13

HUMAN WASTE DISPOSAL ON BEACHES OF THE COLORADO RIVER IN GRAND CANYON

Robert A. Phillips and Cynthia Sortor Lynch

Technical Report No. 11

Colorado River Research Program

REPORT SERIES
GRAND CANYON NATIONAL PARK

United States
Department of the Interior
National Park Service

[redacted]

COLORADO RIVER RESEARCH PROGRAM
Grand Canyon National Park
Grand Canyon, Arizona 86023

The Colorado River Research Program was initiated by the National Park Service in 1974 to secure scientific data to provide a factual basis for the development and the implementation of a plan for appropriate visitor-use of the Colorado River from Lee's Ferry to Grand Wash Cliffs and for the effective management of the natural and cultural resources within the Inner Canyons. The intensified research program consists of a series of interdisciplinary investigations that deal with the resources of the riparian and the aquatic zones and with the visitor-uses including river-running, camping, hiking, and sight-seeing of these resources, as well as the impact of use and upstream development upon canyon resources and visitor enjoyment.

Final reports that result from these studies will be reproduced in a series of Program Bulletins that will be supplemented by technical articles published as Program Contributions in scientific journals.

Merle E. Stitt, Superintendent
R. Roy Johnson, Program Director

Cover Drawing by J.G. Carswell, University of Virginia

HUMAN WASTE DISPOSAL ON BEACHES OF THE COLORADO RIVER IN GRAND CANYON

Robert A. Phillips and Cynthia Sortor Lynch

Technical Report No. 11

Grand Canyon National Park
Colorado River Research Series
Contribution No. 39

Final Report

HUMAN WASTE DISPOSAL ON BEACHES OF THE COLORADO RIVER IN GRAND CANYON

Submitted to

National Park Service
Grand Canyon National Park
Grand Canyon, Arizona

Purchase Order No. 8210-6-1426

Prepared by

Robert A. Phillips, Ph.D.
Principal Investigator

&

Cynthia Sartor Lynch
Graduate Assistant

Department of Civil Engineering & Engineering Mechanics
The University of Arizona
Tucson, Arizona 85721

February 1977

TABLE OF CONTENTS

LIST OF TABLES

LIST OF TABLES--continued

LIST OF ILLUSTRATIONS

ABSTRACT

An estimated 20 tons of human waste solids are generated by the more than 14,000 persons participating in river trips on the Colorado River through the Grand Canyon National Park. The objectives of this research were to investigate the potential for public health hazards due to waste burial on the beaches, the efficiency of presently used disposal methods, and the feasibility of alternative disposal methods.

A review of the literature shows that the survival of fecal coliform and pathogenic bacteria in soils is reduced by low moisture and high temperatures. Fecal coliform levels in actual disposal sites in the summer were shown to be reduced by greater than 99.99% within one month. Although low temperatures result in reduced die-off rates, few organisms in September dumps survive the winter to carry over into the next season. The level of fecal coliforms on the beach surfaces were generally found to be close to or below the minimum detectable density of 0.02 fecal coliforms/g of sand (dry weight).

The major methods of consolidation and disposal presently used include burial of the contents of disinfected or untreated holding-toilets, or using dry-hole latrines. These methods appear to be satisfactory provided that the waste matter is buried sufficiently deep.

Improvements in present practice are recommended, however. Latrines or holding toilets should be set up at attraction and lunch stops to discourage surface dumping by individuals. The installation of pit privies should be considered, but only at major attractions such as at Havasu Creek or where backpackers add to the visitor impact. The carryout procedure of all waste solids as practiced by at least one concessionaire should be encouraged. This will reduce the loading on the beaches and is especially important if visitor usage is increased. Under no circumstances should direct disposal of human wastes to the river be allowed.

Trip leaders must assume greater responsibility for maintaining a waste management discipline for their party. Private parties must be better informed regarding the methods of, and the necessity for, satisfactory waste disposal. Finally, improper food sanitation must be recognized as a link between improper waste disposal and enteric diseases.

Table I

Number of People Running the Colorado River Through the Grand Canyon*

	1975			1976	
Month	Recreational Users	Research + crews	Length of trip, days	Recreational Users	Research + crews
January	-	-	-	-	14
Feb	-	-	-	-	25
March	219	95	10.0	152	45
April	328	113	9.0	346	114
May	1772	330	8.4	1876	483
June	2163	399	9.8	2832	523
July	2357	412	8.8	2675	538
Aug	2581	483	8.5	2618	459
Sept	1097	272	9.0	862	260
Oct	6	198	6.5	-	-
Nov	-	6	-	-	-
Dec	-	-	-	-	-
Total	10523	2308		11361	2461

* Data for average length of trip, ref. 2; otherwise, ref. 1.

CHAPTER 1

INTRODUCTION

In 1975, a total of 14,305 people participated in river trips through the Grand Canyon [1]. Approximately 95% of this river running took place between May and September. The 1975-1976 data of those persons starting at Lee's Ferry are presented in Table I [1,2]. Assuming an average of 8.8 days per trip and 150 grams of feces per person per day, this represents 20 tons of solid human wastes which require disposal during this five-month period.

Historically, the predominant method of human waste disposal has been beach burial. National Park Service regulations, instituted in 1972, require river parties to consolidate their body wastes and bury it in holes at least two-feet deep, six-feet above the normal high river fluctuation line, 50-feet from the river bank and at least 200-feet from normal camping areas [3].

There are two major methods of consolidation and disposal. Most of the larger commercial parties carry a portable, chemical, holding-toilet which is set up on the beach every evening. In the morning, a hole is dug and the toilet is dumped and rinsed. Other parties use a latrine-type system. When the river party makes camp a hole is dug and a portable toilet seat is set over it. When the camp is abandoned, the hole is filled.

Although the majority of human waste is disposed of in compliance with regulations, there is a certain amount of clandestine dumping by individuals. This is unavoidable in some instances as toilets are not set up during lunch or at attraction stops. At other times river-runners either do not want to take the time and trouble to dig adequate holes, or they find the portable toilets unattractive because of odors and flies or, in general, incongrous with their "wilderness experience." The clandestine disposals take the form of either shallow holes, i.e., "sneaker" or "cat" holes, or direct surface dumping.

The problem with any of these methods is that sewage solids end up at or relatively close to the surface and, since the beach sand is unconsolidated, may be readily exposed by wind and water erosion or by trampling. Another common occurrence is re-exposure of waste solids by a second burial attempt at the same site. All problems are compounded as the usage rate of a particular beach increases.

A research report submitted to the National Park Service in 1974 [4] indicated that enteric organisms associated with human body wastes could survive the colder winter months and actually increase in numbers as temperatures increased in the spring. Therefore, it was concluded that the health of river-runners was potentially endangered. This investigation was initiated by the Park Service in June 1976 to determine the survival rates of enteric organisms in dump sites during the warm summer months.

The primary objective of this investigation was to better define the risk of disease transmission among river-runners due to present methods of human body-waste disposal. To this end, several factors were analyzed: the probable frequency of occurrence of various enteric pathogenic organisms in association with the dumps and the survival of these organisms in the beach environment, the general contamination level on the beaches, the efficiency of the major methods of waste disposal and the resulting levels of surface contamination, the probability of contact, and the relationship between the number of disease organisms ingested and the occurrence of the specific disease. Based on the results of this investigation, the advantages and disadvantages of present and alternative methods of disposal are described, and some recommendations as to a suggested course of action are made.

CHAPTER 2

OCCURRENCE OF PATHOGENIC MICROORGANISMS IN ASSOCIATION WITH DUMPS

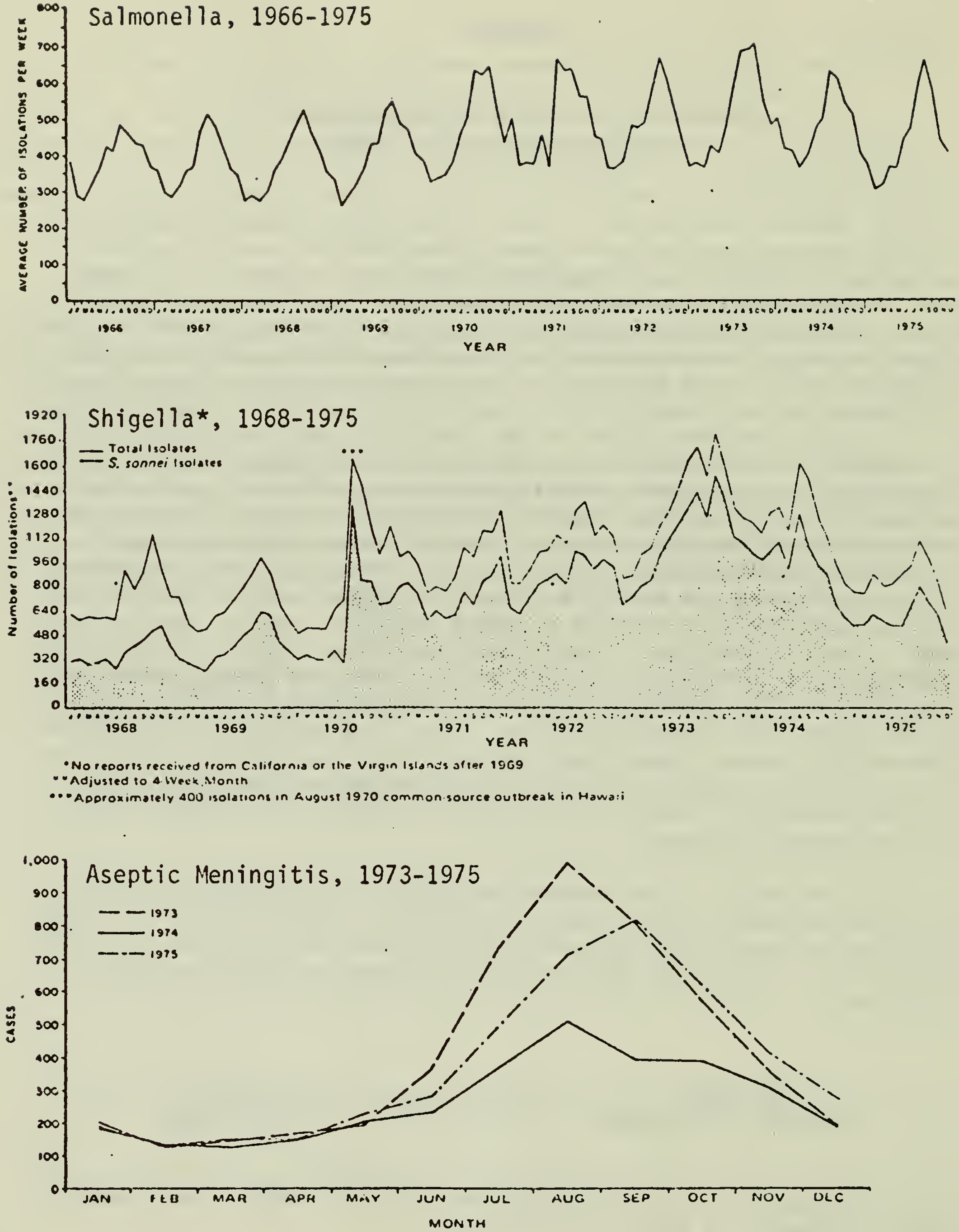
For general information, a list of pathogenic enteric bacteria, viruses, and protozoa (with the associated clinical conditions) which are relevant to this investigation is presented in Table II. "Relevant" illnesses would be those which may be transmitted via human waste. This means that the pathogenic organisms involved must be able to pass through the acid barrier of the stomach and multiply in the intestinal tract to be excreted in the feces or urine of the infected individual.

The occurrence of disease organims in beach dumps is directly proportional to the number of infected persons who enter the Canyon and the sanitary conditions during the course of the trip. Data for the reported incidence of enteric disease for the United States in 1974 and 1975 are presented in Table III. Although it may be assumed that river-runners are healthier than the general population, the level of occurrence of enteric diseases could be close to the national level. As refrigeration of meats and vegetables, as well as sanitary practices in drinking and food preparation, are not optimum among most river parties, the incidence levels of salmonellosis and shigellosis may be higher than those nationwide. Also, the seasonal incidence of salmonellosis shows a consistent pattern, with the greatest number of isolations being reported in July through November--the peak months for river-running. This is also true for shigella and aseptic meningitis as shown in Figure I.

As an example, if one assumes the level of occurrence of hepatitis to be the same on the river as nationally, the probability in any given year of anyone coming down with hepatitis while on the river is about 0.09, or about one case in every 10 years, assuming constant conditions.

$$p = \frac{25 \text{ cases}}{100{,}000 \text{ people-years}} \cdot 14{,}300 \text{ people} \cdot \frac{8.8 \text{ days}}{356 \text{ days/year}} = .09$$

In 1975, a member of a commercial river party did suffer the onset of infectious hepatitis while on the river.



*No reports received from California or the Virgin Islands after 1969

**Adjusted to 4-Week Month

***Approximately 400 isolations in August 1970 common-source outbreak in Hawaii

Figure 1. Seasonal Trends in Salmonella, Shigella, and Aseptic Meningitis [7]

Table II

Enteric Diseases and Infections

Organism	Disease or Infection
Bacterial infections [5]	
Klebsiella pneumoniae	Highly fatal type of pneumonia or lesions which may occur in any part of the body.
Proteus spp.	Genito-urinary and gastro-intestinal tract diseases.
Salmonella (180 different serotypes reported in 1973)	
S. typhi	Typhoid fever may be transmitted by flies; incubation period, 10-14 days.
S. paratyphi (type A)	Paratyphoid fever - resembles typhoid, but less severe.
S. schottmuelleri (type B)	
S. hinschfeldii (type C)	
S. typhimurium	Three most commonly isolated serotypes causing salmonellosis, an acute gastrointeritis with diarrhea. Symptoms occur within a few hours of infection.
S. newport	
S. enteritidis	
Shigella dysenteriae	Most severe form of shigellosis or bacterial dysentery. Several other species of Shigella may cause the disease. The incubation period is 1-7 days (4 av.)
Virus diseases [6]	
Poliovirus	Paralytic poliomyelitis, aseptic meningitis.
Coxsackie	
Group A	Herpangina, aseptic meningitis.
Group B	Pleurodynia, aseptic meningitis.
Infectious Hepatitis	Infectious hepatitis. The incubation period is 15-50 days (25 av.)
ECHO	Aseptic meningitis, "summer" rash, diarrheal disease.
Adenovirus	Respiratory and eye infection.
Protozoan diseases [5]	
Entamoeba histolytica	Amebiasis with symptoms ranging from abdominal discomfort to severe dysentary.

Table III

Reported Cases of Enteric Diseases for the United States in 1974 and 1975 (from Morbidity and Mortality Report [7])

DISEASE	1975	1974
Amebiasis	2,775	2,743
Aseptic meningitis	4,475	3,197
Hepatitis A	35,855	40,358
Hepatitis B	13,121	10,631
Hepatitis, unspecified	7,158	8,351
Poliomyelitis, total	8	7
Paralytic	8	7
Salmonellosis, excluding typhoid fever	22,612	21,980
Shigellosis	16,584	22,600
Typhoid fever	375	437

For many diseases, the reported incidence may not reflect the actual incidence. For example, although only 1.3 cases of amebiasis/100,000 population were reported in 1975, authorities estimate that 10 million persons in the United States harbor the parasite and two million have symptomatic cases of the disease [5]. Although these persons have few clinical indications of the infection, they have the potential to transmit the infective cysts to others for whom the parasite may have graver consequences. Based on the above estimates, one out of every 21 people who run the river harbor the parasite.

CHAPTER 3

DEATH RATE OF ENTERIC MICROORGANISMS IN THE BEACH ENVIRONMENT

Literature Review

Die-off Rate of Enteric Pathogens in Soil

The possibility that current body waste disposal practices may present a potential hazard to the health of river-runners is directly dependent upon the die-off rate of pathogenic organisms in the beach environment. In investigations of stormwater pollution and of wastewater reuse by land application, the survival of intestinal pathogens in soil has been the subject of extended studies. Such investigations date back as far as 1889. However, the results of many of the early investigators are of only relative importance since they are reported as lengths of survival rather than as survival rates.

