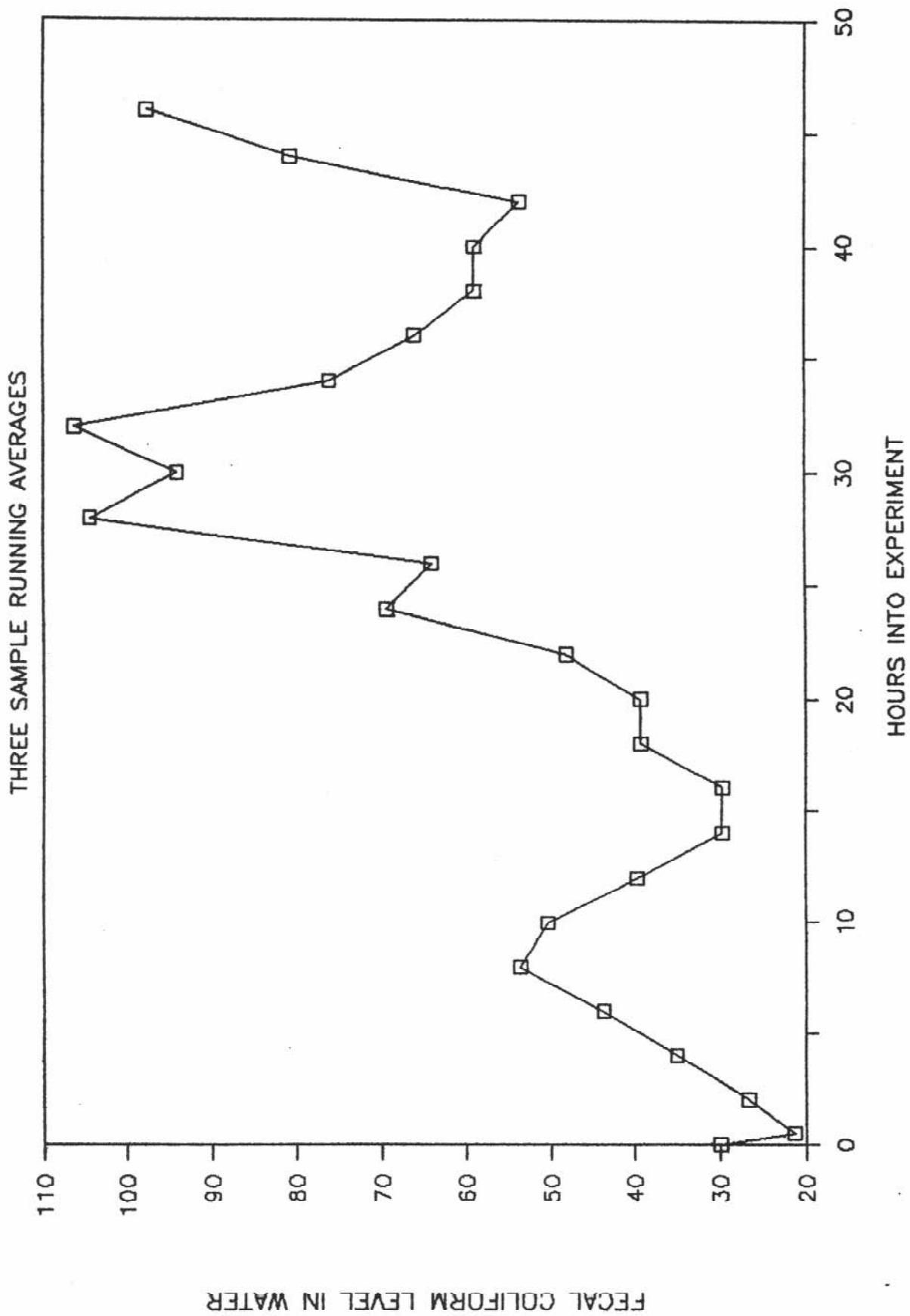


Figure 10. Fecal coliform densities in the captive seal tank over time, plotted as three-sample running averages to better show overall trends.

Fecal coliform concentrations in the tank were much lower than calculated from the estimated input (loading) from seals and the tank water volume. Loading calculations were based on number of seals, time period, defecation rate, and fecal coliform density. These calculations were conducted for the period when maximal concentrations were detected and for the end of the experiment; only 0.2% and 0.08%, respectively, of the estimated fecal coliform production was detected in the water column of the tank. Because the loading calculations did not adjust for feces not dissolving, observed fecal coliform densities would be expected to be lower than indicated from the loading estimates. Dissolving rates reported earlier in this report, however, were generally much greater than suggested by this experiment. These results indicate that bacterial loading calculations overestimate the concentration of bacteria occurring in the captive environment and may also overestimate contributions by seals in the wild.

There are a number of hypotheses for the low concentrations observed as compared to those calculated. Possible hypotheses are: 1) the high die-off of bacteria kept concentrations low, 2) only a very small proportion of the feces dissolved in the water column (lower than found in the dissolving experiments), and 3) fecal coliform densities in feces were overestimated. The latter was possible because only three samples collected immediately before or after the experiment were averaged for the loading calculation. A larger sample size from captive seals or inclusion of earlier samples from captive seals (which had much lower concentrations) could have produced more accurate results. Regardless of the cause of the difference between the concentrations observed in the tank and that predicted from loading calculations, this experiment demonstrates the potential inaccuracies inherent in comparing fecal coliform loadings from different sources.

Fecal coliform densities in water and shellfish in Still Harbor

Water and shellfish sampling in Still Harbor provided information on fecal coliform levels at a site where large numbers of harbor seals congregate in a small embayment. The control station, located in Carr Inlet outside of Still Harbor, had fecal coliform concentrations below detection limits during all samplings (<1.8 per 100 ml).

Fecal coliform concentrations in marine water from the nine sampling stations varied widely (Table 11 and Figure 11). Stations 2, 3 and 4 generally had fairly low concentrations of fecal coliforms with more than half of the samples from each station falling below detection limits. These stations all occupy the deeper and more open portion of Still Harbor.

Table 11. Concentrations of fecal coliforms per 100 ml of marine water taken from sampling stations in Still Harbor. Samples were taken twice daily; A - ebb to low tide, B - flood tide. Samples failing water quality criteria for shellfish production are marked.

Station	5/08		5/22		7/10		8/02		Geometric mean
	A	B	A	B	A	B	A	B	
1	<1.8	<1.8	<1.8	<1.8	<1.8	<1.8	<1.8	<1.8	1.8
2	<1.8	<1.8	<1.8	4.5	<1.8	4.5	<1.8	<1.8	2.3
3	<1.8	<1.8	<1.8	<1.8	2	<1.8	<1.8	<1.8	1.8
4	<1.8	4	2	2	<1.8	<1.8	<1.8	<1.8	2.0
6*	920	350	33	33	2	23	7.8	17	35
7*	79	11	7.8	<1.8	<1.8	33	23	49	13
8*	>2400	540	79	33	<1.8	540	540	49	131
9*	95	70	33	79	4.5	2	13	23	22
10*	17	46	6.8	<1.8	4.5	3.6	<1.8	<1.8	5.2

* Station fails water quality criteria for commercial shellfish production.