It is very important at this point to emphasize the concept of death as a rate process. Temperature, moisture, etc., affect the rate of death of organisms. An adequate designation of the rate of death would be a rate constant obtained from an equation such as Chick's Law: $N_2 = N_1 \exp(-kt)$, where N_1 equals the initial density of organisms; N_2, the density of organisms at time t; and k (base 10) the rate constant. The rate can be denoted also by recording the time required for the death of a given percentage of the organisms. The length of survival time, in itself, is not an adequate specification of the death rate as it is a function of the initial concentration of organisms as well as the rate of death. The same holds true for the number of organisms at any given time. If it can be assumed that the initial density of organisms in a number of samples was the same, then the length of survival would impart some information regarding the relative hostility of the conditions under which the soil samples were held. In this context, Table IV summarizes some of the early studies as reported by Rudolfs et al. [8], Beard [9], and Pound and Crites, [10].

In 1940, Beard [9] studied the survival of S. typhi in various types of soil in clay flower pots exposed outdoors. Some of his results are summarized in Table V.

Table IV

Survival of Pathogenic Microorganisms Cited in Early Literature

Organism	Survival	Reference
Salmonella typhi in soil		
a. soil	5½ months	(11)
b. natural soil	3 months	(12)
c. various soils	12-42 days	(13)
d. dry soil	28 days	(14)
moist soil	88 days	
e. soil with frequent addition of nutrients	315 days	(15)
soil without nutrient addition	86 days	
f. soil	404 days	(16)
sterilized soil	<404 days	
g. sterilized soil	216 days	(17)
unsterilized soil	100 days	
h. dry sand	25 days	(18)
moist sand	≤74 days	
soil exposed to 122 hours of sunlight	≥22 days	
i. dry soil	99% destroyed in 2 weeks	(19)
moist soil	>2 weeks	
j. sandy soil with addition of sterile sewage-hot house conditions	74 days	(20)
k. garden soil	30-36 days	(21)
sterile sand	55 days	
l. moist soil	80 days	(22)
dry soil	20 days	
acid soil	10 days	
Salmonella typhi in feces or sewage sludge		
a. feces in privy vault	5 months	(23)

Table IV--*continued*

b. feces in privy vault	5 months	(24)
feces - 10 days in privy vault		
+ 20 days on surface	30 days (total)	
+ 40 days buried	50 days (total)	
c. feces	10 days	(22)
septic tanks	8-15 days	
d. sludge at 50-60° F	99.9% reduction in 28 days	(25)
sludge at 68-72° F	99.9% reduction in 7 days	
e. raw sewage on surface of soil	46 days	(10)
raw sewage on lower layers of soil	70 days	
air-dried digested sludge	≥17 weeks	
<u>Shigella dysenteriae</u>		
a. in feces	8 days	(22)
b. in raw sewage on grass	6 weeks	
Bovine tubercle bacilli	≥5 months (winter)	(26)
a. in feces	≥2 months (spring)	
	≥4 months (autumn)	
	<2 months (summer)	
b. in dung	178 days	(27)
with 75% soil & 25% dung	122 days	
in soil	85 days	
Enteroviruses in soil	12 days	(10)
Streptococci		
a. in soil	35-63 days	(10)
b. on surface of soil	38 days	(10)

Table V

Survival of S. typhi in Various Soils Exposed Outdoors (from Beard [9])

Precipitation	Loam	Loam-Sand Mix	Sand
	(Days to 99% Reduction)		
10.1 in. in 120 days	49	33	6
3.2 in. in 49 days	17	-	2
2.0 in. in 28 days	8	-	1
0.7 in. in 42 days	10	6	1

Beard attributes the higher die-off rate of organisms in sand versus loam and in dry versus wet seasons to lower moisture; however, soil moisture was not monitored. Average minimum-maximum temperatures for the 3.2 and 2.0 in. precipitation seasons were 4-14 and 12-23°C, respectively. No comment was made on the possibility that temperature might exert an influence on death rate beyond the drying effects. It should be noted that these soils were inoculated with saline suspensions of S. typhi rather than fecal suspensions.

Several other facets of Beard's investigation are of interest in the context of this study. In experiments with loam-peat mixtures, the death rate was 36% higher in the top layers of samples placed in the sun than of those in the shade, although moisture loss was the same. In samples stored under freezing conditions, it took 18 months for a 99% reduction of S. typhi in loam. When fecal suspensions of S. typhi were used to inoculate loam, survival was reduced in the wet season and unaffected in the dry.

In another study [28], where soil moisture and temperatures were maintained at constant levels (not specified), a 99% reduction of S. typhi in sand, sandy-loam, clay-loam and muck occurred in approximately 1, 2, 3 and 6 days, respectively.

The survival of Salmonella enteritidis and fecal coliforms under controlled conditions was studied by Dazzo et al. [29] in 1973. Sand samples were obtained from established fields receiving 0, 1.27, 2.54 and 5.08 cm of cow manure slurry per week. The samples were maintained in sealed bottles with a soil moisture of 10.2% and incubated at 22°C in the dark.

Chemical analyses of the samples revealed increasing percentages of organic matter and organic carbon, and total nitrogen, and increasing cation exchange capacities and exchangeable phosphorus corresponding with increasing rates of irrigation. Data on times required for 90 and 99% reductions of _S. entiritidis_ and fecal coliforms are shown in Table VI.

Table VI

Survival of Fecal Coliforms and _S. enteritidis_ in Soils Previously Receiving Various Amounts of Manure Slurry (from Dazzo et al. [29])

Previous Irrigation Rates (cm/wk)	Time Required for Reduction (days) Fecal Coliforms 90%	99%	S. enteritidis 90%	99%
0.00	4.0	7.5	2.0	4.5
1.27	4.0	8.0	3.5	8.0
2.54	6.5	10.5	4.5	10.0
5.08	8.5	13.5	6.0	13.0

Hence, survival of salmonella and of fecal coliforms is extended by the addition of nutrients.

With soils held at 28°C, Rhines [30] recorded 90% reduction times for the avian tubercle bacillus ranging from 11 days in dry soil to 30 days in soil at 25% moisture.

Mirzoev [31] investigated the self-purification capabilities of soil and water in the far north (USSR). His results indicate that low temperatures (ranging from -40 to 5°C) are not only very favorable for the preservation of shigella in the environment (feces, soil and water), but also contribute to inhibiting the variability of these organisms in the direction of saprophytization.

Bagdasaryan [32] showed that the survival of enteroviruses in soil is affected by pH, temperature, and moisture. Five strains of enterovirus were tested: one pathogenic and one attenuated strain of Poliomyelitis 1, ECHO-7, ECHO-9, and Coxsackie B3. Sterile and non-sterile 10-g portions of either a sandy or loamy soil were inoculated with one of the virus strains and incubated at either 18-23°C or 3-10°C. Among the five strains of enterovirus, no one strain evidenced consistently greater survival for the various combinations of soil conditions.

In one set of experiments, air dried, non-sterile, sandy soil at pH 5 was incubated at 18-23°C. The 90% reduction times of TCD_{50}/gram were 6 to 12 days. Survival was more than doubled at 10.42% moisture.

For the results in Table VII, the moisture was constant at 10.4%. Although Bagdasaryan reported the majority of his data on survival by giving the last day of survival, the average values shown in Table VII may be regarded as the time required for a 99.9% reduction.

Table VII

Survival of Enterovirus in Sterile and Non-sterile Sandy Soil Under Various Temperature and pH Conditions (from Bagdasaryan [27])

	Average of Last Day of Isolation for 5 Strains of Enterovirus			
	18-23°C		3-10°C	
	pH5	pH7.5	pH5	pH7.5
Sterile	62	106	132	160
Non-sterile	41	81	114	134

It was also found by Bagdasaryan that desorption of the enteroviruses from the soil particles was better in an alkaline medium. This may account in part for the increased length of survival at pH 7.5 over that at pH 5. It was concluded by Bagdasaryan that the temperature factor has a much greater influence over survival of enteroviruses in soil than the reaction of the soil within pH values of 5 to 7.5.

Similar results were reported by Duboise et al. [33]. Dechlorinated final effluent from wastewater treatment plant was inoculated with Poliovirus L and applied to loamy-fine-sand cores. At 20°C, the density of poliovirus was reduced by 90% after 8 days in the soil column and 9 days in the effluent. After 84 days, the survival in these samples was less than .00001% of initial density. At 4°C, the poliovirus survival is prolonged. The survival after 84 days was 75% in the effluent and 12% in the soil column. Two other reports indicate equal or more extended poliovirus survival in soil [34,35].

That virus survival is highly temperature dependent, also has been shown by Sobsey et al. [36]. Survival of enterovirus in landfill leachate was reduced by only 50% after 28 days at 4°C, by 90% after 16 days 20°C, and by 90% after only one day at 37°C.

Die-off Rate of Indicator Organisms in Soil

As has been indicated, a wide variety of intestinal pathogens are of concern to investigators in studies of stormwater and soil pollution. The presence or absence and density of any specific type of pathogen is not predictable except under controlled conditions. The detection of many types of intestinal pathogens may require involved and time consuming procedures or, as is the case of viruses, prohibitively expensive equipment. For these reasons, expedience dictates the use of indicator bacteria in many investigations of water or soil contamination.

By definition, the presence of an indicator organism signifies that pollution has occurred. The relationship of fecal coliform and, to a lesser extent, the total coliform density in soils to probable human and animal contamination has been demonstrated by Geldreich [37].

The most important property of indicator bacteria in the context of this study is that ecological factors which influence the survival of intestinal pathogens have similar effects upon the indicator organisms. For example, note the similarity in the survival rates of the Salmonella species and the fecal coliforms in the study by Dazzo et al. [29], presented in Table VI. Soil studies conducted with indicator organisms--coliforms, fecal coliforms, and fecal streptococci--have shown the importance of such factors as sunlight, temperature, rainfall, soil moisture, pH, organic matter and the presence of other microorganisms to survival.

Cuthbert et al. [38] examined the influence of pH on the survival of fecal coliforms and fecal streptococci in bottled soil incubated at 18°C in the dark. In five soil samples at pH 3.8-4.5, the survival of fecal coliforms was 0.1% in 24-50 days and the survival of fecal streptococci was 0.1% in 45 to 63 days. In five soil samples from acid moorlands (pH 2.9-3.7), 99.9% or more of the fecal coliforms or fecal streptococci disappeared within 10 days. Raising the pH of peat soils to 5.6-6.3 resulted in fecal coliforms multiplying to a high level and persisting at that level for at least 110 days. Fecal streptococci showed no multiplication, but the survival to the 0.1% level was increased to 68-110 days.

A summary of some of the results of Ostrolenk et al. [39] is presented in Table VIII. This study showed that lower temperatures result in extended survival of E. coli, that addition of unsterilized manure decreases survival, and that survival is shorter in feces than in soil. Fecal streptococci were also monitored, and it was shown that they were most persistent in soil and equal or less persistent in feces.

Table VIII

Survival of E. coli in Soil and Feces (from Ostrolenk et al. [39])

Condition	Temperature	Survival of E. coli
Virgin soil	room	.1% at 109 days
Soil fertilized with chicken manure	room	.1% at 21 days and .01% at 40 days
	7°C	.1% at 109 days
Rat feces	room	1% at 66 days
	7°C	10% at 130 days
Mouse feces	room	.1% at 66 days
	7°C	.1% at 130 days

Using soil in bottles with cotton stoppers, Young and Greenfield [40] found that optimum moisture conditions for survival of coliforms was 10 to 40% of saturation. In samples held at 60 to 100% of saturation, the die-off rates were very rapid, and were higher in unsterilized than sterilized soil.

Mallman and Litsky [28] treated Fox sandy loam in large metal cyclinders located outdoors with added sterilized raw sewage sludge in concentrations of 0, 2, 5, 10 and 20%, based on total weight. Their results, as shown in Table IX, indicate that an increase in the nutrient content of the soil results in an increase in the survival of coliform bacteria. The importance of added soil nutrients has also been documented by Dazzo et al. [29] in their study with S. enteritidis and fecal coliforms which was discussed previously.

Table IX

Survival of Coliforms in a Sandy-loam Receiving Various Doses of Sterile Raw Sewage Sludge (from Mallman and Litsky [28])

Conc. of Raw Sludge Added %	Weeks to 90% Reduction of Coliforms
0	3.6
2	3.9
5	5.5
10	6.2
20	6.0

Mallman and Litsky also concluded that the natural occurring organic content of various soils (sand, loams and clays) would effect the survival of fecal streptrococci and coliforms. This conclusion, however, was based on the last day of isolation of the organisms from various soils; and, by chance, higher numbers of streptococci and coliforms were seeded into the loam than into the sand. Table X shows their results in terms of weeks to a 90% reduction.

Table X

Survival of Coliforms and Fecal Streptococci in Various Soil Types (from Mallman and Litsky [28])

Soil Type	Weeks to 90% Reduction	
	Coliforms	Fecal Streptococci
Oshtemo Sand	2.6	1.3
Fox Sandy-loam	3.3	1.4
Isabella Loam	3.4	1.3
Brookston Clay-loam	3.8	1.8

There is actually very little difference between the real death rates for the various natural soils, especially when compared with the increase in death rates caused by addition of nutrients as was shown in Table IX. The naturally occurring organic content of a soil seems to be unavailable to enteric organisms as a nutrient source.

An in depth investigation of the survival of indicator bacteria in soil under natural conditions was carried out by Van Donsel et al. [41] over three-years. Two outdoor plots--one in a shaded site on the north slope of a steep wooded hill and another in a level lawn area with sparse vegetation and no protection from direct sunlight--were dosed repeatedly with specifically identifiable "tracer" fecal coliform and fecal streptococci strains. Death rates for both organisms were calculated for different seasons at both sites.

The 90% reduction times are shown in Table XI. The effects of environmental extremes at the exposed site is most notable in the summer and autumn when the death rate is approximately twice as fast as that of the protected hillside site. During the summer, the soil moisture in the shaded site was consistently higher and subject to less fluctuation than the exposed site. In the shaded sites, soil temperatures were also lower.

There also existed a seasonal shift in the relative survival of the two indicator organisms. In the summer, the fecal coliforms survived slightly longer than the fecal streptococci; in the autumn, there is little difference; and in winter and spring, the fecal streptococci survived about twice as long as the fecal coliform.

When the levels of the fecal coliform tracer were low, it was possible to monitor the non-fecal coliform population. Under non-freezing conditions, an increase in non-fecal coliforms would occur after a rainfall. The magnitude of the increase was not proportional to the amount of precipitation, but to the temperature conditions following the rainfall. Very warm weather following a rain shower would cause a considerable increase of up to 100-fold in soil coliforms, whereas the increase would be more moderate when cool weather followed rain. High atmospheric humidity in conjunction with elevated temperatures also initiated increases in non-fecal coliforms. An exception to the positive association of soil moisture with coliform counts occurred under sub-freezing conditions, when very high (above 35%) soil moisture levels caused by frost or snow were associated with decreasing numbers.

While there appeared to be occasional occurrences of aftergrowth for both tracer organisms in wet weather it was not so evident as with the non-fecal coliforms.

Table XI

Death Rates for Fecal Coliforms and Streptococci for Different Seasons at Exposed and Shaded Sites
(from Van Donsel et al. [41])

	Days to 90% Reduction of Organisms			
	Fecal Coliforms		Fecal Streptococci	
Season	Shaded	Exposed	Shaded	Exposed
Spring	4.2	6.4	14.8	12.1
Summer	7.6	3.4	5.5	2.8
Autumn	13.4	5.4	12.7	6.8
Winter	10.4	10.4	19.6	19.6

Summary of Factors Affecting Survival of Enteric Organisms in Soil

The results of the studies presented in the literature suggest several factors which affect the survival of enteric organisms, applying equally, it seems, to Salmonella typhi, fecal coliforms, and enteroviruses, although resistance of individual organisms may vary:

1. Soil moisture. Optimum soil moisture for survival of enteric organisms appears to be about 10 to 20% of saturation. Drier conditions result in increased die-off rates.

2. Nutrients. Addition of organic increases the survival of organisms, not only because they serve as nutrient source, but because the increased organic content of the soil results in increased moisture-holding capability.

3. pH. While extremely acidic or basic conditions (pH < 5 or > 10) definitely result in low survival rates, the pH within the range of ordinary soils is not a dominating factor in the survival of enteric organisms.

4. Sunlight. A significantly higher death rate is shown for enteric organisms in soil exposed to sunlight, aside from its heating and drying effects.