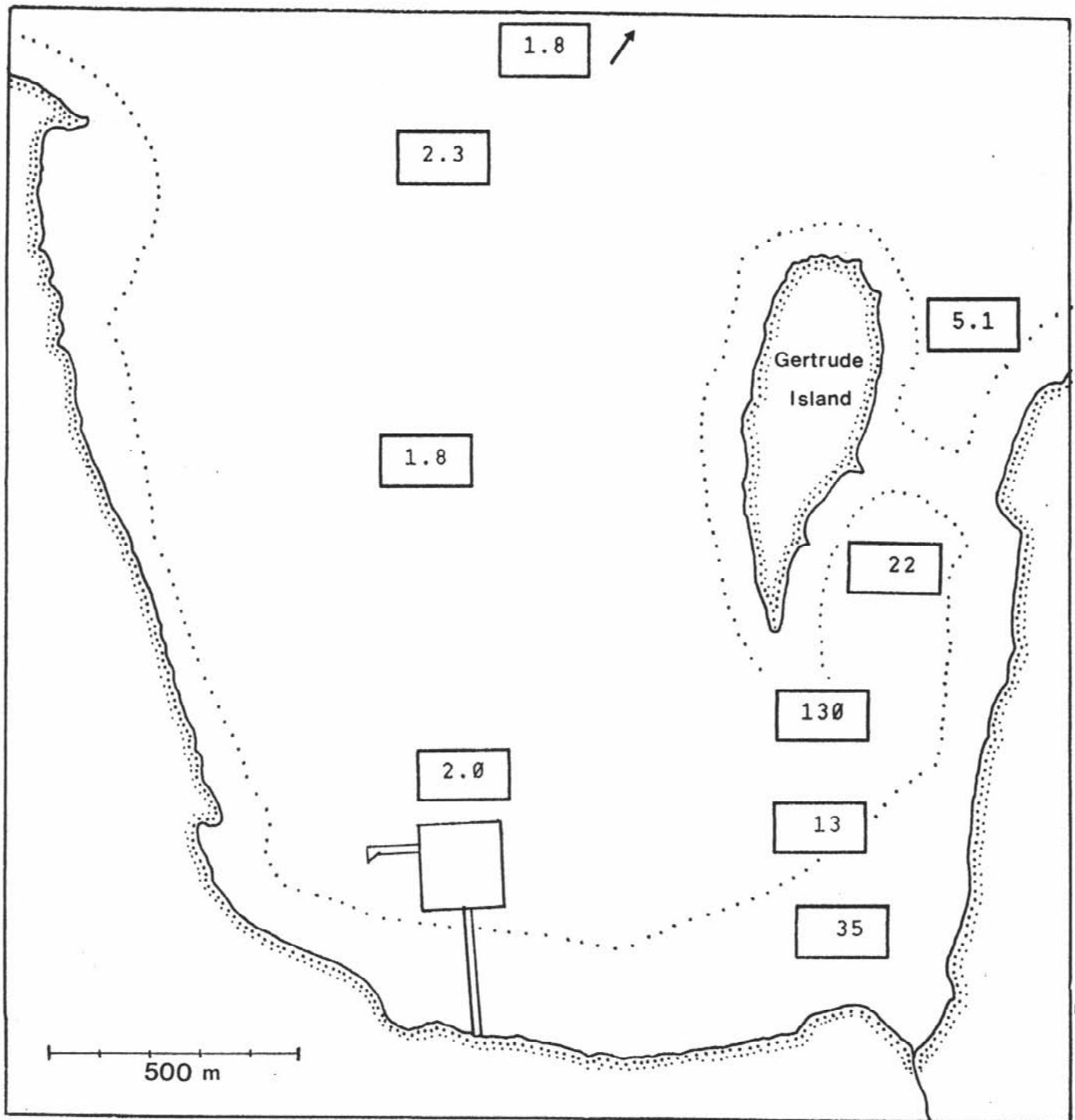


Figure 11. Geometric mean fecal coliform densities found in marine water samples in Still Harbor, $n=8$ for each station. All five sampling stations on the east side of the harbor fail one or both of the water quality standards for commercial shellfish production.

This portion of the Harbor is more open to the influx of water from Carr Inlet, and also is more removed from the harbor seal haul-out area on Gertrude Island in the east portion of Still Harbor. Concentrations were consistently higher at the stations in the east side of Still Harbor, near Gertrude Island. The primary source of bacterial contamination in Still Harbor appeared to be the haul-out area. The highest concentrations were consistently at the seal haul out area, where levels exceeded 100 per 100 ml during 5 of 8 samplings. The next highest concentrations in water were found at station number 6 at the southeast end of Still Harbor and about 100 meters from the haul-out area.

All five stations on the east side of the harbor failed one or both water quality standards for commercial shellfish production. Three exceeded the standard of mean concentrations of less than 14 per 100 ml. All five stations failed the standard of less than 10% of samples having less than 43 fecal coliforms per 100 ml.

There were differences in the concentrations of fecal coliforms among the four different days sampled. This was significant for the stations in the eastern portions of Still Harbor where concentrations were highest (ANOVA, $p < 0.01$, using log values by day). The day with the lowest concentrations of fecal coliform was the only day when large numbers of harbor seals were not present at Gertrude Island. Over 350 seals were hauled out at Gertrude island on three of the four days and only 70 were seen in the water on the fourth day.

Results of fecal coliform concentrations in shellfish followed a pattern similar to the water samples (Table 12). Three shellfish samples taken on different days from the haul-out area at the south end of Gertrude Island had fecal coliform concentrations of 49,000 to > 240,000 per 100 grams of tissue. Two samples taken a short distance away from the main haul-out area (occasionally used by seals) had slightly lower concentrations, 4,900 and 33,000 per 100 g. A site at the southeast end of the Harbor was sampled twice and had intermediate concentrations, 170 and 330 per 100 g. The control site at Baldwin Point, just outside the entrance to Still Harbor, had fairly low concentrations of 20 to 93 per 100 g. The shellfish results were less variable among days than the water samples and clearly indicated the highest concentrations were in the vicinity of the haul-out area.

Overall, the flow rates of streams found entering Still Harbor and the fecal coliform concentrations in these streams were low (Table 13). Only 4 of 15 samples taken revealed concentrations of greater than 200 per 100 ml. Three samples that exceeded 900 per 100 ml were taken from the southeast

Table 12. Concentrations of fecal coliforms (per 100 grams of tissue) in shellfish collected from Still Harbor. See Figure 4 for locations of sites.

Location	Collection date			Seal use of area
	5/22/88	7/10/88	8/02/88	
S. end Gertrude	>240,000	70,000	49,000	Main haul-out area
W. side Gertude	-	33,000	-	Secondary haul-out area
E. side Gertrude	-	-	4,900	Secondary haul-out area
S. end Still Harbor	330	170	-	200-500 m from haul-out
Baldwin Pt.	20	93	92	>1,000 m from haul out

Table 13. Flow rates (liters per minute) and fecal coliform concentrations (per 100 ml) for fresh water inputs to Still Harbor. For all but 8 May, entire shoreline of Still Harbor checked for evidence of freshwater inputs. - indicates not checked (for flow rates on 5/8) or not sampled (for concentrations).

Input code	5/08/88		5/22/88		7/10/88		8/02/88	
	Flow l/m	Conc. /100ml	Flow l/m	Conc. /100ml	Flow l/m	Conc. /100ml	Flow l/m	Conc. /100ml
A	12	920	20	2400+	0	-	0	-
B	4	33	10	46	6	49	8	1600
C	-	-	2	33	0	-	0	-
D	-	-	15	49	4	540	0	-
E	-	-	30	130	28	79	3	33
F	-	-	10*	49	0	-	1	-
G	-	-	15	-	4	130	3	110
H	-	-	20	-	8	-	1	-

* Rough estimate

end of Still Harbor just across from the haul-out area. The highest rate of flow into the harbor was found on 22 May when eight inputs were found with a total estimated flow of just over 60 liters per minute. From two to five inputs were found on the other three days with total flows of less than 50 liters per minute.

The stream outlet into the southeast end of Still Harbor is the major freshwater source of fecal coliforms into the harbor, based on flow rates and fecal coliform concentrations in the water. The origin of this contamination appears to be cattle grazing on a field that drains into the stream. Although this stream may contribute some of the bacterial levels found in water and shellfish, it appears to be secondary to the seal contribution. A water and shellfish sampling station at the southeastern end of Still Harbor were both near the mouth to this stream. Levels in water and in shellfish were lower at these sites compared to those closer to the haul-out area. Even the contamination near the stream outflow likely is not all from the stream as this is also an area where groups of up to 100 harbor seals congregate in the water both before and after haul out on Gertrude Island.