5. Temperature. In every instance (for S. typhi, E. coli, infectious hepatitis, bovine tubercle bacillus and enteroviruses in soil and in feces) low temperatures (above freezing) favor survival.

In general, higher temperatures result in increased die-off rates. Although the literature reveals a few cases of multiplication of organisms when higher temperatures occur in conjunction with adequate moisture and nutrients, this phenomenon is most often associated with the indigenous soil bacteria. When the population of the soil bacteria increases, the survival of the enteric organisms in the soil is decreased. In any case, aftergrowth is short-term and usually followed by an accelerated death-rate as nutrients are exhausted, drying effects become more pronounced, or other organisms more favored by the soil environment predominate.

Previous Investigation of Waste Disposal on Beaches in Grand Canyon

In September 1975, the National Park Service initiated an investigation into the potential health hazard of human waste buriel on the beaches along the Colorado River in the Grand Canyon [4]. The chemically-treated contents of a portable, holding-toilet serving 30 (first trip) or 16 (second trip) people were disposed of following established procedures for the commercial party involved. Nine of the dump sites were suitably marked so that the sites could be easily relocated. Once a month--except December--for 9 months (September 1975 through May 1976), one core was taken at each dump site. The surface and bottom temperatures were determined; and the top, middle, and bottom portions of each core were examined for total and fecal coliforms. These analyses were carried out on the day of sample collection after camp had been set up.

Several observations should be made concerning the methodology of this investigation and the reliability of the data presented in the report.

1. Because only one incubator was available, the total coliform plates were incubated at the same temperature as the fecal coliform plates--namely 44.5°C. Since the ability to grow at 44.5°C separates fecal coliforms from non-fecal coliforms, the total coliform plate counts might be regarded as duplicate fecal coliform analyses.

2. Due to limited incubation space, only one dilution (namely 1:16) was run. Hence, the numbers reported represent membrane filter counts per 1/16 gram of beach soil (wet weight). However, each determination was carried out in duplicate.

3. Membrane filter colonies are best counted with a magnification of 10-15 diameters [42]. Because a microscope and good light source were not available, the plate counts were considered "Too Numerous to Count (TNTC)" if there were more than 25 colonies, and the results were reported as zero, 1-25, or TNTC. These correspond to < 16, 16-400, or > 400 organisms/gram (wet weight).

4. It would be helpful to know the initial density of the fecal coliforms in the dumps. This number was not determined in the field, but can be estimated. Assuming 13.0×10^6 FC/g (wet weight) of feces, 150 g (wet weight) of feces/24 hr/person [37], 16 people, a 10-in. diam. by 2-ft deep hole with fecal matter spread homogenously through the bottom 16 in., and sand with S.G. of 2.6 and 40% porosity, the initial density of fecal coliforms would be approximately 800,000/g dry weight of sand.

5. Only a summary of the data (which combined the total and fecal coliform data in an unexplained way) was presented in the report. However, for the months of April and May, the original data were available. Interpretation of the data was difficult as the growth on the fecal coliform plates was not usually in the form of typical colonies. "Blue ring" or "2 blue squares" are examples. However, in comparing the original data with the summary given, four of the analyses would seem to be incorrectly summarized in April, and eight of eight in May.

In any case, from the results of this study, it would be hoped that the cold-weather death rates of fecal coliforms might be obtained. According to a corrected summary, the fecal coliform densities in three out of nine dumps were down to the 1-25 or zero level by February. This represents a decrease from about 800,000 to 400 or less FC/g, or a 99.95% decrease in five months. Three more sites had decreased to that level by April, and all but one were less than 16 FC/g by May (a 99.98% decrease). The April and May data would not seem to support the stated hypothesis of multiplication of organisms.

From the results indicated, the death rate is very slow over the colder winter months; but the number of enteric organisms is reduced 99.9%, or more by the time the next river-running season begins. Unless low levels of pathogenic organisms can cause disease, the health hazard of wastes from past seasons is probably not significant.

Death Rates of Enteric Organisms in Beach Disposal Sites

In order to determine the warm-weather die-off rates of enteric organisms in established beach disposal sites, the study initiated by Knudson [4] was continued during the summer of 1976. The sites were established and sampling was carried out by the same procedure as that used in the September 1976 study; however, the authors found it necessary to make certain modifications in the analytical procedures.

General Procedure

The microbiological analysis of samples taken on any field survey within the Grand Canyon presents unique problems to the investigator which must be considered. These include:

1. The daily schedule for the research trips while on the river is generally a compromise between the needs of different investigators.

2. The research trips are usually of at least a week's duration, with only one contact (Phantom Ranch) for possible resupply or sample shipment. If anything goes wrong on the river--the power source proves unreliable, the dilution water becomes contaminated, the equipment provided is inadequate, etc.--the isolation of the boating party in the Grand Canyon magnifies the problem.

3. The nature of the transportation (rafts) limits the space available for, and the sensitivity of, equipment, unless prohibitively-expensive special apparatus is considered.

4. Samples cannot be shipped out of the Canyon within the 6-maximum delay [43] between sampling and analysis unless expensive helicopter transport is used. Therefore, analyes must be performed in the field. Often, late camps, rising water, rain, wind and blowing sand may make it difficult for the analysts to maintain quality control and sterile conditions.

Selection of bacteriological method of analysis. Two methods are available for the enumeration of microorganisms. These are the membrane filter (MF) technique and the multiple-tube fermentation or "Most Probable Number (MPN)" test.

The MPN method generally does not lend itself to field analyses, especially under the conditions imposed by the locale of this research. The logical choice is the MF method which has found wide acceptance for field measurements.

Although the MF procedure has the advantage in terms of equipment and time necessary to carry out an analysis, the accuracy of this method is prohibitively limited by turbidity and excessive numbers of interfering organisms [43]. Interfering organisms may be controlled somewhat by the use of freshly prepared rosalic acid [45], but there is no practical method of removing turbidity without also removing an unpredictable number of organisms. Other investigators have experimented with prefiltering after decanting supernatant from settled turbid samples. Geldreich et al. [42] found that a 20-80% loss of organisms results from such treatment.

An attempt was made by the authors to determine the effects of pretreating to remove the turbidity from samples composed of 53 to 105 g of beach sand and 95 ml of buffered dilution water dosed with fecal coliforms to given an initial density of 4.4 x 10^9 FC/100 ml. The samples were shaken for 1 min., then settled for 1 min., and diluted 1:20. This procedure was repeated and then 25 ml of the 1:400 dilution was prefiltered through 12 µm membrane filters. This was the procedure used by Knudson [4]. The filtered and unfiltered, settled portions were analyzed using the membrane filter technique [43]. The results of this study are given in Table XII.

The results would indicate that allowing the sand to settle and analyzing the supernatant results in a loss of organisms as shown by the 5 to 18% decrease in the density of FC in the unfiltered portions with increasing amounts of sand added. Prefiltering results in an additional loss of 5 to 19% as shown by the ratios of FC/ml in unfiltered to FC/ml in filtered portions, F/UF.

Table XII

Effect of Sample Prefiltration on Fecal Coliform Recovery (Initial Density (ID = 4.4×10^9 FC/100 ml)

		Fecal Coliforms/100 ml (in billions)			
Beach	Sand Added (g)	Unfiltered (UF)	Filtered (F)	F/UF	F/ID
After Lava Falls	53	4.4	4.2	0.95	0.95
Hermit	78	4.3	3.7	0.86	0.84
Hance	101	3.8	3.5	0.92	0.80
Lower Havasu	105	3.8	3.1	0.82	0.70
Deer Creek	106	3.7	3.4	0.92	0.77

The effects of turbidity on determinations of low densities of fecal coliforms (<200-800 FC/g) were very unpredictable. In parallel analyses of samples by the "Most Probable Number (MPN)" five-tube dilution method and by the membrane filter technique, the membrane filter plates were consistently masked by sediment. Colonies were rarely those of typical fecal coliforms. For example, in one case where there should have been approximately 60 colonies (according to the density determined by the MPN method), about 1/3 of the grid squares (randomly scattered) on the filter were blue and no real growth was observed. In other cases, there was discrete colony formation, but they seemed only faintly blue or yellow. In short, an excessive amount of personal judgement was required in counting the plates. For these reasons it was decided that the MPN 5-tube dilution method would be used in this investigation.

Effect of prolonged sample storage. As the MPN method could only be carried out in the laboratory, the samples would have to be stored on ice for the duration of the trip (1-12 days). It was necessary to ascertain that storage in an ice-chest did not result in an appreciable reduction in the number of organisms. In one study, 6 samples were analyzed for fecal coliforms by the MPN method after 0, 10 and 20 days of storage at 4°C. The MPN and 95% confidence limits are shown in Table XIII.

Table XIII

Effect of Beach Sand Sample Storage at 4°C on Fecal Coliforms

Beach	Time (Days)	FC/g (dry Weight)		
		Lower Limit	MPN	Upper Limit
Lava Canyon	0	.126	.377	1.06
	10	.126	.377	1.06
	20	.052	.180	.480
Papago	0	2.65	7.97	20.7
	10	11.8	>23.5	-
	20	2.62	7.88	20.4
Lower Bass	0	.119	.358	1.01
	10	1.95	5.88	15.2
	20	.0108	.076	.184
Deer Creek Overhang	0	946	>1891	-
	10	41.3	154	357
	20	294	1290	4200
Lower Havasu	0	.337	1.06	2.52
	10	.593	2.36	6.78
	20	.388	1.30	3.03
Mile 173.5	0	.0795	.261	.795
	10	-	<.227	.795
	20	-	<.027	.204

To reduce the effects of vagaries in the MPN method, six replicate fecal coliform analyses were performed at 0 and 14 days on a sample taken at Deer Creek Overhang and stored in the laboratory at 4°C. The results are summarized in Table XIV.

Since, the death-rate of the organisms is almost negligible at 4°C, it was determined that the delay between sampling and processing (providing the samples were kept iced) would not significantly effect the results of this investigation; and the cores could be transported back to the laboratory for bacteriological analysis by the preferred MPN method.

Table XIV

Effect of Sample Storage at 4°C on Fecal Coliform Content of Deer Creek Overhan Sample

	TC/g (dry weight)		FC/g (dry weight)	
Days	Ave. MPN	Standard Dev.	Ave. MPN	Standard Dev.
0	87	26	4.8	1.5
14	79	10	4.4	1.3

Results of Field Study

The investigation initiated in June 1976 by the National Park Service [4] involved analyses of die-off rates for chemically treated dumps, dumps with liquid and feces only (no disinfectants), and dumps with liquid and chemicals only. The latter were established by straining the contents of a chemically-treated holding toilet to separate the larger fecal solids. Liquid included the fecal solids passing the strainer and urine, as well as the river water charged to the toilet. The dumps were established by National Park Service personnel according to the same procedure used in September 1975. Sampling procedures were also the same, with one core collected at each site in June, July, August and September. June samples were taken at the sites before dumps were established to determine background contamination, but not immediately after waste disposal. Hence, the initial densities of organisms were again not established. The monitoring of four of the September 1975, sites was continued during this period.

The authors entered into this investigation in July and initiated the use of the MPN procedure to determine the densities of total and fecal coliforms. In July, fecal streptococci were also determined. In August and September, the Chemical Oxygen Demand (COD) of the samples was determined as an index of the self-purification capability of the beaches.

Table XV shows the coliform densities and COD values for the dumps sampled during July, August, and September 1975, according to the type of dump. The initial values for these sites were unavailable but, for fecal coliforms, have been estimated as approximately 800,000 per gram dry soil. By July, however, the fecal coliform densities were less than 14/g in all of the sites established in June excepting that at Deer Creek Overhang. This was stated to have been a "larger than average-sized" burial treated with Lysol disinfectant.

Table XV

Coliform and COD Content of Beach Disposal Sites
According to Dump Type

Beach Mile-Side	Month Sampled (1976)	Temp. °C Top-Bottom	Moisture %	Coliforms per Gram Total	Fecal	COD mg/g
	<u>Feces - Liquid - Chemical</u> Established Sept. 1975.					
LAVA	July	33-32	6.3	17.5	0.54	-
65.5-R	Aug.	32-29	6.8	18.3	0.38	3.07
	Sept.	31-29	7.9	0.36	<0.03	1.86
PAPAGO	July	37-31	4.2	3.58	0.81	-
76-L	Aug.	29-31	3.9	0.18	<0.02	0.70
	Sept.	29-29	10.3	>60.5	0.50	0.68
173.5	July	42-36	2.0	0.21	<0.21	-
173.5-L	Aug.	33-34	7.9	>181.6	0.26	0.44
	Sept.	28-31	16.3	>922.	2.77	0.19
	Established June, 1976					
CRASH	July	32-34	4.2	70.8	13.1	-
CANYON	Aug.	33-33	1.8	1.78	<0.02	1.70
62.5-R	Sept.	36-33	3.5	9.43	<0.03	1.23
DR.CK.	July	34-33	15.6	>20860.	>20860.	-
OVERHANG	Aug.	36-33	5.5	>946.	>946	3.01
137-L	Sept.	30-30	2.9	1280.	7.48	1.07
BELOW	July	38-37	6.0	16.5	2.97	-
PUMPKIN	Aug.	40-38	6.55	1.15	<0.03	2.5
214-R	Sept.	-	-	-	-	-

Table XV--continued

	Feces - Liquid Only Established June, 1976					
INDIAN	July	-	-	-	-	-
DICK	Aug.	33-32	7.1	16.4	<0.88	2.20
28-L	Sept.	29-31	2.5	5.72	<0.02	1.94
PRES.	July	33-29	5.3	37.7	2.22	-
HARDING	Aug.	28-30	16.3	0.33	<0.03	5.66
44-L	Sept.	28-29	9.4	4.95	<0.05	0.99
LOWER	July	38-38	7.4	1.38	<0.26	-
BASS	Aug.	33-34	4.0	>17.4	0.36	4.13
108-R	Sept.	37-33	2.5	<0.03	<0.02	0.92
LOWER	July	34-34	4.1	>16.7	0.98	-
HAVASU	Aug.	39-34	16.1	73.	1.06	0.45
158-R	Sept	38-33	1.6	4.05	1.00	0.33
	Liquid - Chemical Only Established June, 1976					
BADGER	July	43-38	5.7	261.	5.33	-
8-L	Aug.	34-31	5.6	16.5	<0.16	1.07
	Sept.	31-30	.63	5.18	<0.02	1.03
LOWER	July	35-36	8.6	1.28	<0.05	-
GARDEN CK.	Aug.	40-40	1.5	0.11	<0.02	0.51
89-R	Sept.	37-33	9.4	8.89	<0.03	0.45
2nd B.	July	33-33	4.6	22.6	1.78	-
AFTER	Aug.	36-34	3.3	>90.6	>88.	1.4
LAVA	Sept.	29-29	13.1	106.	1.28	0.48
183-R						

Without the initial concentration of organisms, actual rates of die-off cannot be calculated; however, if one uses the value of 800,000 FC/g as previously estimated the decrease to 14 FC/g in one month would give a 99.998% reduction in numbers. In subsequent months there is a tail-off in the densities with smaller reductions occurring as the more resistant organisms survive. This two-phase death rate for a population has been shown by Frost and Streeter [45] and others. By September, the fecal coliform densities were reduced to values comparable to those in sites established in September of the previous year. The high death rates occurring during the summer are consistent with reported literature values for soil temperatures averaging about 35°C (95°F). During the colder months, death rates are reduced and few organisms are carried over into the following season.

Figure 2 shows the temperature variations occurring during the year for cores taken at beach disposal sites. The bottom temperatures were taken approximately 2 ft from the surface and the surface temperatures were about 4 in. from the surface.

The total coliform group shows a much lower death rate in general than fecal coliforms. The non-fecal fraction of this group includes a number of naturally occurring genera and species and, hence, has little sanitary significance. The relationship between fecal and total coliforms in July for the June 1976 dumps is shown in Figure 3. The ratio for the month is roughly one fecal coliform to 16 total coliforms or 15 non-fecal coliforms. In August and September, this relationship is not in evidence. However, the probable occurrence of fecal coliforms in low densities could be predicted for samples with high total coliform levels (>100 TC/g).

In July, the fecal streptococci were included in the analysis; however, the individual values are not presented here. In general the values were 100 to 1000 times greater than for the fecal coliforms, indicating a greater survival since the FC/FS ratio for human feces is about 4/1 [37]. It has been shown by Smith et al. [46] that the survival characteristics of salmonella more closely resemble those of fecal coliforms than those of fecal streptococci. This conclusion is supported by others for salmonella and shigella [41,47]. Therefore, determinations of fecal streptococci densities were not carried out after July.

The effect of chemical disinfectants is not apparent in this field study. The role of the chemicals used in holding toilets is to suppress decomposition orders. While chemicals may adequately disinfect the liquid portion of the waste, it is unlikely that they penetrate to the interior of clumps of cells or fecal matter. If the disinfectant in the three dumps established in June did significantly reduce the initial density of organisms, assuming similar death-rates, the fecal coliform densities in those dumps in July should be lower than in the dumps which

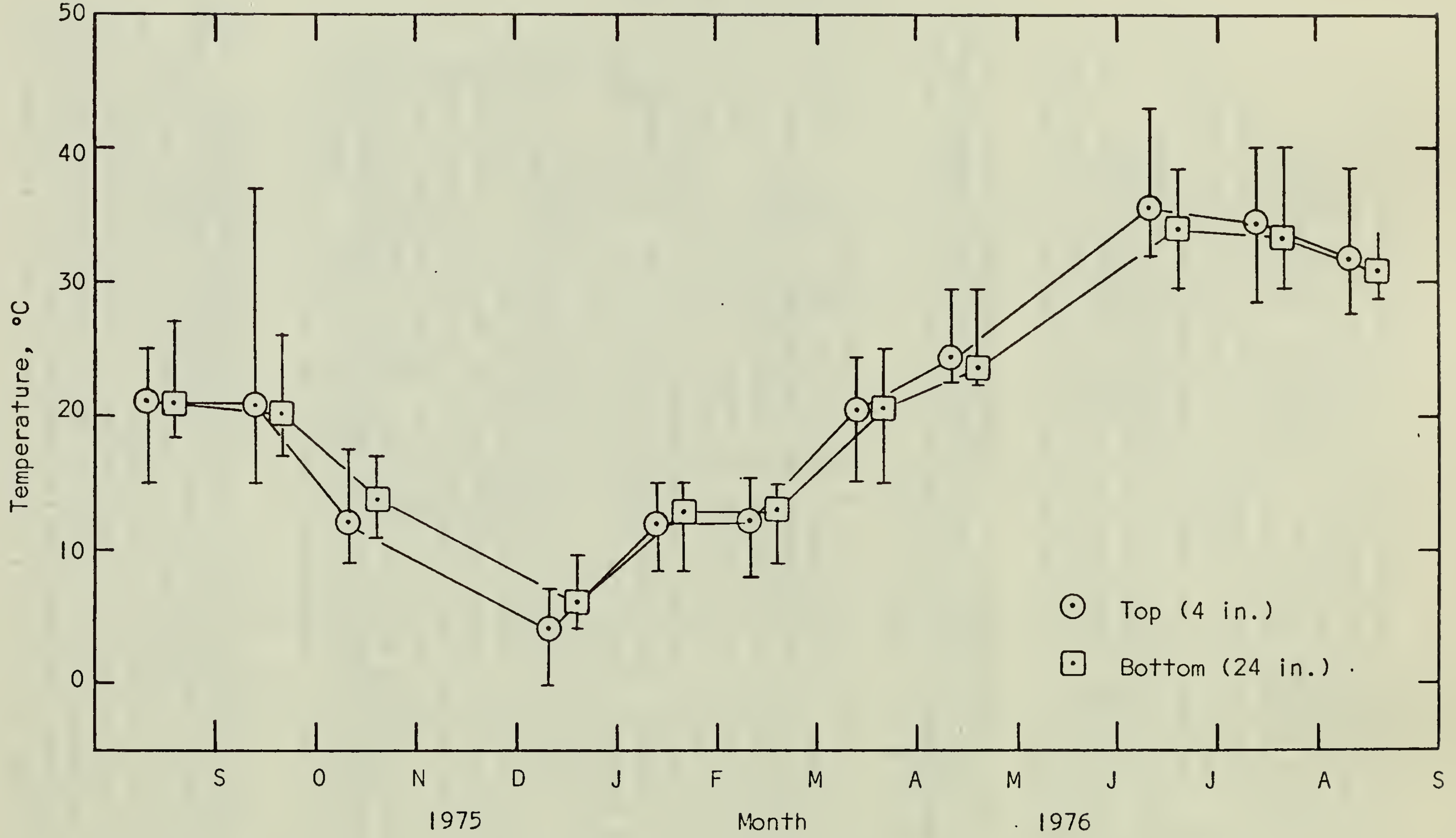


Figure 2. Average Temperature Of Cores Taken At Waste Disposal Dumps (Sept., 1975 to May, 1976, data from Knudson [4])

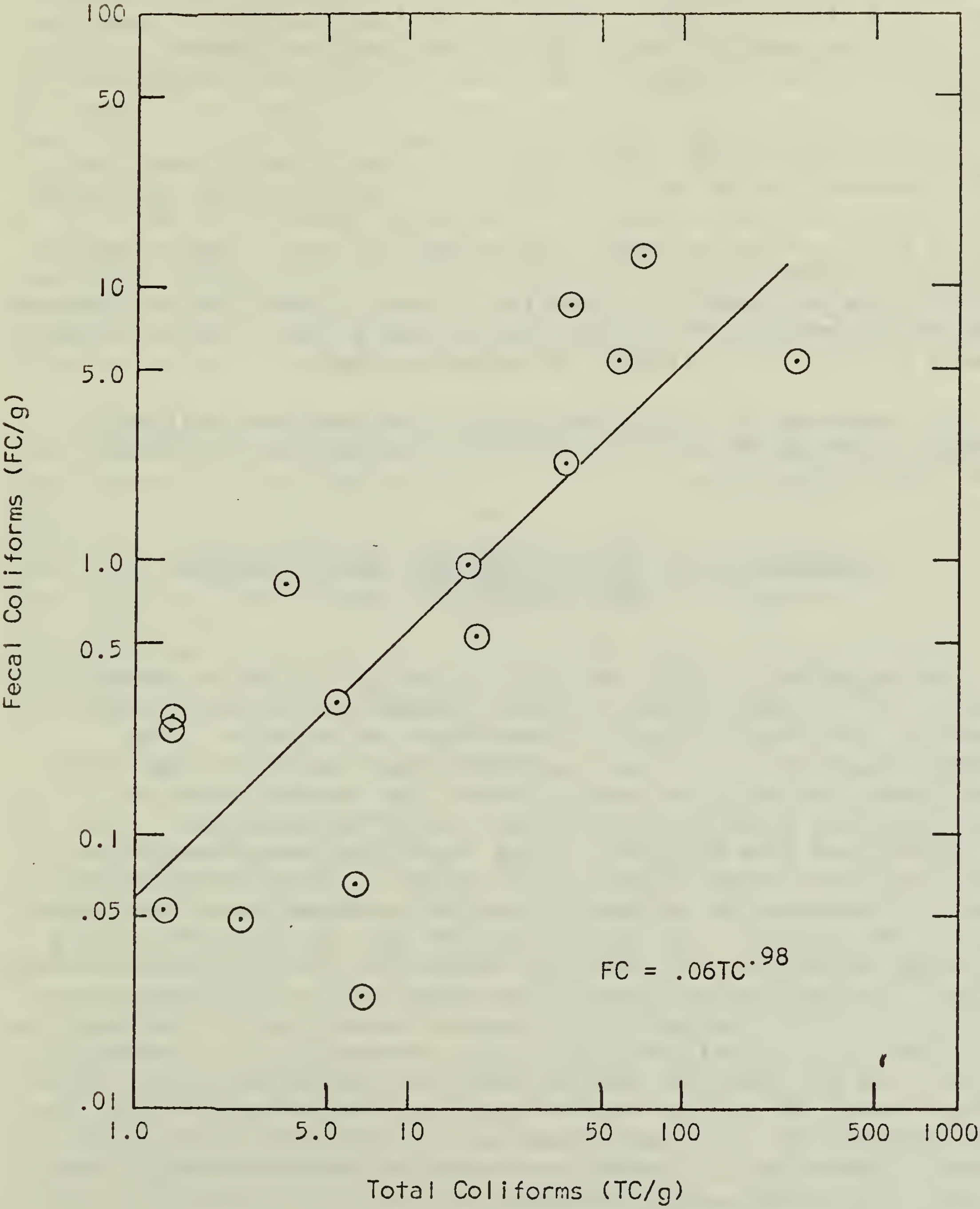


Figure 3. Fecal versus Total Coliforms In Month-Old Dump Sites

were not chemically treated. In fact, on the average, the chemically treated dumps had higher numbers of total and fecal coliforms in July. This might indicate utilization of the disinfectants as a food source or suppression of competing organisms and, consequently, a slow death rate. More likely, however, larger quantities of waste were used in setting up the chemical dumps than for those without chemicals. It is documented that the dump at Deer Creek Overhang was larger than average.

The COD content was determined for 13 dump sites in August and 12 in September and is shown in Table XV. The initial COD content at the sites is not known; however, it can be estimated to be about 25 mg/g of soil. After two months the average COD was 2.3 mg/g, showing about a 90% destruction of the organic content. By September the average COD value was about 0.94 mg/g for a total of about 96% destruction. These can be compared to a value after almost a year of about 0.9 mg/g, showing rapid stabilization of the organic content.

The presence of disinfectants did not affect the stabilization process, although the exclusion of solid fecal matter did result in lower final COD values.

Death Rates of Enteric Organisms Under Simulated Beach Conditions

The objective of this laboratory investigation was to establish numerically the rate of death of enteric organisms in fecal matter spread in sand as a function of temperature and moisture. Such information, hopefully, would supplement the scarcity of data in the field study, and might be used to predict the survival of enteric organisms under a variety of burial and climatic conditions.

Four dumps were simulated in the laboratory using homogenized concentrated raw sewage sludge (10.2% solids). One-hundred and fifty grams of sludge was spread evenly over 500 g of Lees Ferry beach sand in a 1-liter beaker. The sludge layer was then covered with a 500 g sand layer to give a total depth of about 4 in. In two of the samples, the sand was saturated with distilled water to give a 20% moisture. The initial moisture of the other two samples was 11.7%. These samples are referred to as "wet" and "dry" dumps, respectively. An initial 0.5 in. core was taken for analysis and the hole filled with an empty culture tube. One wet and one dry sample was maintained at 20°C; the other two at 3.5°C. The wet samples were covered with plastic film to prevent moisture loss. Samples were cored at intervals over 21 days and moisture content, total and fecal coliforms and 20 and 35°C standard plate counts (SPC) determined.

The moisture content of the wet samples was maintained at saturation. Figure 4 shows the gradual moisture loss in the dry samples. In both dry dumps, the sludge layer rapidly shrank to a fibrous cake. After 79 days, a follow-up analysis showed that, while the overall core moisture content in the dry samples at 20 and 35°C was 0.2%, the moisture content of the organic layer itself was 2.5% at 20°C and 2.3% at 35°C. This demonstrates the moisture retaining capacity of organic matter in dry sand.

The results of the bacteriological analyses are presented in Table XVI. That a large proportion of the total coliform population is fecal in origin is shown by the similarity in the data shown in Figures 5 and 6. The 20 and 35°C SPC (Figures 7 and 8) reflect the dynamics of the total population in the dump.

The only simulated dump which showed a rate of death of fecal coliforms of the same magnitude as that observed in the field study was that at 35°C and 20% moisture. From an initial density of 10,000,000/g (dry weight) the numbers dropped to less than 3/g in 21 days. The total coliform group showed similar reductions. The total population of the 35°C wet dump decreased initially in parallel with the decrease in coliforms; but, as the 35°C SPC (Figure 8) shows, a non-fecal group of organisms rapidly became predominant. The set of circumstances producing this rapid kill of fecal coliforms (namely saturation of the dump in conjunction with high temperatures) would only rarely occur on the beaches of the Colorado River.

In the other three simulated dumps, the die-off of fecal coliforms was either slow or (99% in 12.6 days for the 20°C wet dump) or near zero in the case of the dry dumps. These results indicate that if conditions remain aerobic and competing populations of non-fecal organisms do not develop, enteric organisms may survive in fecal matter in sand for extended periods at constant temperatures of 20 to 35°C. Ostensibly, the fecal matter may provide a source of nutrients and retain moisture which protects the enteric organisms from desiccation. Fecal coliforms cannot survive indefinitely, however. After 122 days, the FC population in the 20°C wet dump was 1.4 FC/g. The fecal coliform levels after 79 days were 768 and 2.01/g in the 20 and 35°C dry simulated dumps, respectively (a 99.99% reduction).

While very little data on the survival of fecal coliforms in feces in soil are available from the literature, Jordan [48] found that survival of E. coli in feces was extended when the feces were covered with garden soil or sand, rather than stored. The addition of 5 to 20% raw sewage sludge in the study by Mallman and Litsky [28] resulted in survivals of 10% of the coliforms after 5 to 6 weeks. Dazzo et al. [29] found that soil at 10.2% moisture previously receiving 5.1 cm/week of manure slurry had only 90% reduction in fecal

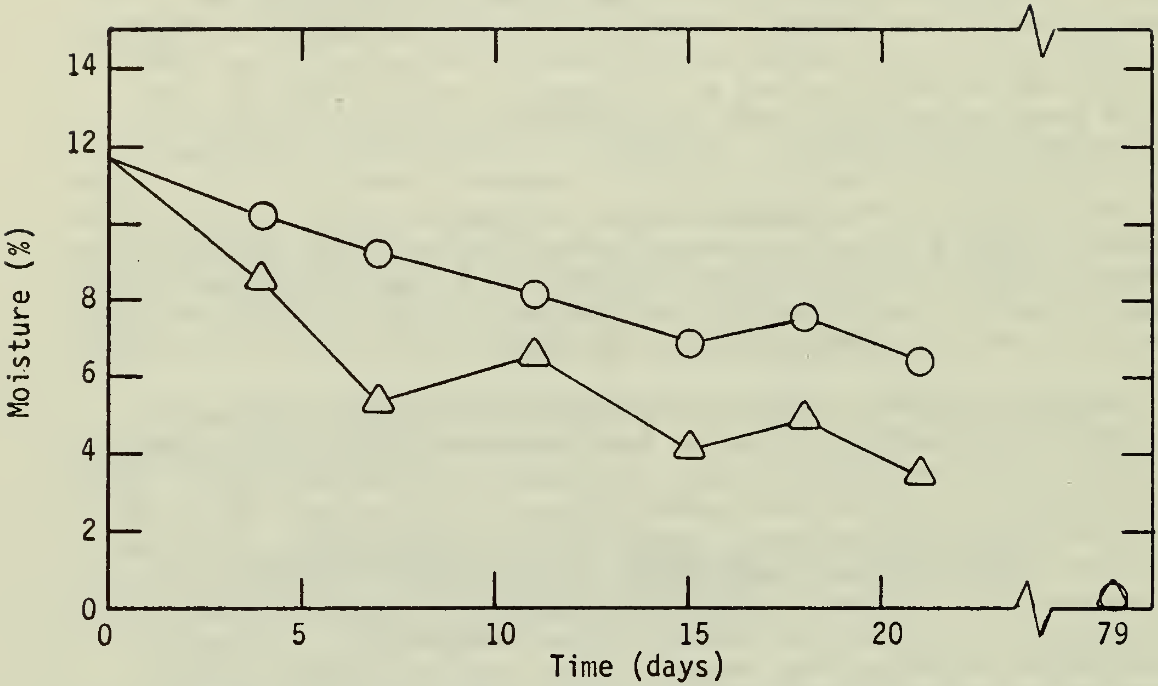


Figure 4. Moisture Loss of Uncovered "Dry" Simulated Dumps at 20°C (○) and 35°C (△)