Despite the high concentrations of fecal coliforms found in water and shellfish at or near the haul-out area, other samples from the west side of Still Harbor showed fairly low concentrations. This appears to reflect the fairly rapid flushing rate of Still Harbor. Still Harbor is relatively open to Carr Inlet and is fairly shallow. A current of several knots flows through portions of Still Harbor, including off the southern end of Gertrude Island, during peak flood and ebb tides. The contamination in water we observed therefore likely reflected only recent contamination. Other than in the immediate vicinity of Gertrude Island the bacterial contamination by seals in Still Harbor does not appear to result in elevated fecal coliform concentrations.

Bacterial contamination at Quilcene and Dosewallips

We examined the temporal and geographic patterns of contamination in marine waters in Quilcene Bay and Dosewallips Delta to evaluate the role of harbor seals. Data on bacterial contamination at these sites were not a part of this study but were available from studies conducted by the Department of Social and Health Services (DSHS 1980, 1988) and Jefferson County (Welch and Banks 1987, Banks et al. 1987, Rubida et al. In prep.).

The fecal coliform levels at Dosewallips Delta were generally consistent with seals being the primary source of the contamination. Fecal coliform concentrations in marine water, shellfish, and sediment were all

highest in the southern portion of the delta (DSHS 1988), areas used as a haul-out site by harbor seals. No major sources of bacterial contamination other than the seals were found (DSHS 1988). The only evidence inconsistent with seals being responsible for the contamination is the low concentrations reported at Dosewallips Delta in 1980 (DSHS 1980). Large numbers of seals have hauled out at the Delta for many years and bacterial contamination from seals should not have changed dramatically in a few years. The sample effort in this earlier study, however, may have been too limited to detect contamination at this site.

The patterns of bacterial contamination in Quilcene Bay were not consistent with seals being the primary source of contamination. Concentrations of fecal coliforms at five marine stations monitored by Jefferson County show a general decrease from the head of Quilcene Bay southward to the mouth of the bay. The station closest to the primary seal haul-out area does not show levels significantly higher than other stations. A decreasing level of contamination with distance from the seal haul-out areas, as was observed in Still Harbor and Dosewallips, did not occur in Quilcene Bay. The distribution of contamination in Quilcene Bay is more consistent with a source at the head of the bay than it is with a source near the locations where seals congregate.

We found some evidence that the marine contamination in Quilcene Bay was related to freshwater inputs. Fecal coliform levels at two marine sampling stations showed an association with the loadings from the nearest river mouths and two stations showed an inverse correlation with salinity, also suggestive of a freshwater source. Concentrations of fecal coliforms off the mouth of the Big Quilcene River (station 3) were significantly correlated to the fecal coliform loading from the river ($n=21$, $r=.69$, $p=0.001$). Marine concentrations near the seal haul-out area (station 2) were significantly associated with both salinity ($p=0.02$) and loading ($p=0.03$) from the Big Quilcene River ($n=18$, multiple $r=0.64$, overall $p=0.02$). Additionally, marine concentrations near the mouth of Quilcene Bay (station 5) may also be related to salinity but too few samples were above detection limits to allow adequate statistical tests. We found no direct correlations, however, between the total freshwater loadings and the levels at the marine stations ($p>0.05$ for all five stations). Concentrations off the Little Quilcene River (station 1), did not show an association with loadings from the river ($n=21$, $r=.04$, $p>0.05$).

Fecal coliform loadings and comparison to other sources -

Fecal coliform loadings from seals were calculated for Dosewallips, Quilcene, and Gertrude and compared to stream loadings (Table 14 and 15).

Table 14. Fecal coliform loading (10^9 organisms per day) in Quilcene Bay, reported by Banks et al. (1987). Samples listed were collected near the mouth of the river/creek. Sampling station codes used in Banks et al. (1987) are given in parentheses.

Date	Fecal Coliform Loading (10^9 org/day)				Total
	Little Quilcene (LQ3)	Big Quilcene (BQ3)	Donovan Creek (DV2)	Cemetery Drain (CD3)	
17 Jul 86	10.6	18.8	9.6	-	39.0
28 Jul 86	75.2	3.5	4.4	-	83.1
11 Aug 86	11.0	6.3	2.6	-	19.9
03 Sep 86	16.6	8.6	-	-	25.2
13 Oct 86	2.8	0.3	0.8	-	3.9
27 Oct 86	24.5	56.5	3.0	-	84.0
13 Nov 86	5.7	69.5	-	-	75.2
25 Nov 86	17.1	27.1	3.2	-	47.4
16 Dec 86	1.2	12.1	0.7	-	14.0
06 Jan 87	13.2	14.5	21.7	18.9	68.3
30 Jan 87	28.1	10.9	14.8	14.0	67.8
02 Feb 87	66.4	7.3	18.9	7.7	100.3
09 Mar 87	12.2	7.1	23.2	5.4	47.9
02 Apr 87	1.7	0.0	0.6	19.4	21.7
28 Apr 87	8.6	4.8	16.7	9.1	39.2

Table 15. Unadjusted fecal coliform loading from harbor seals at different sites. Seal FC production is not adjusted for portion dissolving in the water column or for proportion of seals defecating at or near the site.

	Mean ^a number seals	Daily ^b feces g/day	Mean FCC ^c density 10 ⁶ per g	FC production 10 ⁹	Mean stream inputs of FC 10 ⁹
Dosewallips	180	350	190	12,000	_d
Quilcene	91	350	13	410	49e
Gertrude	345	350	63	7,600	0.33 ^f

a Based on the mean number of seals counted during visits in 1988

b Daily feces produced is based on average size of a seal (50 kg, see Calambokidis and McLaughlin 1987).

c Fecal coliform density is based on the arithmetic mean density found at that site.

d Loading reported as minimal based on low concentrations (DSHS 1988).

e Calculated from 15 samplings of freshwater inputs into Quilcene Bay from 7/17/86 to 4/28/87 reported in Banks et al. (1987).

f Calculated from 4 samplings of freshwater inputs into Still Harbor 5/8/88 to 8/2/88 conducted in this study, values from several streams were extrapolated based on results on other days.

These comparisons have a number of potential inaccuracies that need to be considered. Stream loading into Still Harbor were calculated from the data presented earlier in this report. Stream loadings into Quilcene Bay were taken from Banks et al. (1987) and are summarized in Table 14. These loading figures should be viewed with some caution because fecal coliform inputs from streams is heavily influenced by rainfall. Averaging the loading figures may not accurately reflect freshwater inputs.

Production of fecal coliforms by harbor seals (based on population sizes, defecation rates, and densities of fecal coliforms in feces) like those from streams, is variable and a number of inaccuracies exist:

- 1) Harbor seal numbers at the haul-out area are highly variable and influenced by season, tides, time of day, and weather conditions. Maximal numbers of seals counted at the haul-out area exceed the mean values by three to four fold.
- 2) Harbor seals also range widely to feed and likely defecate in areas other than near the haul-out site. The time from food consumption to initial defecation has been reported as 5-6 hours (Helm 1984) and Harvey (1987) found that 90% of fish otoliths were excreted within 24 hours of consumption. Harbor seals feeding long distances from the haul-out area may defecate away from the haul-out area. Average rather than maximum numbers of seals were used in the loading calculation to partly account for this factor.
- 3) The fecal production rate used in these calculations is taken from Calambokidis and McLaughlin (1987) and is based on the percent assimilation of prey (Pastukhov 1974, Ashwell-Erickson and Elsner 1981) and the fish consumption rates for a 50 kg harbor seal (see Ashwell-Erickson and Elsner 1981 and Calambokidis et al. 1984 for summary). Most of the consumption and defecation data comes from captive harbor seals which may differ from seals in the wild.
- 4) We found significant variations in bacterial densities by site which may reflect real differences or artifacts of the "age" of the collected feces at different sites.

Despite these limitations, the calculated fecal coliform loading from harbor seals does provide a basis for comparison to other sources of bacterial contamination. Seal fecal coliform production tended to be higher than average daily loadings found in freshwater inputs to Quilcene Bay and Still Harbor. The difference between the loading from freshwater inputs and seals is most dramatic at Still Harbor and indicates fecal

coliform contamination from streams entering Still Harbor is dwarfed by the contamination from seals. Average freshwater loadings into Quilcene are less than the loadings for seals, but the difference in loadings is not large enough to safely conclude seals are the major contribution of fecal coliforms to Quilcene Bay. The freshwater loadings indicate that there are significant human or domestic animal sources of contamination into Quilcene as reported by (Banks et al. 1987, Welch and Banks 1987).

The fecal coliform loading from harbor seals would likely accumulate in sediment near the haul-out area as well as dissolve in the water column. The fairly low recovery of seal feces at haul-out areas and observations of captive seals suggest harbor seals are more likely to defecate shortly after entering the water after having been hauled out. Experiments with seal feces in marine water indicated that not all the fecal material readily dissolved (<1% to >50%); undissolved portions settled to the bottom. In addition to the bacterial contamination of the water column, an accumulation of bacteria would be expected in the sediment, especially in the vicinity of the haul-out area.

Disease pathogens in seals

Fecal coliforms serve only as a convenient indicator of the potential presence of pathogenic bacteria. Unfortunately laboratory support was not available to test for the presence of pathogenic bacteria in the fecal samples collected for this research. Calambokidis and McLaughlin (1987) reported on bacteria isolated in 10 harbor seal feces from the Dosewallips River Delta; these samples were examined for the presence of Salmonella spp. and Yersinia spp., which were not found. Although this sample was small, bacteria that had not been reported previously in marine mammals were isolated. They included Bacillus subtilis and Aeromonas hydrophila, that are potentially pathogenic to humans. We have found no other studies that have examined the bacteria present in feces of wild harbor seals. Van Pelt and Dieterich (1973) did report bacteria isolated in the gut flora of an abandoned harbor seal pup. Because little is known about bacteria present in the gastrointestinal tracts of harbor seals in the wild, it is difficult to make any conclusions about the potential pathogens in this species. The potential of disease transmission from harbor seals to humans also is not known. An examination of the pathogenic bacteria reported from marine mammals provides some insights into the potential health risk from zoonoses (transmission of a disease from animals to human).

Many bacteria have been isolated in marine mammals; most have been isolated in the species most thoroughly studied: the small cetaceans (porpoises and dolphins) and otariids (sea lions and fur seals). A summary

of bacteria reported in marine mammals is listed in Appendix A. Studies of bacteria present in wild harbor seal populations have been more limited (Van Pelt and Dieterich 1973, Stroud and Roffe 1979, Stroud and Stevens 1980, Geraci et al. 1982, Calambokidis et al. 1985, Calambokidis and McLaughlin 1987, and Steiger et al. In press).

Bacteria that are potentially pathogenic to land mammals have been isolated in marine mammals. Neisseria mucosa var heidelbergensis, isolated in two species of wild dolphins, also has been implicated as the cause of pneumonia in children, although it was commonly found in the nasopharynx of healthy adults (Vedros et al. 1973, Smith et al. 1978). The bacterium, Erysipelothrix rhusiopathiae, found in dolphins and commonly found in fish, causes a condition in humans known as erysipeloid or "fish handlers' disease", which is usually transmitted through wound infections (Medway 1980, Seibold and Neal 1956). Salmonella (serotypes S. bovis morbificans, S. enteritidis, S. typhimurium, S. newport) have been reported in California sea lions, northern fur seals, grey seals, and bottlenosed dolphins (Jellison and Milner 1958, Johnston and Fung 1969, Schroeder et al. 1973, Sweeney and Gilmartin 1974, Smith et al. 1978, Anderson et al. 1979, Baker et al. 1980, Stroud and Roffe 1979). For salmonellae, zoonosis has been reported from domestic animals (Taylor 1984, Williams 1979); however, transmission from wildlife to humans is either rare or underreported (Williams 1979). We found no reported cases of infection from these bacteria from marine mammals to humans. Staphylococcus aureus has been isolated in a number of wild marine mammal species (Johnston and Fung 1969, Sweeney and Gilmartin 1974, Colgrove and Migaki 1976, Stroud and Roffe 1979, Baker et al. 1980), including harbor seals (Van Pelt and Dieterich 1973, Geraci et al. 1982); however, no evidence of transmission was found from captive dolphins to oceanarium personnel in a study of these bacteria (Streitfeld and Chapman 1976). Other pathogenic bacteria isolated in marine mammals include: pathogenic forms of Pseudomonas sp. (Smith et al. 1978, Medway 1980); Clostridium sp. (Keyes 1965, Hsu et al. 1974, Smith et al. 1978, Geraci et al. 1982, Buck et al. 1987); Klebsiella pneumoniae (Sweeney and Gilmartin, 1974), and Pasteurella multocida (Keyes et al. 1968).

The zoonotic potential of Leptospira interrogans, a spirochete bacterium that causes leptospirosis, from marine mammals to humans is clear. Leptospira interrogans serovar pomona has been reported in free-swimming California sea lions (Vedros et al. 1971, Smith et al. 1974a) and northern fur seals (Smith et al. 1977), and has been transmitted to humans (Smith et al. 1978). Leptospirosis is classified as an anthroozoonosis (Torten 1979), or a disease that can be transferred to humans under natural conditions. It can be spread through direct contact with urine, where

leptospire are present, or through contact with contaminated water, soil, or utensils (Torten 1979). Although every mammal is potentially capable of being infected with each leptospiral serovar (Torten 1979), pomona is the only serovar known to infect marine mammals (Smith et al. 1978).

In addition to bacteria, many viral diseases have been diagnosed in marine mammals (Smith et al. 1973, Smith et al. 1978, Smith and Skilling 1979, Britt et al. 1979, Dierauf et al. 1981, Geraci et al. 1982, Borst et al. 1986, Osterhaus et al. 1988, Kennedy et al. 1988), although little information has been reported on the zoonotic potential of these viruses. Much research has focussed on caliciviruses because of their potential for transmission to other mammals. One calicivirus, San Miguel Sea Lion Virus (SMSV), which has been isolated from five pinniped species, is readily transmissible to domestic swine and is indistinguishable from vesicular exanthema of swine virus (VESV) (Smith et al. 1973, 1974b, Prato et al. 1974, Smith and Skilling 1979, Sawyer 1976, Skilling et al. 1987). Monkeys have been infected in laboratory experiments and three research workers have developed antibodies after exposure to SMSV (Smith and Skilling 1979). Since the early 1970s, 14 calicivirus serotypes have been isolated from marine mammals, including the California sea lions, northern fur seals, northern elephant seals, Pacific walrus, and Steller sea lions (Smith 1981, Smith and Skilling 1979, Skilling et al. 1987). We have found no reports of calicivirus isolation in harbor seals; Steiger et al. (In press) reported no evidence of these viruses from 42 harbor seal pups sampled from the inland waters of Washington.

There are few cases of zoonosis reported from marine mammals (Smith et al. 1978, Medway 1980), whereas many transmissible diseases have been reported from domestic animals to humans (Taylor 1984). Salmonellae, for example, can be directly transmitted from animals to humans (Williams 1979), and has been reported as transmissible to humans from domestic cows, horses, pigs, sheep and goats, dogs, cats, and fowl (Taylor 1984, Williams 1979). Except for leptospirosis, we found no reports of disease transmission from marine mammals to humans.

It is not possible at this point to determine if seal fecal contamination poses any human health risks. This determination requires a better understanding of the diseases present in harbor seals, the presence of pathogenic bacteria in harbor seal feces, and the typing of these bacteria to determine if they are similar to human pathogens. Research reviewed here, however, indicates a wide variety of potentially pathogenic bacteria have been isolated from some marine mammal species.

CONCLUSIONS

Bacterial contamination at Dosewallips River Delta and at Still Harbor appears to be caused primarily by harbor seals. The source of the contamination at Quilcene Bay is harder to determine because a number of other sources of contamination, including domestic animals and failing septic systems, have been identified as other sources (Welch and Banks 1987). Fresh water inputs into Quilcene Bay also have high levels of bacterial contamination. The total loading of fecal coliforms from these streams appears to be smaller than the theoretical estimates of loading from harbor seals, but the calculations of loading from seals may be overestimated.