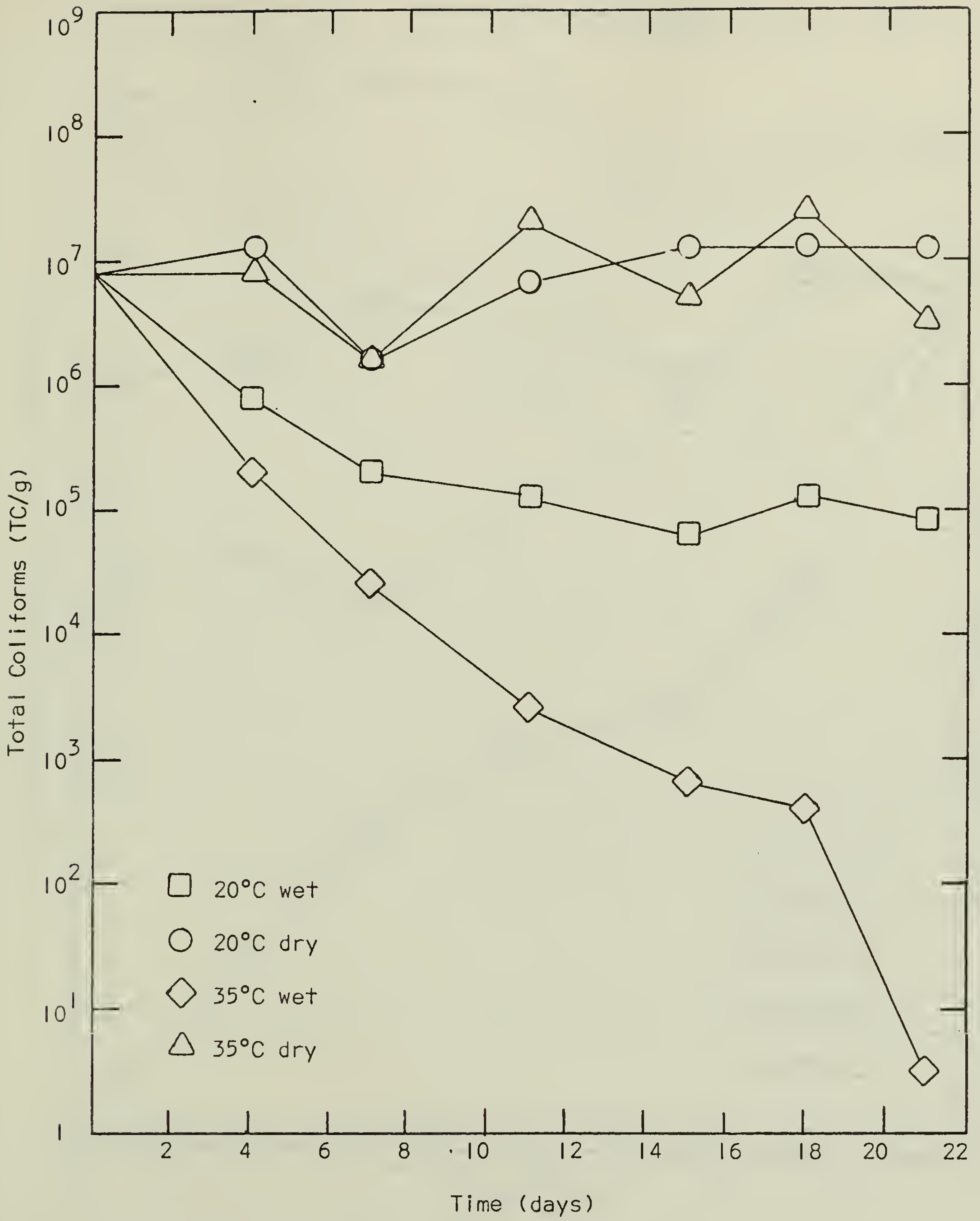


Figure 5. Total Coliforms in Simulated Dumps vs. Time

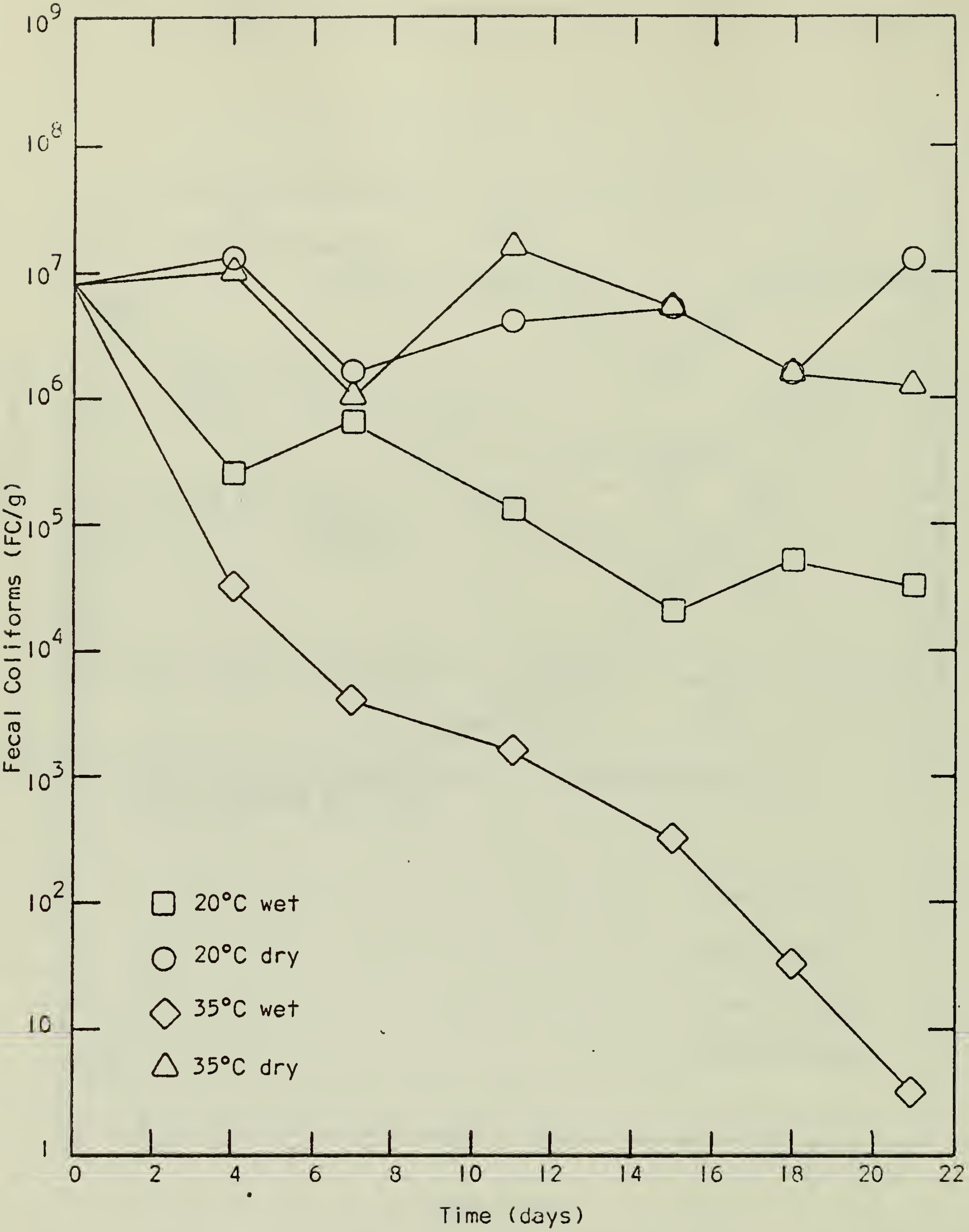


Figure 6. Fecal Coliforms in Simulated Dumps vs. Time

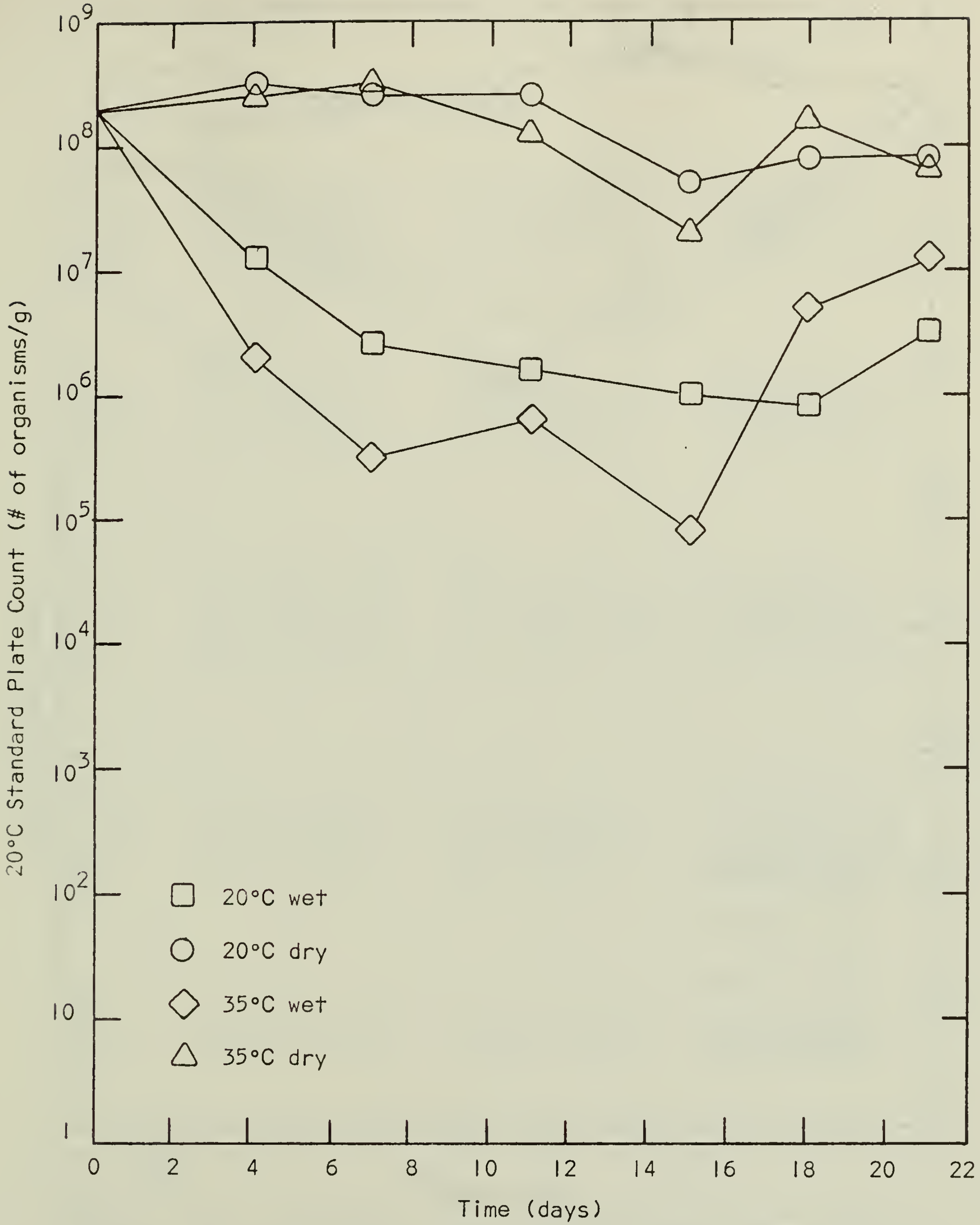


Figure 7. Standard Plate Count (20°C) Of Simulated Dumps vs Time

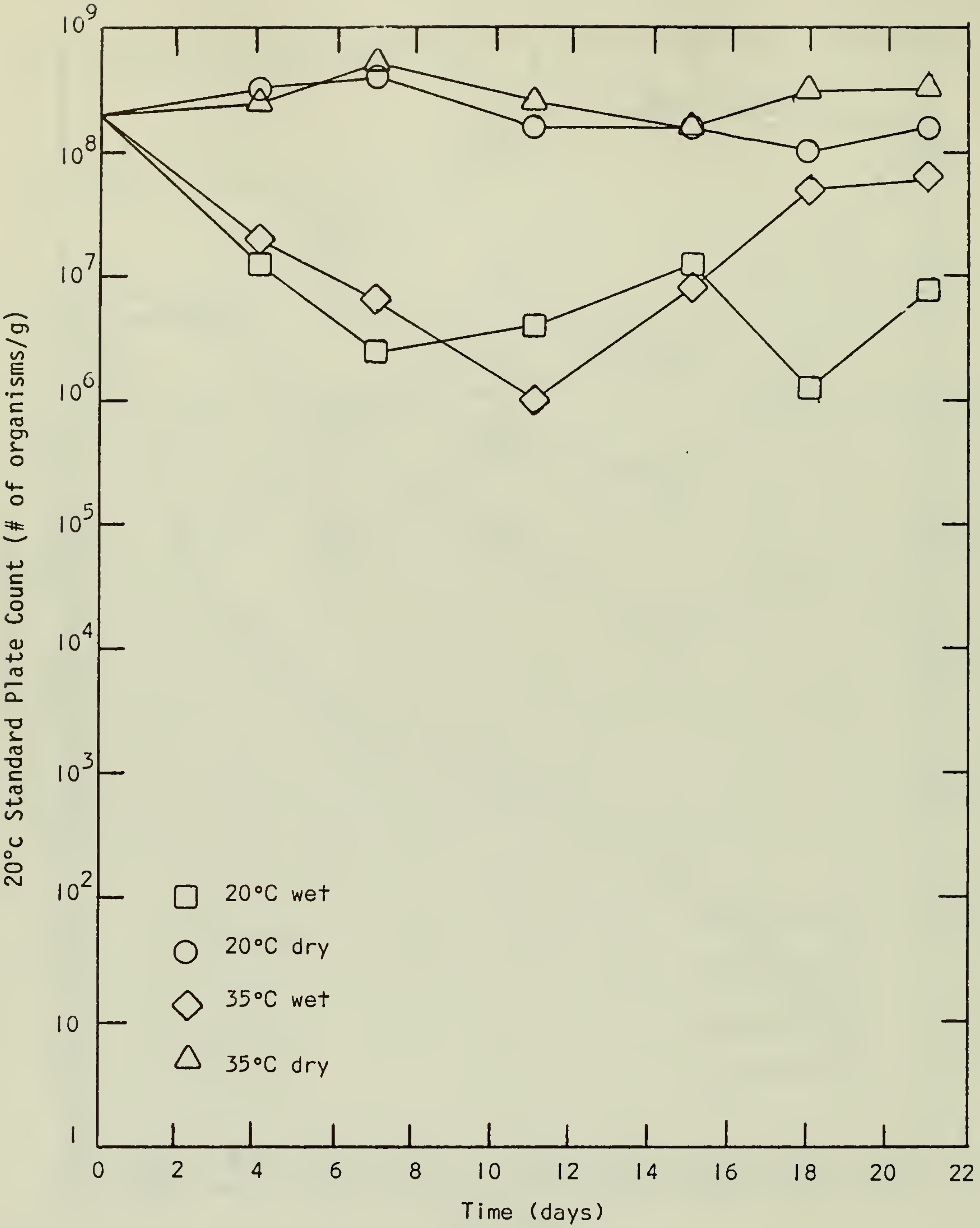


Figure 8. Standard Plate Count (35°C) In Simulated Dumps vs Time

Table XVI

Simulated Dumps Data

Time (days)	Moisture (%)	Total Colif. (TC/g)	Fecal Colif. (FC/g)	SPC(20^oC) (#/g)	SPC(35^oC) (#/g)
20^oC - WET					
0	20.0	8.0×10^6	8.0×10^6	2.2×10^8	2.0×10^8
4	20.3	7.1×10^5	2.7×10^5	1.2×10^7	1.3×10^7
7	19.8	$>2.0 \times 10^5$	6.7×10^5	2.6×10^6	2.4×10^6
11	19.4	1.3×10^5	1.3×10^5	1.5×10^6	4.1×10^6
15	18.3	5.9×10^4	2.2×10^4	9.3×10^5	1.3×10^7
18	18.9	1.4×10^5	5.2×10^4	8.0×10^5	1.3×10^6
21	17.5	7.5×10^4	3.3×10^4	3.0×10^6	9.0×10^6
122	17.0		1.4×10^0		
20^oC - DRY					
0	11.7	8.0×10^6	8.0×10^6	2.2×10^8	2.0×10^8
4	10.2	1.3×10^7	1.3×10^7	3.1×10^8	3.8×10^8
7	9.2	$>1.7 \times 10^6$	$>1.7 \times 10^6$	2.6×10^8	3.6×10^8
11	8.1	6.2×10^6	4.0×10^6	2.7×10^8	1.6×10^8
15	6.8	1.3×10^7	4.8×10^6	5.5×10^7	1.8×10^8
18	7.5	1.2×10^7	1.7×10^6	8.2×10^7	1.1×10^8
21	6.4	1.3×10^7	1.3×10^7	7.1×10^7	1.5×10^8
79	.2		7.6×10^2		
35^oC -WET					
0	20.0	8.0×10^6	8.0×10^6	2.2×10^8	2.0×10^8
4	19.3	2.0×10^5	7.5×10^4	2.0×10^6	2.1×10^7
7	22.2	2.8×10^4	4.4×10^3	3.4×10^5	6.9×10^6
11	18.9	2.5×10^3	1.6×10^3	7.0×10^5	1.1×10^6
15	21.5	6.0×10^2	3.0×10^2	7.3×10^4	8.1×10^6
18	19.1	3.8×10^2	3.2×10^1	$>4.8 \times 10^6$	4.9×10^7
21	17.7	$<3.0 \times 10^0$	$<3.0 \times 10^0$	1.3×10^7	6.1×10^7

Table XVI--continued

$35^{0}C$ - DRY					
0	11.7	$8.0x10^{6}$	$8.0x10^{6}$	$2.2x10^{8}$	$2.0x10^{8}$
4	8.5	$9.2x10^{6}$	$9.2x10^{6}$	$2.8x10^{8}$	$2.5x10^{8}$
7	5.3	$>1.7x10^{6}$	$9.7x10^{5}$	$3.1x10^{8}$	$4.9x10^{8}$
11	6.6	$>1.8x10^{7}$	$>1.7x10^{7}$	$1.4x10^{8}$	$2.3x10^{8}$
15	4.1	$4.9x10^{6}$	$4.9x10^{6}$	$2.2x10^{7}$	$1.6x10^{8}$
18	4.9	$2.3x10^{7}$	$1.6x10^{6}$	$1.5x10^{8}$	$3.0x10^{8}$
21	3.4	$3.5x10^{6}$	$1.3x10^{6}$	$6.9x10^{7}$	$3.5x10^{8}$
79	.2		$2.0x10^{0}$		

coliforms in 8.5 days. These results are comparable with those found in this laboratory investigation.