Factors consistent (+) or inconsistent (-) with seals being responsible for bacterial contamination of three regions; (?) indicates unknown:

Factor	Dosewallips	Quilcene	Still Hbr.
High contribution by seals	+	+	+
Lack of other sources	+	-	+
Contamination concentrated near seal haul-out areas	+	-	+
Historical contamination consistent with seals	-	?	?

Calculations of fecal coliform loading contributed by seals can be conducted with current data but are subject to some error. The large differences in fecal coliform concentrations in harbor seal feces from different sites is surprising. These may be the result of true differences in the bacterial populations at these sites or may reflect the differences in sampling techniques required by the varying behavior of harbor seals at sites. Until this is resolved, the accuracy of the bacterial concentrations found is subject to question and should only cautiously be extrapolated to other sites.

Future research may further refine calculations of fecal coliform loading from harbor seals. The high variability and uncertainty found in this study, even with larger sample sizes, indicates that it may be difficult to refine these loading calculations. Loading figures are valuable when there is a dramatic difference between the loads contributed by seals and those from other sources, such as in Still Harbor. In areas like Quilcene Bay, where a number of other major sources also exist,

loading figures may never be precise enough to resolve the exact role of each source.

Captive seal studies provided useful information on the behavior of feces in the water column in a closed environment. There were large differences in fecal coliform densities in captive compared with wild seals. This limits the application of results from a captive setting to the environment. Other studies using captive animals should be viewed cautiously before extrapolation to the natural environment.

The continued increase in harbor seal numbers in Puget Sound will only increase the potential for conflicts involving seals and shellfish production. Large concentrations of seals at numerous other areas of Puget Sound and Washington State exist. An increasing potential exists for seal bacterial contamination at these sites.

The human health threat posed by seal fecal contamination cannot be determined with existing data. A variety of disease pathogens potentially infectious to humans have been identified in harbor seals and other marine mammals. The degree to which these strains can be transmitted to humans through exposure to water and shellfish contaminated by seal feces is not known.

FUTURE DIRECTIONS FOR RESEARCH

Research reported here indicates that harbor seals are responsible for bacterial contamination in at least two areas of Washington State. This research, however, was limited to only three specific sites. We were also unable to evaluate if seal fecal contamination posed any human health risk. Current techniques to evaluate seal contributions remain fairly crude and are not effective where potential seal contributions are present with large inputs from other sources. Three avenues of future research would help resolve these uncertainties:

- 1) Develop techniques to identify whether bacteria in marine water and shellfish is from seals or from humans and domestic animals. Potential techniques include identification of unusual forms of *E. coli* in seal wastes using biochemical and serological techniques as well as the potential use of plasmid markers. Identification of host specific species and subspecies of bacteria, especially of the fecal streptococcus group, may also be of use in identifying the presence of seal feces. This research will require a broad spectrum of potential techniques to be tested.
- 2) Examine the geographic distribution of bacterial contamination at other locations where contamination exists in conjunction with harbor seals. Examination of fecal coliform concentrations in marine water and shellfish at stations specifically chosen to test whether seals were the source of the contamination at Dosewallips and Still Harbor were extremely valuable. A similar sampling design in Quilcene Bay and other sites would allow better determination of the source of bacterial contamination. The research needed would consist of replicate sampling of water and shellfish from sampling stations established at the locations of seal concentrations and at distances away from these locations. Counts of seals before and during the sampling would allow more specific correlation of concentrations to seal numbers. Sampling stations would also be set up at the locations of potential sources of bacterial contamination from human and domestic animals. Sampling would be conducted both during both wet and dry seasons.
- 3) Test seal wastes for the presence of pathogenic organisms to evaluate the human health significance of this contamination. There is currently only limited information on the degree to which contamination from seal feces poses a risk to human health. Fecal coliforms serve as an indicator of the presence of potentially

pathogenic bacteria that are present in human and domestic animal wastes. This research would consist of both the identification of pathogenic bacteria in seals as well as typing the bacteria to determine if they are the same form as the human pathogens.

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APPENDIX TABLE A-1

SUMMARY OF CENSUS COUNTS AT QUILCENE IN 1988

Mo	Dy	Yr	Begin time	End time	Max. time	Haul	Water	Total Pups	Tide time	Tide ht.	Comments
1	16	88	1438	1500	1445	138	3	141	0	1236	11.1
4	6	88	730	820	814	24	5	29	0	657	10.6
4	12	88	1435	1505	1440	11	4	15	0	1344	8.8
4	14	88	1700	1730	1705	4	5	9	0	1617	10.1
4	15	88	1800	1850	1845	13	12	25	0	1720	10.8
4	17	88	730	805	800	116	2	118	0	520	11.7
4	20	88	705	720	710	11	17	28	0	703	10.3
4	24	88	1203	1300	1205	40	18	58	0	1104	7.6
5	1	88	1800	1830	1805	3	2	5	0	1821	10.7
5	3	88	1800	2025	2015	21	16	37	0	1949	11.4
5	6	88	640	930	900	27	4	31	0	659	10.0
5	11	88	1510	1600	1530	0	15	15	0	1406	8.3
5	22	88	725	800	735	108	12	120	0	900	7.7
5	28	88	1545	1835	1830	29	7	36	0	1658	9.8
6	4	88	755	820	800	271	22	293	0	652	9.7
6	14	88	1700	2030	2025	21	6	27	0	1918	11.9
6	18	88	732	910	850	145	70	215	0	650	9.0
6	22	88	1050	1120	1115	5	16	21	0	1135	7.0
6	27	88	1832	1900	1855	4	9	13	0	1739	10.9
6	29	88	1920	1955	1940	41	19	60	0	1859	12.0
7	3	88	800	850	817	16	80	96	0	705	9.7
7	6	88	745	1310	1000	81	20	101	0	1116	7.9 SEE SJJ COUNT
7	10	88	1650	1805	1735	72	7	79	0	1657	10.8
7	14	88	1818	2045	2040	5	23	28	0	1933	11.9
7	20	88	828	1100	850	0	27	27	0	944	7.7
7	23	88	1500	1530	1520	12	20	32	0	1423	8.5
9	13	88	805	820	815	178	13	191	36	656	9.9 AERIAL 112 ON RAFTS
9	14	88	730	750	740	148	5	153	25	741	9.9 AERIAL 95 ON RAFTS
9	18	88	1135	1235	1210	141	3	144	22	1148	9.6
11	28	88	1405	1500	1415	100	23	123	0	1842	8.9 AFTER COUNT
12	4	88	1400	1530	1410	395	8	403	0	1235	10.8 AFTER COUNT
12	6	88	1440	1515	1447	157	4	161	0	1327	10.7 AFTER COUNT

APPENDIX TABLE A-2

SUMMARY OF CENSUS COUNTS AT DOSEWALLIPS IN 1988

Mo	Dy	Yr	Begin time	End time	Max. time	Haul	Water	Total Pups	Tide time	Tide ht.	Comments	
4	12	88	1315	1350	1345	138	74	212	0	1344	8.8	MOST P. HAULED
4	14	88	1515	1600	1550	20	38	58	0	1617	10.1	
4	15	88	1545	1650	1645	79	33	112	0	1720	10.8	
4	20	88	820	910	825	216	47	263	0	702	10.3	DIST BY PEOPLE
4	24	88	1128	1138	1130	0	52	52	0	1104	7.6	
5	3	88	1915	1945	1940	88	27	115	0	1949	11.4	SOME IN SEC. C ?
5	6	88	730	800	732	216	19	235	0	659	10.0	
5	11	88	1410	1445	1440	31	52	83	0	1406	8.3	SEALS P. HAULED
5	22	88	825	840	832	0	34	34	0	900	7.7	
5	28	88	1710	1732	1715	243	11	254	0	1658	9.8	
6	4	88	720	850	735	227	44	271	1	652	9.7	
6	14	88	1900	1935	1910	73	37	110	0	1918	11.9	
6	18	88	650	715	650	252	35	287	0	650	9.0	
6	22	88	1227	1300	1255	92	17	109	0	1135	7.0	
6	27	88	1735	1815	1810	166	32	198	0	1739	10.9	
6	29	88	1820	1900	1825	106	47	153	0	1859	12.0	
7	3	88	700	735	730	200	33	233	0	705	9.7	
7	6	88	1128	1200	1153	131	11	142	0	1116	7.9	DIST BY TOURISTS
7	14	88	1935	1950	1940	6	18	24	0	1933	11.9	? SEALS IN SEC. C
7	20	88	950	1010	956	144	55	199	0	944	7.7	DIST AT 1008 BY ?
7	23	88	1417	1430	1420	172	54	226	0	1423	8.5	DIST BY PEOP AT 1425
9	13	88	825	840	830	345	8	353	34	656	9.9	
9	14	88	800	810	805	372	0	372	29	741	9.9	

APPENDIX TABLE A-3

SUMMARY OF CENSUS COUNTS AT DUCKABUSH IN 1988

Mo	Dy	Yr	Begin time	End time	Max. time	Haul	Water	Total Pups	Tide time	Tide ht.	Comments	
4	12	88	1355	1410	1400	38	25	63	0	1344	8.8	
4	14	88	1602	1640	1635	27	15	42	0	1617	10.1	HAULED ON LOGS
4	15	88	1700	1735	1730	25	38	63	0	1720	10.8	16 H. ON LOGS
4	17	88	625	715	700	137	35	172	0	520	11.7	DIST BY ?
4	20	88	740	815	745	201	12	213	0	702	10.3	DIST BY RADIO
4	24	88	1100	1120	1105	14	35	49	0	1104	7.6	HAULED ON LOGS
5	3	88	1840	1910	1908	164	3	167	0	1949	11.4	
5	6	88	808	832	830	179	6	185	0	659	10.0	
5	11	88	1325	1405	1400	13	53	66	0	1406	8.3	BOAT OFF DELTA
5	22	88	845	900	849	31	11	42	0	900	7.7	BOATS OFF DELTA
5	28	88	1650	1700	1654	0	70	70	0	1658	9.8	BOATS NEAR DELTA
6	4	88	655	855	715	258	15	273	0	652	9.7	
6	14	88	1845	1900	1850	0	17	17	0	1918	11.9	
6	18	88	630	645	635	230	7	237	0	650	9.0	
6	22	88	1140	1220	1215	0	49	49	0	1135	7.0	
6	27	88	1640	1720	1715	127	25	152	0	1739	10.9	
6	29	88	1735	1815	1810	5	83	88	0	1859	12.0	BOAT IN RIVER
7	3	88	610	650	645	195	20	215	0	705	9.7	
7	6	88	1037	1120	1115	60	49	109	0	1116	7.9	P HAULED&ON LOGS
7	14	88	1904	1925	1910	123	25	148	0	1933	11.9	
7	20	88	920	945	930	154	61	215	0	944	7.7	
7	23	88	1350	1410	1355	0	68	68	0	1423	8.5	BOATS NEAR DELTA
9	13	88	745	800	750	312	0	312	30	656	9.9	
9	14	88	815	825	820	322	0	322	33	741	9.9	

APPENDIX TABLE A-4
SUMMARY OF FECAL COLIFORM DENSITIES IN HARBOR SEAL FECES

Location: 14-Gertrude, 23-Dose., 24-Quillcene, 99-Captive

Loc. code	Sample no.	Date Mo Dy Yr	FC concent. per gram	Log FC per g	Log Total coliform per gram
14	1	5 8 88	2.4E+08	8.38	8.380
14	2	5 8 88	<2E+3	3.30	3.300
14	4	5 8 88	2.3E+05	5.36	5.361
14	5	5 8 88	1.3E+08	8.11	8.113
14	6	5 8 88	<2E+3	3.30	3.300
14	7	5 8 88	3.3E+07	7.52	7.518
14	8	5 8 88	1.3E+07	7.11	7.113
14	10	5 8 88	3.3E+07	7.52	7.518
14	11	5 8 88	4.9E+07	7.69	7.690
14	12	5 22 88	1.3E+07	7.11	7.113
14	13	5 22 88	2.3E+07	7.36	7.361
14	14	5 22 88	2.3E+07	7.36	7.518
14	15	5 22 88	7.0E+06	6.84	6.845
14	22	7 10 88	3.3E+07	7.52	7.897
14	24	7 10 88	2.0E+04	4.30	8.806
14	27	8 2 88	3.5E+08	8.54	8.544
14	28	8 2 88	1.3E+08	8.11	8.113
23	1	5 2 88	7.3E+07	7.86	7.863
23	2	5 2 88	5.4E+07	7.73	7.544
23	3	5 2 88	7.3E+07	7.86	7.863
23	4	5 16 88	9.5E+06	6.98	6.977
23	5	5 16 88	1.7E+08	8.23	8.230
23	6	5 16 88	5.4E+08	8.73	8.732
23	7	5 16 88	<2E+4	4.30	4.300
23	8	5 16 88	1.1E+08	8.04	8.041
23	9	5 16 88	1.1E+08	8.04	8.041
23	10	5 16 88	1.3E+08	8.11	8.113
23	10	9 3 86	4.9E+06	6.69	6.690
23	11	5 16 88	9.2E+08	8.96	8.963
23	11	9 3 86	9.2E+08	8.96	8.964
23	12	6 19 88	2.3E+06	6.36	6.361
23	13	6 20 88	2.4E+08	8.38	8.380
23	13	9 15 86	3.3E+07	7.52	7.518
23	14	6 20 88	7.9E+06	6.90	7.113
23	14	9 15 86	3.3E+07	7.52	7.518
23	15	6 20 88	1.3E+08	8.11	8.113
23	15	9 15 86	7.9E+06	6.90	7.114
23	16	6 20 88	5.4E+08	8.73	8.732
23	17	6 20 88	2.2E+07	7.34	7.342
23	18	7 24 88	2.2E+08	8.34	8.342
23	19	7 24 88	7.0E+07	7.84	7.845
23	20	7 24 88	2.2E+08	8.34	8.342
23	21	7 24 88	5.4E+08	8.73	8.146
23	3	7 23 86	4.9E+06	6.69	6.898
23	4	8 19 86	9.2E+07	7.96	7.964
23	5	8 19 86	5.4E+08	8.73	8.732
23	6	8 19 86	2.2E+07	7.34	7.342

APPENDIX TABLE A-4
SUMMARY OF FECAL COLIFORM DENSITIES IN HARBOR SEAL FECES

Location: 14-Gertrude, 23-Dose., 24-Quilcene, 99-Captive

Loc. code	Sample no.	Date			FC concent. per gram	Log FC per g	Log Total coliform per gram
		Mo	Dy	Yr			
23	9	9	3	86	4.0E+06	6.60	7.690
24	11	12	4	88	4.9E+06	6.69	6.690
24	12	12	4	88	1.7E+06	6.23	6.690
24	13	12	4	88	2.2E+07	7.34	7.342
24	14	12	4	88	4.9E+05	5.69	5.690
24	15	12	4	88	2.3E+07	7.36	7.361
24	21	12	5	88	3.3E+07	7.52	7.518
24	23	12	6	88	3.3E+06	6.52	6.518
99	1	5	2	88	<1E+2	2.30	2.300
99	2	5	2	88	<2E+2	2.30	2.300
99	3	5	9	88	1.7E+03	3.23	3.230
99	4	5	9	88	4.6E+02	2.66	2.662
99	5	5	16	88	7.9E+03	3.90	3.897
99	6	5	16	88	4.9E+04	4.69	4.690
99	7	11	6	88	1.7E+04	4.23	4.690
99	8	11	8	88	7.9E+04	4.90	4.897
99	9	11	8	88	3.3E+05	5.52	5.518

Appendix Table A-5. Bacteria reported in marine mammals. References cited here are listed in References section.