As average temperature and moisture conditions for the sites in the June 1975 field study are most similar to those of the dry simulated cores, it would seem that the rapid die-off observed in the field study is in contradiction to the results of this laboratory study. The difference in death rates may be partially explained in terms of the variation in temperature and moisture extremes in nature, but it may also be due to the physical differences between the simulated dumps and the field study dumps.

In the field study, the contents of a holding toilet were used in setting up the dumps. The fecal matter, in this case, after sitting in the toilet and being mixed and strained by the flushing mechanism, no longer retains its initial solidity. When a dumps is established, the few intact feces are usually the less dense "floater," and they end up near the surface where they are subject to the most extreme heating and drying conditions. In contrast is the consolidated layer of organic matter in the simulated dump which by it's density protects the enteric organisms from the hostile soil environment.

The simulated dumps duplicate more closely the second major method of consolidation and burial used on the river where a portable toilet seat is set over a dry hole. Liquid readily filters into the sand and the solids form an intact layer in the bottom of the hole. While the changes and extremes in temperature and moisture would increase the death rate, the simulated cores indicate that the survival of organisms in these dumps may be longer than in the holding type toilet which breaks up the feces.

CHAPTER 4

CONTAMINANT LEVELS ON BEACHES

To assess the general level of beach contamination, a total of 52 beach samples were collected in July, August and September 1976. Included in August were three samples taken at small beaches at miles 221.5, 223 and 22.5 for background data. Stops by boat parties at these three locations would be unlikely.

Sampling was done by scraping away approximately 1 in. of surface sand, taking the temperature, and filling a sterile Whirl-pak plastic bag with sand. The samples were analyzed in the laboratory for moisture and total and fecal coliforms. Fecal streptococci densities were also determined in July, and COD analyses were carried out in August and September. The results of these analyses are presented by month in Tables XVII, XVIII and XIX.

During the three months of field study, the temperature and moisture conditions on the beaches were generally hot and dry. Of 52 samples, 29 had moistures of less than 0.5%, and 11 were less than 0.05%. Sand temperatures were as high as 59°C (138°F), with 55% of the samples having temperatures greater than 35°C (95°F).

The total number of samples and the number of samples with fecal coliforms greater than approximately 0.02 FC/g for various ranges of moisture and temperatures are shown in Table XX. The proportion of samples positive for fecal coliforms increases (up to 63%) as the moisture content increases and temperature decreases. That the low moisture-high temperature conditions on the beaches result in low fecal coliform densities is also shown by the fact that only five of the samples had greater than 1 FC/g (>1.21, >1.17, 5.41, 1.95 and 1.32 FC/g). All of the three control beaches showed fecal coliforms to be below the minimum detectable density of <0.02 FC/g.

Although survival of fecal coliforms on the surface of the beaches is limited in the summer months, a high proportion of the samples (32%) showed the presence of fecal coliforms. This may be significant in that the occurrence of fecal coliforms in the soil is directly related to human or animal pollution, and because under cooler and moister weather conditions (spring and fall), concentrations of enteric organisms would remain at high levels for a longer period.

Table XVII

Bacterial Contamination of Beaches, July, 1976

Beach	Moisture (%)	Temp. (^{0}C)	Bacterial Densities, MPN/g soil Total Coliform	 Fecal Coliform	 Fecal Strep	Exposure
Badger Ck.	9.5	50	0.97	<0.03	>1.57	
Pres. Harding	0.0	40	<0.03	<0.03	0.09	
Crash Canyon	0.1	42	0.74	<0.03	0.52	sun
Lava Canyon	0.0	35	>1.26	0.77	>1.26	
Papago Beach	0.0	47	<0.02	<0.02	0.09	
Hance	0.1	30	>1.21	<0.03	0.74	shade
Monument-Granite	8.0	29	>1.48	<0.03	>1.48	
Hermit (Upstream)	0.3	32	0.51	<0.03	1.19	
Lower Bass	4.0	43	0.81	0.07	0.23	
Deer Ck. Overhang	0.3	26	>1.21	>1.21	>1.21	shade
Old Canyon	0.0	38	0.50	<0.02	>1.16	
Lower Havasu	3.7	36	0.12	<0.03	0.22	
Fern Glen	18.2*	42	0.55	<0.04	>1.77	shade
173.5 Mile	0.0	47	<0.02	<0.02	0.197	
Lava Falls	0.0	46	<0.02	<0.02	0.15	
2nd After Lava	0.2	28.5	<0.02	<0.02	0.12	
Pumpkin	0.0	47	<0.03	<0.02	0.21	
Upper Diamond	5.0	23	>1.17	1.17	>1.17	

* sample wetted in transport

Table XVIII

Bacterial Contamination of Beaches, August, 1976

Beach	Moisture (%)	Temp. (°C)	Bacterial Densities, MPN/g		COD (mg/g)	Exposure
			TC	FC		
Badger	18.4	35	1.11	<0.03	0.11	sun after rain
Indian Dick	0.4	36	0.08	<0.02	0.26	sun
Pres. Harding	21.5	34	0.21	<0.03	1.15	shade
Crash Canyon	1.8	33	>1600.	5.41	0.34	shade
Lava Canyon	1.6	44	244.	<0.02	0.19	partial shade
Papago	2.1	31	1.76	<0.02	0.08	sun
Hance	3.3	31	0.12	<0.02	0.16	shade
Garden Creek	0.9	43	1.32	0.05	0.30	
Granite 1 in.	0.0	38	34.	0.77	-	shade
3 in.	21.4*	38	523.	1.95	0.97	shade
Lower Bass	4.9	31	10.	0.25	0.92	shade
Lower Dr. Ck.	1.1	33	295.	0.07	0.18	shade
Lower Olo 5 in.	2.7	29	2.18	0.23	0.38	shade
Upper Olo 4 in.	2.4	30	0.11	<0.02	0.39	sun
Lower Havasu 1 in.	16.7*	59	1.69	0.64	0.09	sun
5 in.	6.6*	41	1.49	0.56	0.13	sun
173.5 Mile	0.3	31	1.53	0.02	0.09	shade
After Lava Falls	1.2	42	>1.57	<0.02	0.16	partial
Pumpkin	2.6	38	0.52	<0.02	0.72	sun
Control No. 1, 221.5 Mile	-	35	>1.55	<0.02	0.09	sun
Control No. 2, 223 Mile	-	35	>1.58	<0.02	0.04	sun
Control No. 3, 224.5 Mile	-	35	0.19	<0.02	0.10	sun

* sample wetted in transport

Table XIX

Bacterial Contamination of Beaches, Sept., 1976

Beach	Moisture (%)	Temp. (°C)	Bacterial Densities, MPN/g		COD (mg/g)	Exposure
			TC	FC		
Pipe Creek	0.2	38	>16.1	<0.02	0.27	sun
Lower Bass	0.1	31	>15.9	0.02	0.47	partial shade
Shinumo	0.2	34	9.24	<0.02	1.62	sun
After Fossil	0.3	31	>16.1	0.05	0.41	shade
Bass	0.1	29	<0.02	<0.02	0.20	partial shade
Havasu	0.1	38	<0.02	<0.02	0.07	sun
National	0.7	37	132.	1.32	0.34	shade
Fern Glen	0.2	36	0.68	<0.02	0.71	shade
173.5 Mile	0.1	26	<0.02	<0.02	0.07	sun
2nd After Lava	0.3	24	9.12	<0.02	0.25	sun
238 Mile	0.1	37	0.02	<0.02	0.27	sun
Scorpion Is.	0.1	41	9.21	<0.02	0.37	shade

Table XX

Total Number and Number of Samples Positive for Fecal Coliforms Under Various Moisture and Temperature Conditions

	Moisture (%)					
	< 0.05		0.05 - 0.45		0.50 - 5.0	
Temp., °C	No. Samples	Pos. FC	No. Samples	Pos. FC	No. Samples	Pos. FC
< 35	11	2	11	4	8	5
> 35	0	0	7	0	7	3

Geldreich [37] has shown that non-fecal coliforms are present in most soils and on vegetation even when there is no indication of recent contamination. Therefore, it is not unexpected that the results of the beach study indicate that 83% of the samples were positive for coliform bacteria, but that no numerical ratio of fecal to total coliforms exists. However, certain generalizations may be made concerning the occurrence of the two organisms. When fecal coliforms are present, the total coliform population usually exceeds them by at least an order of magnitude. This is in contrast to the ratio in actual fecal matter where 96% of the total coliform population are fecal coliforms [37]. Also, for the five samples with levels of total coliforms greater than 100/g, fecal coliforms were detectable (>.02 FC/g) in four samples and were greater than 1 FC/g in three samples. These results may be attributed to the multilication of the soil non-fecal coliforms when nutrients associated with fecal contamination are added to the soil, as documented by Van Donsel et al. [41].

In contrast to the fecal coliform results, in July, 100% of the beach samples had fecal streptococci present. Several factors indicate that fecal streptococci levels are not valid indicators of the occurrence and duration of beach contamination in this case. While it has been shown that the occurrence of fecal coliforms may be strictly related to the fecal wastes from warm-blooded animals, there are several sources and species of fecal streptococci which are probably of no sanitary significance. Geldreich et al. [49] found large numbers of fecal streptococci on six orders of insects which spent part of their life cycle in contact with fecal matter. Also, the ratio of fecal streptococci to fecal coliforms for humans is less than one, while the same ratio for warm-blooded animals (including birds) is almost always greater than one [37]. Lastly, as has been stated before the survival of salmonella and shigella most closely parallels that of fecal coliforms rather than the extended survival of fecal streptococci. For these reasons, the analysis of fecal streptococci in beach samples was not carried out after July. The high background level of fecal streptococci indicated by this general beach contamination study was also a decisive factor in the termination of fecal streptococci analyses in the dump sites.

The COD of the beach samples ranged from 0.06 to 1.6 (average, 0.35) mg/g of dry sand. Three samples obtained from the isolated control beaches had COD contents of 0.04, 0.08 and 0.10 mg/g of dry sand. Although there is no obvious correlation between COD and the occurrence of total or fecal coliforms, the COD level may be regarded as an indicator of the general organic content of the sand. In comparison to the average background level of COD, 0.07 mg/g, and the average level for general beach samples, 0.35 mg/g, the average COD for a one-month old dump including feces was 2.8 mg/g and that for a one-year old dump was 0.91 mg/g.

CHAPTER 5

COMPARISON OF BURIAL EFFICIENCY FOR CURRENT CONSOLIDATED DISPOSAL METHODS

The probability of a river-runner coming in contact with pathogenic organisms associated with human body wastes must be directly related to the frequency of the occurrence of fecal matter on or near the surface of the beaches. For this reason, the efficiency of burial for the two major methods of consolidation and disposal currently being used must be a prime consideration.

The foremost difficulty with any "deep" burial is that it may be difficult in cohesionless beach sand to dig a 2-ft deep hole and to utilize that hole successfully. If a sufficiently deep hole is not dug, or if it collapses before or during the disposal of human wastes, then the situation deteriorates to shallow or nearly direct surface dumping.

Even if an acceptable hole can be dug, there are additional problems with the disposal of holding toilets. Including the initial charge of a holding toilet of approximately 2 gal and the wastes of an average river party, the volume of the contents of the holding toilet may fill a regulation size hole to within 6 in. or less of the surface. This would be acceptable if at least some of the liquid portion of the wastes would infiltrate into the sand, reducing the volume and lowering the waste level. However, this does not occur; and, as the hole is filled, the wastes (including low density floating fecal matter) are pushed up closer and closer to the surface. The end result may be a dump covered by only a thin layer of sand. Sometimes, if highly colored disinfectants are used in the holding toilets, blue- or aqua-dyed sand may warn succeeding river parties of a contaminated area for months afterward. However, the appearance of such dye in the beach sand does not constitute an aesthetically agreeable solution to this problem.

It has been suggested that the limited infiltration of the holding toilet contents is due to the clay content of the beach sand which, upon contact with water, swells to block the passage of fluids. To test this hypothesis, infiltration tests were carried out during the July field study.

Holes were dug approximately 2-ft deep by 1-ft diameter. The holes were then filled to a given level with river water and the drop in surface level with time was monitored. This process was repeated until a constant rate of infiltration was achieved.

The results of this study from Table XXI are shown graphically in Figure 9. The values for permeability were calculated according to Darcy's Law: Q/(AH) = K, were Q/A is the rate of infiltration in gal/min/sq ft, H is the pressure head in ft, and K is the permeability of the sand in gal/min/sq ft/ft of head. The average permeability of the beach sand is 0.50 gal/min/sq ft/ft. Such permeability is not indicative of a significant clay content for the beach sand. A mechanical analysis of the sand from the beach at mile 126 (Table XXII and Figure 10) reinforces this conclusion.

The analysis indicates that the sand is very uniform (this size uniformity should result in optimum infiltration) with only 1.2% by weight finer than 0.007 mm size uniformity should result in optimum filtration. Yet, when the contents of a chemically treated holding-toilet were emptied into the second infiltration study hole on the beach at mile 126, the level of the waste dropped less than 0.5 in. before infiltration stopped completely. In five additional minutes, no observable drop in the level of the dump occurred in the same hole where the rate of river water infiltration averaged 0.4/gal/min/sq ft It may be concluded that the sand was clogged, not because of its clay content, but because of the colloidal and organic materials suspended in the waste liquid itself.

This problem is not apparent in the dumps established by digging a hole and setting a toilet seat over it (dry-hole dumps). As the solid and liquid matter are not intimately mixed by any flushing mechanism, urine rapidly filters into the sand leaving an intact mass of solids in the bottom of the hole. Hence, the efficiency of burial for dry hole dumps is potentially much higher than that for holding-toilet burials.