BACTERIA	HOST	REFERENCE
Achromobacter sp.	bottlenosed dolphin	Johnston and Fung 1969
Acinetobacter sp.	California sea lion	Sweeney and Gilmartin 1974
Acinetobacter sp.	harbor seal	Steiger et al. In press
Acinetobacter sp.	northern elephant seal	Stroud and Roffe 1979
Acinetobacter paraptus	California sea lion	Sweeney and Gilmartin 1974
Actinobacillus actinomycetemcomitans	harbor seal	Geraci et al. 1982
Actinomyces mallei	California sea lion	Smith et al. 1978
Aeromonas sp.	harbor seal	Calambokidis and McLaughlin 1987
Aeromonas hydrophila	harbor seal	Calambokidis and McLaughlin 1987
Alcaligenes faecalis	California sea lion	Sweeney and Gilmartin 1974
Alteromonas putrefaciens	harbor seal	Calambokidis and McLaughlin 1987
Bacillus sp.	harbor seal	Calambokidis and McLaughlin 1987
Bacillus sp.	northern elephant seal	Stroud and Roffe 1979
Bacillus cereus	harbor seal	Calambokidis and McLaughlin 1987
Bacillus licheniformis	harbor seal	Calambokidis and McLaughlin 1987
Bacillus subtilis	harbor seal	Calambokidis and McLaughlin 1987
Bordetella sp.	Antarctic fur seal	Baker and Doidge 1984
Bordetella bronchiseptica	harbor seal	Geraci et al. 1982
Citrobacter sp.	California sea lion	Sweeney and Gilmartin 1974
Citrobacter sp.	harbor seal	Calambokidis and McLaughlin 1987
Clostridium sp.	Antarctic fur seal	Baker and Doidge 1984
Clostridium sp.	harbor seal	Geraci et al. 1982
Clostridium chauvoei	northern fur seal	Smith et al. 1978
Clostridium novyi	marine mammal	Smith et al. 1978
Clostridium perfringens	bottlenosed dolphin	Buck et al. 1987
Clostridium perfringens	harbor seal	Geraci et al. 1982
Clostridium perfringens	harbor seal	Hsu et al. 1974
Clostridium perfringens	northern fur seal	Keyes 1965
Corynebacterium sp.	Antarctic fur seal	Baker and Doidge 1984
Corynebacterium sp.	grey seal	Baker et al. 1980
Corynebacterium sp.	harbor seal	Steiger et al. In press
Corynebacterium sp. type 1	grey seal	Anderson et al. 1979
Corynebacterium sp. type 1	grey seal	Baker et al. 1980
Corynebacterium bovis	grey seal	Baker et al. 1980
Corynebacterium bovis type 1	grey seal	Anderson et al. 1979
Corynebacterium equi	Antarctic fur seal	Baker and Doidge 1984
Corynebacterium phocae	Antarctic fur seal	Baker and Doidge 1984
Corynebacterium phocae type 1	grey seal	Anderson et al. 1979
Corynebacterium pyogenes	California sea lion	Sweeney and Gilmartin 1974
Corynebacterium pyogenes	seal	Bonner 1970
Edwardsiella tarda	bottlenosed dolphin	Johnston and Fung 1969
Edwardsiella tarda	bottlenosed dolphin	Buck et al. 1987
Edwardsiella tarda	sea lion or porpoise	Stroud and Roffe 1979
Edwardsiella tarda	California sea lion	Wallace et al. 1966
Enterococcus sp.	seal	Bonner 1972

Appendix Table A-5. Continued.

BACTERIA	HOST	REFERENCE
<i>Enterobacter</i> sp.	Antarctic fur seal	Baker and Doidge 1984
<i>Enterobacter</i> sp.	California sea lion	Rand 1975
<i>Enterobacter</i> sp.	harbor seal	Calambokidis and McLaughlin 1987
<i>Enterobacter</i> sp.	harbor seal	Steiger et al. In press
<i>Enterobacter aerogenes</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Enterobacter aerogenes</i>	northern fur seal	Keyes 1965
<i>Erysipelothrix rhusiopathiae</i>	bottlenosed dolphin	Seibold and Neal 1956
<i>Escherichia coli</i>	Antarctic fur seal	Baker and Doidge 1984
<i>Escherichia coli</i>	California sea lion	Sweeney and Gilmartin 1974
<i>Escherichia coli</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Escherichia coli</i>	grey seal	Baker et al. 1980
<i>Escherichia coli</i>	harbor seal	Calambokidis and McLaughlin 1987
<i>Escherichia coli</i>	harbor seal	Steiger et al. In press
<i>Escherichia coli</i>	harbor seal	Geraci et al. 1982
<i>Escherichia coli</i>	harbor seal	Hsu et al. 1974
<i>Escherichia coli</i>	harbor seal	Stroud and Stevens 1980
<i>Escherichia coli</i>	northern elephant seal	Stroud and Roffe 1979
<i>Escherichia coli</i>	northern fur seal	Keyes 1965
<i>Escherichia coli</i>	sea lion or porpoise	Stroud and Roffe 1979
<i>Escherichia coli</i>	seal	Bonner 1970
<i>Escherichia coli</i> 0147:K:H52	California sea lion	Diamond et al. 1980
<i>Escherichia coli</i> anaerogenic	harbor seal	Calambokidis and McLaughlin 1987
<i>Klebsiella</i> sp.	California sea lion	Rand 1975
<i>Klebsiella</i> sp.	harbor seal	Geraci et al. 1982
<i>Klebsiella</i> sp.	sea lion or porpoise	Stroud and Roffe 1979
<i>Klebsiella oxytoca</i>	harbor seal	Calambokidis and McLaughlin 1987
<i>Klebsiella pneumoniae</i>	California sea lion	Smith et al. 1978
<i>Klebsiella pneumoniae</i>	California sea lion	Sweeney and Gilmartin 1974
<i>Leptospira interrogans</i> serovar pomona	California sea lion	Smith et al. 1978
<i>Leptospira interrogans</i> serovar pomona	northern fur seal	Smith et al. 1978
<i>Leptospira interrogans</i> serovar pomona	California sea lion	Dierauf et al. 1985
<i>Micrococcus</i> sp.	California sea lion	Sweeney and Gilmartin 1974
<i>Micrococcus luteus</i>	harbor seal	Calambokidis and McLaughlin 1987
<i>Moraxella</i> sp.	harbor seal	Calambokidis and McLaughlin 1987
<i>Moraxella kingii</i>	California sea lion	Stroud and Roffe 1979
<i>Morganella morganii</i>	bottlenosed dolphin	Buck et al. 1987
<i>Neisseria</i> sp.	grey seal	Anderson et al. 1979
<i>Neisseria</i> sp.	harbor seal	Geraci et al. 1982
<i>Neisseria</i> sp.	seal	Appleby 1964 in Bonner 1972
<i>Neisseria</i> sp.	seal	Bonner 1972
<i>Neisseria flavescens</i>	Antarctic fur seal	Baker and Doidge 1984
<i>Neisseria mucosa</i>	dolphin	Vedros et al. 1973
<i>Neisseria mucosa</i> var heidelbergensis	dolphin	Vedros et al. 1973
<i>Paracolon</i> sp.	bottlenosed dolphin	Johnston and Fung 1969

Appendix Table A-5. Continued.

BACTERIA	HOST	REFERENCE
<i>Pasteurella</i> sp.	Antarctic fur seal	Baker and Doidge 1984
<i>Pasteurella</i> sp.	harbor seal	Geraci et al. 1982
<i>Pasteurella hemolytica</i>	harbor seal	Steiger et al. In press
<i>Pasteurella hemolytica</i>	seal	Bonner 1970
<i>Pasteurella multocida</i>	California sea lion	Smith et al. 1974b
<i>Pasteurella multocida</i>	dolphin	Keyes et al. 1968
<i>Pasteurella multocida</i>	seal	Bonner 1970
<i>Proteus</i> sp.	harbor seal	Calambokidis and McLaughlin 1987
<i>Proteus</i> sp.	harbor seal	Steiger et al. In press
<i>Proteus</i> sp.	harbor seal	Van Pelt and Dieterich 1973
<i>Proteus</i> sp.	seal	Bonner 1970
<i>Proteus mirabilis</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Proteus mirabilis</i>	bottlenosed dolphin	Buck et al. 1987
<i>Proteus mirabilis</i>	harbor seal	Steiger et al. In press
<i>Proteus mirabilis</i>	northern fur seal	Keyes 1965
<i>Proteus rettgeri</i>	harbor seal	Calambokidis and McLaughlin 1987
<i>Proteus vulgaris</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Proteus vulgaris</i>	grey seal	Anderson et al. 1979
<i>Proteus vulgaris</i>	harbor seal	Calambokidis and McLaughlin 1987
<i>Providencia</i> sp.	bottlenosed dolphin	Buck et al. 1987
<i>Pseudomonas</i> sp.	harbor seal	Geraci et al. 1982
<i>Pseudomonas</i> sp.	harbor seal	Hsu et al. 1974
<i>Pseudomonas</i> sp.	harbor seal	Steiger et al. In press
<i>Pseudomonas</i> sp.	harbor seal	Van Pelt and Dieterich 1973
<i>Pseudomonas</i> sp.	California sea lion	Sweeney and Gilmartin 1974
<i>Pseudomonas aeruginosa</i>	California sea lion	Rand 1975
<i>Pseudomonas aeruginosa</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Pseudomonas aeruginosa</i>	bottlenosed dolphin	Streitfeld and Chapman 1976
<i>Pseudomonas aeruginosa</i>	seal	Bonner 1970
<i>Pseudomonas aeruginosa</i>	seal	Bonner 1970
<i>Pseudomonas bejerinckii</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Pseudomonas iridescens</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Pseudomonas mallei</i>	California sea lion	Smith et al. 1978
<i>Pseudomonas marinoglutinosa</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Pseudomonas nigrogaciens</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Pseudomonas pseudomallei</i>	marine mammal	Medway 1980
<i>Pseudomonas stutzeri</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Salmonella</i> sp.	California sea lion	Smith et al. 1978
<i>Salmonella</i> sp.	California sea lion	Sweeney and Gilmartin 1974
<i>Salmonella</i> sp.	northern fur seal	Smith et al. 1978
<i>Salmonella bovis morbificans</i>	grey seal	Anderson et al. 1979
<i>Salmonella bovis morbificans</i>	grey seal	Baker et al. 1980
<i>Salmonella enteritidis</i>	northern fur seal	Jellison and Milner 1958
<i>Salmonella enteritidis</i>	California sea lion	Schroeder et al. 1973
<i>Salmonella enteritidis</i>	northern fur seal	Stroud and Roelke 1980
<i>Salmonella enteritidis</i> Gaertner	bottlenosed dolphin	Johnston and Fung 1969
<i>Salmonella newport</i>	California sea lion	Schroeder et al. 1973
<i>Salmonella typhimurium</i>	California sea lion	Schroeder et al. 1973

Appendix Table A-5. Continued.

BACTERIA	HOST	REFERENCE
<i>Serratia</i> sp.	bottlenosed dolphin	Johnston and Fung 1969
<i>Serratia</i> sp.	California sea lion	Sweeney and Gilmartin 1974
<i>Staphylococcus</i> sp.	California sea lion	Sweeney and Gilmartin 1974
<i>Staphylococcus</i> sp.	harbor seal	Calambokidis and McLaughlin 1987
<i>Staphylococcus</i> sp. coagulase positive	dolphin	Smith et al. 1978
<i>Staphylococcus</i> sp. hem and non-	seal	Bonner 1972
<i>Staphylococcus</i> sp. hemolytic	harp seal	Wilson and Long 1970
<i>Staphylococcus albus</i>	grey seal	Anderson et al. 1979
<i>Staphylococcus albus</i>	seal	Appleby 1964 in Bonner 1972
<i>Staphylococcus aureus</i>	California sea lion	Stroud and Roffe 1979
<i>Staphylococcus aureus</i>	California sea lion	Sweeney and Gilmartin 1974
<i>Staphylococcus aureus</i>	bottlenosed dolphin	Colgrove and Migaki 1976
<i>Staphylococcus aureus</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Staphylococcus aureus</i>	bottlenosed dolphin	Streitfeld and Chapman 1976
<i>Staphylococcus aureus</i>	grey seal	Baker et al. 1980
<i>Staphylococcus aureus</i>	harbor seal	Geraci et al. 1982
<i>Staphylococcus aureus</i>	harbor seal	Van Pelt and Dieterich 1973
<i>Staphylococcus aureus</i>	harp seal	Wilson and Long 1970
<i>Staphylococcus aureus</i> coagulase positive	bottlenosed dolphin	Buck et al. 1987
<i>Staphylococcus aureus</i> coagulase-positive	harbor seal	Van Pelt and Dieterich 1973
<i>Staphylococcus epidermidis</i>	bottlenosed dolphin	Johnston and Fung 1969
<i>Staphylococcus epidermidis</i>	bottlenosed dolphin	Buck et al. 1987
<i>Staphylococcus epidermidis</i>	harbor seal	Geraci et al. 1982
<i>Staphylococcus epidermidis</i>	Antarctic fur seal	Baker and Doidge 1984
<i>Staphylococcus hyicus</i>	bottlenosed dolphin	Buck et al. 1987
<i>Staphylococcus pyogenes</i>	Antarctic fur seal	Baker and Doidge 1984
<i>Streptococcus</i> sp.	Antarctic fur seal	Baker and Doidge 1984
<i>Streptococcus</i> sp.	California sea lion	Sweeney and Gilmartin 1974
<i>Streptococcus</i> sp.	grey seal	Anderson et al. 1979
<i>Streptococcus</i> sp.	grey seal	Baker et al. 1980
<i>Streptococcus</i> sp.	harbor seal	Hsu et al. 1974
<i>Streptococcus</i> sp.	harbor seal	Stroud and Stevens 1980
<i>Streptococcus</i> sp.	northern elephant seal	Stroud and Roffe 1979
<i>Streptococcus</i> sp.	seal	Bonner 1970
<i>Streptococcus</i> sp. Group A	California sea lion	Smith et al. 1978
<i>Streptococcus</i> sp. Group F	Antarctic fur seal	Baker and Doidge 1984
<i>Streptococcus</i> sp. alpha	bottlenosed dolphin	Johnston and Fung 1969
<i>Streptococcus</i> sp. alpha	harbor seal	Steiger et al. In press
<i>Streptococcus</i> sp. alpha	harbor seal	Geraci et al. 1982
<i>Streptococcus</i> sp. alpha, hemolytic	seal	Bonner 1972
<i>Streptococcus</i> sp. alpha, hemolytic	harbor seal	Van Pelt and Dieterich 1973
<i>Streptococcus</i> sp. beta	harbor seal	Steiger et al. In press
<i>Streptococcus</i> sp. beta	harbor seal	Geraci et al. 1982
<i>Streptococcus</i> sp. beta, hem and non-	harbor seal	Calambokidis and McLaughlin 1987
<i>Streptococcus</i> sp. beta, hemolytic	harbor seal	Stroud and Roffe 1979
<i>Streptococcus</i> sp. beta, hemolytic	seal	Bonner 1972
<i>Streptococcus</i> sp. beta, hemolytic	California sea lion	Rand 1975

Appendix Table A-5. Continued.

BACTERIA	HOST	REFERENCE
Streptococcus sp. gamma	bottlenosed dolphin	Johnston and Fung 1969
Streptococcus sp. types 1-6	grey seal	Anderson et al. 1979
Streptococcus equisimilis	Antarctic fur seal	Baker and Doidge 1984
Streptococcus faecalis	Antarctic fur seal	Baker and Doidge 1984
Streptococcus faecalis	Antarctic fur seal	Baker and Doidge 1984
Streptococcus faecalis	grey seal	Anderson et al. 1979
Streptococcus faecalis	northern fur seal	Keyes 1965
Streptococcus faecalis	bottlenosed dolphin	Johnston and Fung 1969
Streptococcus thermophilus	Antarctic fur seal	Baker and Doidge 1984
Vibrio alginolyticus	bottlenosed dolphin	Schroeder et al. 1985
Vibrio alginolyticus	bottlenosed dolphin	Buck et al. 1987