Table XXI

Results of Infiltration Tests

Time (min)	Depth to Water (in)	Rate (gal/min/sq ft)	Head (ft)	Permeability (gal/min/sq ft/ft)
Nankoweap: 9-in. diam., 2-ft. depth				
0.00	15.00			
0.33	16.13	0.45	0.70	0.64
0.83	17.00	0.25	0.61	0.40
1.33	17.56	0.17	0.56	0.30
2.08	18.75	0.28	0.49	0.58
4.58	16.63			
5.08	17.13	0.15	0.59	0.25
Beach above Kaibab Bridge: 9.5-in. diam., 20.5-in. depth				
0.00	12.31			
0.50	13.25	0.27	0.64	0.42
1.00	13.88	0.20	0.57	0.35
1.50	14.63	0.26	0.52	0.49
2.00	15.19	0.20	0.46	0.44
2.50	15.75	0.22	0.42	0.53
3.00	16.13	0.15	0.38	0.41
4.00	18.88	0.18	0.33	0.53
Mile 126: 11-in. diam., 2-ft. depth				
0.00	10.25			
0.35	12.5	0.50	1.05	0.48
1.00	14.0	0.38	0.89	0.43
1.42	15.5	0.51	0.77	0.66
5.25	9.75			
5.50	11.0	0.52	1.14	0.46
5.75	12.0	0.45	1.04	0.43
6.00	12.75	0.36	0.96	0.37
6.25	13.5	0.39	0.88	0.44
6.50	14.25	0.41	0.84	0.49
Mile 126 (site #2): 10-in. diam., 27-in. depth				
0.00	16.5			
0.25	17.75	0.62	0.82	0.75
0.50	18.25	0.25	0.75	0.46
0.75	18.75	0.27	0.71	0.38
1.00	19.25	0.28	0.67	0.42
1.25	19.75	0.29	0.63	0.47
1.50	20.25	0.31	0.58	0.53
1.75	21.0	0.54	0.53	1.01
2.25	22.0	0.41	0.46	0.90

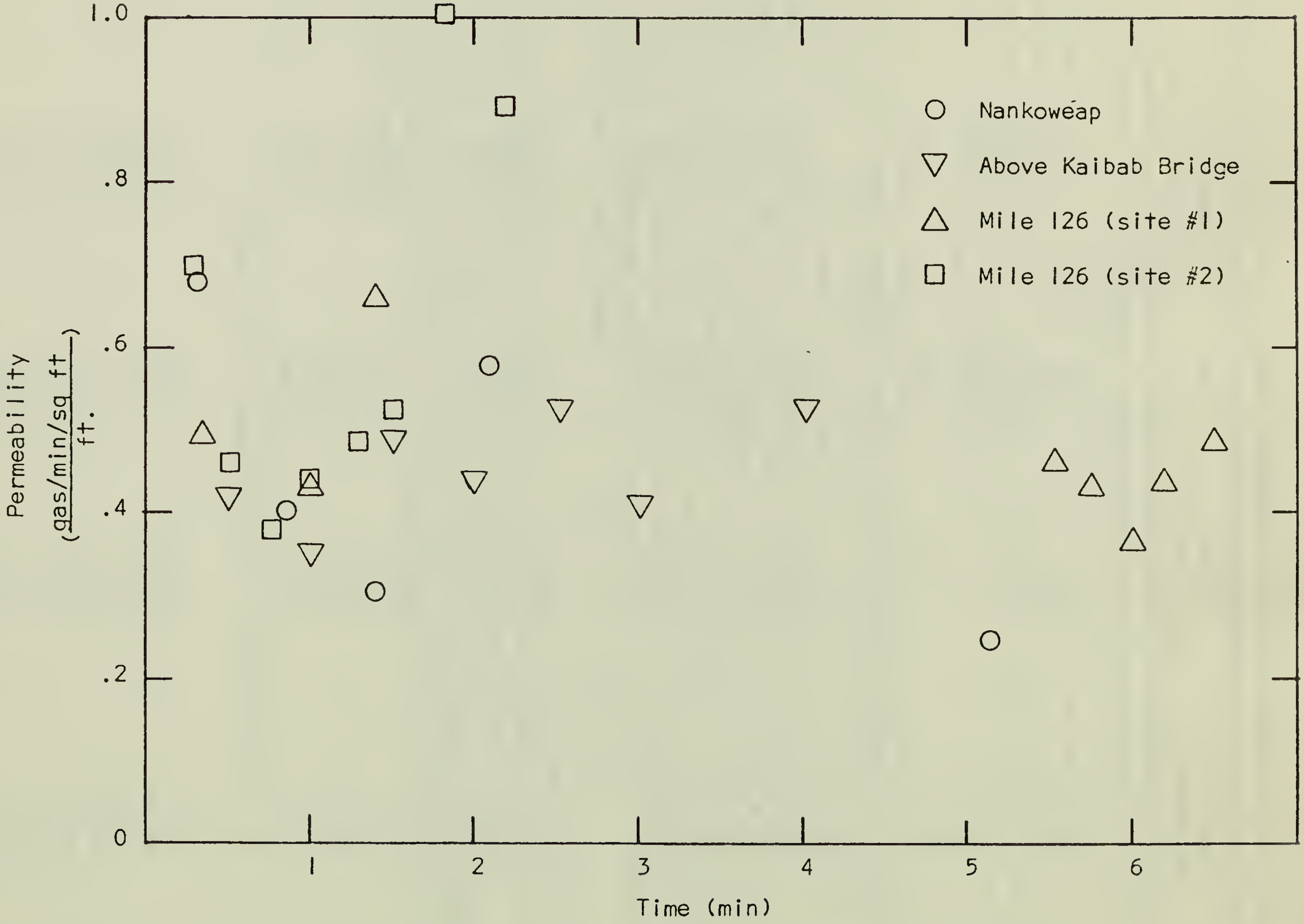


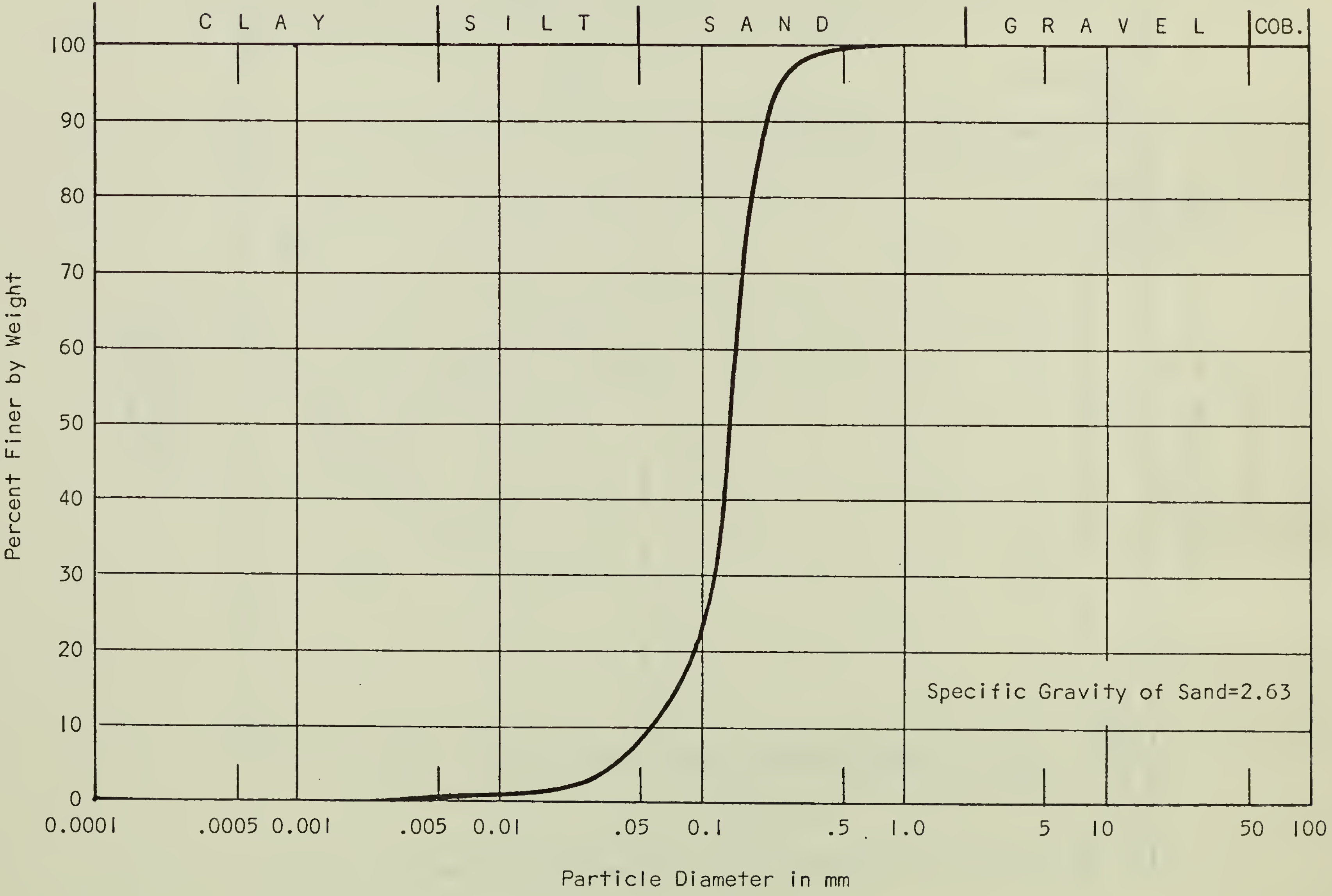
Figure 9. Permeability of Beach Soils as Determined by Infiltration Tests

Table XXII

Grain Size Distribution Analysis Of Sand

Particle Diameter (mm)	% Finer by Weight
2	100
0.83	99.9
0.42	99.8
0.23	96.5
0.10	24.1
0.073	16.6
0.039	3.87
0.028	2.41
0.018	1.8
0.010	1.5
0.007	1.2

Figure 10. Mechanical Analysis of Grain Size Distribution of Beach Sand



CHAPTER 6

PATHS OF DISEASE TRANSMISSION AND THE RISK OF INFECTION

There are numerous paths by which enteric pathogenic organisms associated with fecal matter may be transmitted from one person to another while on the river. The most direct route, which can in no way be affected by altering present disposal methods, is a person harboring a disease who returns from the latrine with fecal matter on his hands and, without taking adequate sanitary precautions, prepares or handles food, or dips a cup and some portion of his hand into unchlorinated drinking water. If, because of the inadequancy of present disposal methods, fecal matter or contaminated sand is on the surface, several other modes of transmission are possible. The hands or gear of an uninfected individual may become contaminated, with subsequent transfer of organisms to eating utensils or food. Pathogens may be picked up and carried by rodents, flies, or other insect vectors. Organisms also may be transferred by wind blown sand and/or fecal matter. Animals, through contact with infected feces, may contract some diseases and serve as reservoirs.

For all the modes of transmission mentioned above, there would seem to be intrinsic limits as to the quantity of fecal matter or the number of organisms which could be transferred. For example, very few organisms and only miniscule amounts of fecal matter could be transferred by flies. It also does not seem likely that river-runners would frequently ingest visible amounts of fresh fecal matter. Therefore, it is important to determine if the number or organisms possibly associated with a miniscule amount of infected fecal matter might cause the onset of disease in others.

Bacterial Diseases

Thomson [52] found that the number of pathogenic organisms in a gram of feces from persons in the early stages of shigellosis, salmonellosis, and partyphoid ranged from 10^4 to 10^8, with 10^6 organisms most frequently encountered (greater than 40% of the specimens in each case). One million organisms per gram is also a good estimate for the density of *Salmonella typhi* found in the feces of asymptomatic typhoid carriers [53]. However, for paratyphoid carriers, Thomson found that the density of *S. paratyphi* B could be as high as 12×10^9/g, with 10^8/g most frequently encountered.

Hence, the number of pathogenic organisms in a small amount of infected feces, perhaps 0.01 to 0.001/g, could vary from an average of 10^4 up to 10^8. Since die-off studies have shown rapid reduction in enteric organism concentrations, the occurrence of the higher numbers of organisms should be infrequent. In this case, the risk of disease transmission is dependent upon whether lower numbers of organisms--around 10,000, for example--constitute an infective dose.

While typhoid is not a frequently encountered disease in the United States, the dose-response studies conducted by Hornick et al. [52,53] are the most complete and have the clearest results. Graded doses of S. typhi were ingested orally by human volunteers and the frequency of infection of typhoid fever noted. The dose response data gathered by Hornick are shown in Table XXIII and, graphically, in Figure 11. These results would indicate that for a challenge dose of 10,000 S. typhi the probability occurrence of typhoid is less than 10%. The results of toher investigators [54-56] indicate that the virulence of strains of Salmonella other than S. typhi may be somewhat less. This result may be ascribable to the lesser severity of the symptoms of various types of salmonellosis and, hence, the greater difficulty in diagnosing the occurrence of disease; or to an increased level of resistence to salmonellosis in the general population due to the more frequent occurrence of the disease organisms. Based on the above data, it may be estimated that a river-runner who ingests 0.01/g of fresh feces--from an individual who has recently had salmonellosis, or from a carrier--has a 1 in 100 chance of contracting the disease.

The actual risk of disease for the average river-runner is much lower due to several factors. Obviously, not every river-runner will directly ingest 0.01/g of feces, as even that small an amount is quite visible. Also, as was explained in Chapter 1, the frequency of occurrence of high numbers of pathogenic organisms in association with fecal matter depends on the number of persons infected with a specific disease. Hence, fecal contamination does not necessarily entail the presence of pathogenic organisms. That the feces must be quite fresh is indicated by the die-off studies. Small masses of feces or contaminated sand on the surface would experience very rapid reductions in the enteric organism content at least under the high temperature--low moisture conditions which predominate in the Canyon over most of the river-running season. Intact feces on the surface would have slower death rates but, due to their visibility, should be easily avoidable. While the transmission of bacterial diseases via direct ingestion of small amounts of fecal matter is possible, the circumstances leading to such an occurrence would be infrequent.

The above analysis is based on the fact that pathogenic bacteria do not multiply in water, sand or fecal matter under normal conditions. However, enteric organisms can multiply in certain foods. Enteric

Table XXIII

Relation of Dosage of *S. typhi* to Disease
(from Hornick et al. [52,53])

S. typhi Strain	Number of *S. typhi* Ingested	Total Volunteers Challenged	Number With Disease	% Volunteers Sick at Challenge Dose
Quailes	10^3	14	0	0
	10^5	116	32	27.59
	10^7	32	16	50.00
	10^8	9	8	88.89
	10^9	42	40	95.24
Quailes	10^7	30	16	53.33
Zermatt	10^7	11	6	54.55
Ty 2V	10^7	6	2	33.33
Quailes	10^5	104	28	26.92
	10^7	30	15	50.00
	10^9	4	4	100.00

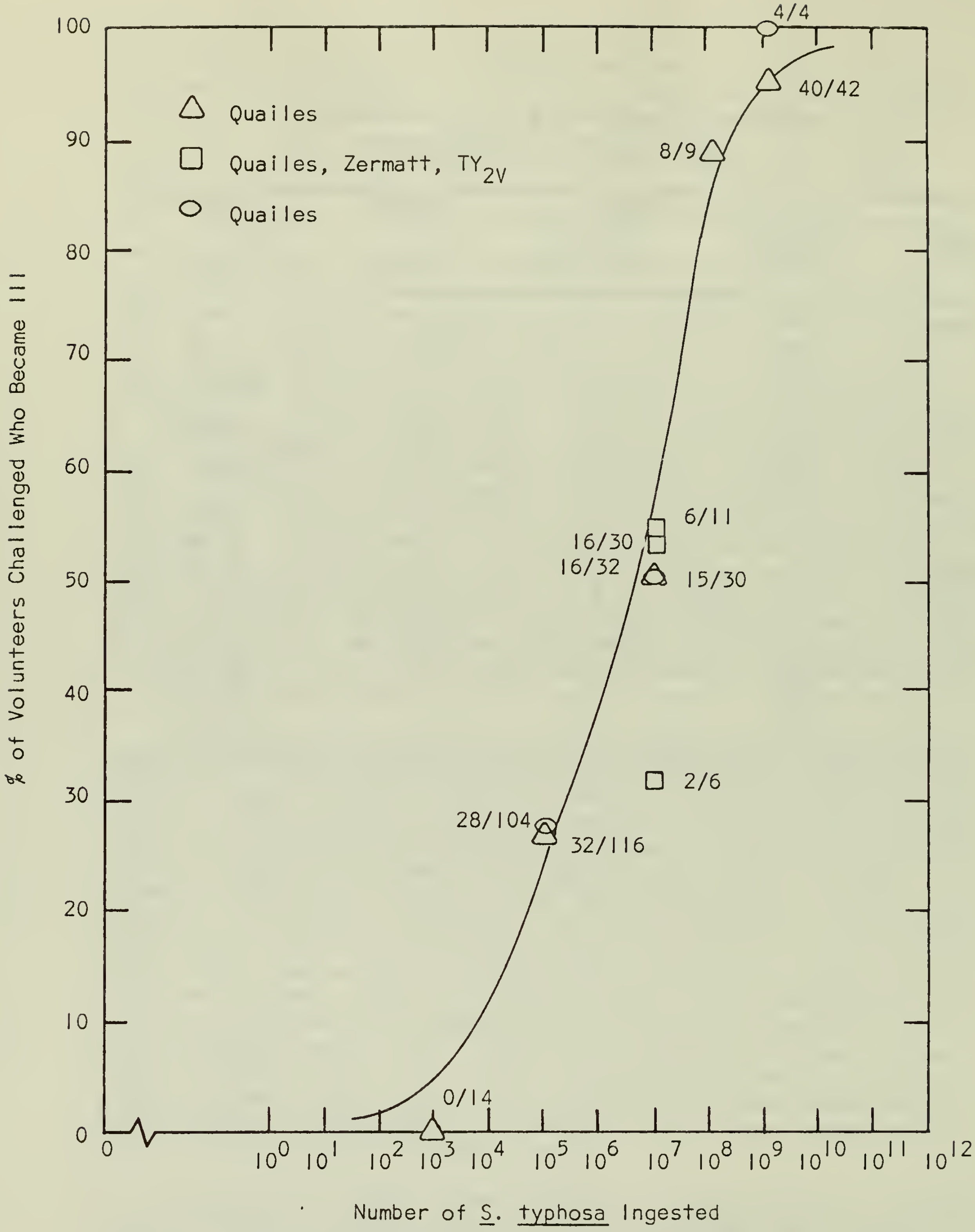


Figure 11. Relation Of Dosage Of S. typhi To Disease (from Hornick et al. [52,53])

diseases, such as salmonellosis, are most often transmitted via food--especially beef, poultry, and dairy products [57]. Small numbers of organisms transferred to the food during preparation or serving by food handlers, or by flies, other insects, or even the wind have a chance to muitiply into infectious doses if the contaminated food is stored long enough without proper refrigeration.

Viral Diseases

There have been several investigations into the relationship between the infectivity for tissue cultures and for man and animals of enteric viruses administered by the oral, respiratory, and conjunctival routes. It is generally recognized that infection regularly follows the oral administration of 100,000 $TCID_{50}$ (100,000 times the concentration required to infect 50% of a set of tissue cultures) [58]. There is evidence, however, that 10 $TCID_{50}$ or less of attenuated poliovirus may cause infection. This is increasingly significant in that attenuated strains of enterovirus may be considerably less infective than their progenitor strains [59]. Sabin [60] found $10^6 TCID_{50}$ of poliovirus per gram of feces in human stools. The agent in infectious hepatitis has not yet been isolated and can not be titrated in tissue culture. Therefore, it cannot be subjected to an analysis of its infective dosage. Given the inability of viruses to multiply in water, the frequency of water-borne transmission [61] of the disease indicates that low numbers of virus can be infective. Hence, the risk of disease transmission due to the ingestion of small amounts of infected fecal matter, contaminated sand or contaminated food may possibly be greater for viral diseases than for bacterial diseases. However, the frequency of occurrence of viral diseases, such as infectious hepatititis or poliomyelitis, is so much less than that of bacterial infections, such as salmonellosis and shigella.

CHAPTER 7

ANALYSIS OF PRESENT METHODS OF BODY WASTE DISPOSAL

Although the risk of disease transmission due to the direct ingestion of fecal matter seems to be very low, surface contamination of the beaches due to improper waste disposal may be significant for three reasons: (1) low numbers of organisms when transmitted to food may multiply into infectious doses, or cause food poisoning; (2) under conditions of low temperature and high moisture, high levels of enteric organisms associated with contaminated sand will not be reduced as quickly as under normal hot-dry conditions, and the risk of disease transmission is increased; and (3) it is possible that low numbers of pathogenic enteroviruses such as might be transmitted by insects, rodents, or on wind blown sand may be infective. Hence, the possibility of contamination resulting from the two major methods of body waste consolidation and disposal, and individual clandestine dumping, will be analyzed based on the efficiency of burial and the results of the die-off studies.

Holding Toilets

The results of the field study indicate that in holding-toilet dumps the die off of enteric organisms in hot, dry weather is about 99.998% in one month. Such results are in agreement with literature studies in which organisms not in association with fecal matter are added to sand and subjected to a high temperature--low moisture conditions. This rapid decrease in microorganisms is aided by the partial physical breakup of fecal solids in this type of toilet. If the solids are not broken up they retain moisture, supply concentrated nutrients, and protect the enteric organisms from the hostile soil environment.

However, it has been shown in the infiltration studies that it is the broken-down fecal matter suspended in the liquid which clogs the sand pore spaces. The infiltration of the liquid portion into the sand is halted and, as the hole is filled, the liquid and floating feces rise closer to the surface. Die-off may be rapid, but for one or two days or more during cool, wet weather the possibility of a person coming in contact with high levels of enteric organisms is increased. The appearance of chemical dyes may decrease the likelyhood of contact, but the occurrence of blue or pink surface sands is not generally regarded as aesthetreally desirable.

The obvious solution to the eruption of wastes onto the surface is to dig larger holes. To obtain at least a foot of cover, the volume of the hole below a depth of 12 in. must be 2.5 times the total waste and rinse volume since the sand has a 40% porosity. The problem of digging adequate holes in the cohensionless soil may be overcome by wetting the soil with river water as the digging progresses. The apparent volume of the hole may then be increased by caving in the lower sides of the hole if the waste volume has been underestimated. This last procedure is not recommended, however, since it is usually the boat crew who are responsible for disposing of wastes as well as food handling. Any process which increases the contact of a food handler with wastes and, hence, the chance of him contracting a disease or transmitting disease organisms to others should be avoided.

The end result of an efficient disposal of the contents of a holding toilet is approximately 1 cu ft of contaminated sand containing little intact fecal matter, and having a rapidly decreasing moisture and enteric organism content.

Dry Hole Burial

The dry hole method of waste disposal involves digging a hole and setting a portable toilet seat over it. The infiltration problem experienced with holding toilets is not apparent with this method. Urine rapidly infiltrates into the sand leaving a layer of intact fecal matter. The simulated dump study indicated that the die-off of enteric, organisms in association with intact fecal matter may be slow. However if the fecal matter is buried deeply so that there is little possibility of exposure and contact, this is not a problem. Because of the visibility of the intact fecal matter, there is little chance that such a dump could be exposed without the digger's knowledge.

There are certain problems with this type of dump, however. The first is the digging of an adequate hole in sand. Because the hole does not have to contain liquid wastes, it can be smaller than that for the holding toilet. Again, the problem of digging a hole in the cohesionless sand can be overcome by wetting the sand with river water as the digging progresses. Other problems result from the length of time which the hole must be left open. Flies and odors can be prevented by a light soil cover after individual use, or by using a solid box-type toilet set.

The end result of this type of burial is a layer of intact fecal matter. Since the urine volume is small, there should be little movement of contamination into the surrounding sand. The stabilization of available organics may be more rapid in this type of dump because of the increased survival of organisms. As the drying process proceeds, the volume of the fecal matter will decrease significantly. For example, if one assumes an average value of 150 g of feces/person/day with a 75% moisture content, the volume of the body wastes from 20 persons after drying should be approximately 0.03 cu ft and, hence, would constitute a layer 0.4 in. thick in a hole with a 1-ft diameter.

Individual Surface and Shallow Dumps

While the majority of wastes are disposed of according to regulation, there is a substantial amount of individual clandestine dumping. This includes direct surface dumps and shallow "sneaker" or "cat" holes.

Aesthetically, the surface dump, or the shallow dump which is exposed by trampling or wind, is the most obnoxious. From a human health point of view, these dumps present the least danger of contamination by direct contact because of their high visibility. Any pathogenic organisms are exposed to the high surface temperatures which sterilize and desiccate. The major problems are: (1) that the "good" places to locate a dump are usually shaded or protected to a degree which slows the death rate of organisms, (2) the danger that insects, rodents and other animals, or the wind might transmit disease organisms to food, drink, or other animals including humans, (3) the exposed dump may be camouflaged by blowing sand or vegetation, and (4) the presence of paper and other sanitary items which remain intact even when desiccated.

The problem with individual dumps covered by only a thin layer of sand is that they lose the advantage of high visibility. The perfect location for such a site may be exactly where a river-runner may decide to sit or sleep that evening, with resulting contamination of body and/or gear. Barring such a development, if the shallow, covered dump stays covered, the problem of flies and insect transmission is avoided, while high surface temperatures sterilize-desiccate any potential pathogenic organisms.

CHAPTER 8

RECOMMENDATIONS FOR DECREASING RISK OF DISEASE TRANSMISSION WITHOUT ALTERNATIVE METHODS

A major pathway by which disease organisms may be transmitted to man is via food. Hence, of importance in reducing the spread of disease is the practice of sanitary precautions in the preparation and serving of food and drink. Certain practices such as proper refrigeration of foods and disinfection of drinking water must be emphasized. Since it has been shown that flies and other insects may transfer disease organisms, it would be beneficial for river parties to cover prepared food with fine-mesh netting before and after the major rush of serving.

For aesthetic as well as health reasons, individual surface and shallow dumps must be eliminated. The first step in any problem to accomplish this end must be the education of the river-runner. Then the occurrence of situations where there is no alternative to individual dumps must be reduced. For lunch and attraction stops, toilets should be set up just as in the evening, or alternative systems devised. One possibility is the use of an ammunition can with an attachable toilet seat. Such a can can be kept accessible in rigging the craft; and, in the evenings, the charge (water plus disinfectant) for a holding toilet could be used to rinse the contents into the holding toilet, or it could simply be emptied into a dry-hole. If an individual dump must be made, a small spade or shovel should be kept available so that a deep hole can be made. In any case, any paper or other sanitary items should be separated and either carried out or burned in a firepit.

At locations along the river where parties stop for prolonged periods, or where backpackers increase the user load, sanitary pit privies should be built. The need for such a facility is most apparent at Havasu Creek. Other locations of heavy impact to be considered are Nankoweap, Hance, Monument, and Lava Falls Rapids and at Deer Creek. Although this type of facility is not in keeping with a "wilderness experience," the condition to which some areas have degraded make consideration of these units an unfortunate necessity. The visual aesthetics can be in part preserved by using natural materials in the construction of the privies, rather than sterile fiberglass. Privies will be further considered as an alternative in a later section.

River parties should be encouraged not only to stop before major attraction stops such as Vassey's Paradise, Deer Creek Falls, Elves Chasm and Matcatameba, but they should also be urged to set up holding toilets, dry hole burials or alternatives such as the ammunition can at such stops.

While certain parts of the regulations concerning the burial of wastes might be relaxed (such as the requirement of 50 ft between the burial site and the river), certain other stipulations should be emphasized and/or added. Because flies are vectors of disease, the importance of keeping the latrine physically away from the kitchen area should be stressed. Also, as much as is possible, flies should be kept away from the waste. Burial holes should be dug only on the level to avoid exposure due to erosion; and holding-tank burials should be made in sunny areas to take advantage of high surface temperatures and the sterilizing radiation. This precaution is not necessary for dry-hole burials. Of course, the need for digging an adequate hole can not be over-emphasized. The use of the wet-method when necessary for getting deep holes in the cohesionless sand should be promoted.

CHAPTER 9

ALTERNATIVE METHODS OF DISPOSAL

Present methods of waste disposal being used on beaches within the Grand Canyon have been discussed in the previous section. Other methods are available and should be considered. Not all involve radical changes in present procedures.

Pit Privy

A pit privy can be constructed to be flyproof, rodentproof, and relatively odorless [62]. Although wastes are consolidated, studies have shown that bacterial travel through soil is minimal even though the pit is within the ground water table; if located above the water table, bacterial movement is restricted to a few feet.

The advantage of pit privies would be that boat crews would be freed of the task of waste disposal, the multitude of disposal sites would be reduced, and sanitary facilities would be available and convenient to river-runners and backpackers.

A major disadvantage of these units would be the problem of maintaining them in a sanitary condition. The erection of privies on all beaches would be neither economical nor desirable. The concept of "wilderness" is contradicted by the presence of sterile, green, fiberglass structures. However, considering the condition of certain locations within the Canyon, compromises with the wilderness concept are needed. Certainly, Havasu Canyon is a case in point. This area has become greatly deteriorated by the large number of individual dumps. This problem can be alleviated by erecting a privy off the trail--preferably out of natural rock material. It would be the responsibility of boat crews to sanitize the seat with an hypochlorite solution. Further consideration should be given to other attraction stops, such as Nankoweap, Hance, Monument, Hermit, and Lava Falls Rapids and Deer Creek and Tapeats Creek.

Carryout of Human Wastes

An obvious solution to the problem of waste disposal in the Canyon is to require river parties on the larger boats (those less likely to be overturned) to carry out their body wastes. This method of disposal is currently practiced by one commercial party on the river. Holding tanks with capacities equal to the volume of wastes generated by the river parties are built into the boats. The contents of holding toilets are drained into the tanks along with a small amount of rinse water. There is a minimum amount of contact between the boatman and the waste. As 90% of those running the Colorado River are members of commercial river parties, this would substantially reduce the number of burials and, hence, the chance of a burial being dug up. The disadvantages of this method might be (1) contact with fecal material during the transfer of wastes from the holding toilet to the tank, (2) the danger of a boat being overturned or a tank punctured and a large quantity of concentrated waste being released into the river, and (3) the practice of discouraging urination in the holding toilet in order to reduce the volume. Without a designated place for urinating, urine and urine odors are spread all over beaches. Aside from aesthetics, pathogenic organisms are infrequently excreted in the urine of infected individuals. If urinating at the edge of the river below high water mark is encouraged, it must be downstream of where water will be drawn for cooking and drinking purposes. Carry-out probably should be encouraged at present; and, if the number of user-days is significantly increased, it should be required in the future.

A modification of the complete carryout method has been tried. This technique involves the separation of the solid and liquid portion of the contents of a holding-toilet by straining the contents through a fine net or sieve. The liquid portion is buried and the solids are carried out. This method shares all the disadvantages of burial of the total contents of a holding toilet (except for a slightly reduced volume) and results in a prohibitatively increased chance of contact between the boatman and the fecal matter. Besides the health aspect, this high degree of contact would make separation aesthetically unacceptable to many boat crews. The reduction of carryout volume is the only advantage of separation, and is an insufficient reason for recommending the application of this practice in the Canyon.

Designated Waste Disposal Beaches

Certain of the smaller, little-used beaches or an area within a large beach might be dedicated solely to the purpose of waste disposal. These might be located at reasonable intervals along the river, such that wastes might need to be stored for only several days, rather than the length of the entire trip.

The wastes from the holding-toilets or boat tanks could then be dumped into marked holes and buried, or into a single constructed tank.

A single-tank disposal unit could be constructed much like a septic tank system. The major function of the tank would be for solid-liquid separation and storage of solids. Unless disinfecting chemicals are used to excess, biological digestion of the solids should occur. This will reduce the volume of solids to be disposed of. The liquid overflow from the tank would be disposed of by a subsurface filter or trench.

The accumulation of solids will necessitate that in time the tank will need to be pumped out to a drying bed to reduce the water content and volume. A sand-lined shallow basin would serve this purpose. After drying the remaining solids would be piled and burned, using added fuel-oil. Since the drying and incineration process would result in odors, it should be performed during the off-season.

Incineration Toilets

This type of toilet has been employed in mobile homes and marine installations and utilizes a propane or natural gas flame to oxidize the wastes to exhaust gases and ash. No water or chemicals are required and relatively low operation and maintenance is necessary.

Several drawbacks exist with this type of unit. In addition to the toilet, which weighs 95 lb, it is necessary that a 12-volt battery and the required fuel tanks be available. An electrical source must be used as an ignitor and to run a 140 cfm blower. Waste gases are exhausted through an 8-10 ft, 4-in. diameter stack. The cost of this type of toilet is about $500.

Although this unit would alleviate a portion of the beach burials, only the larger craft could utilize this system. Power and fuel would be an additional problem, since both would be necessary throughout the trip. Conversations with one manufacturer's representative were not at all encouraging as to the suitability of this type of unit for this application.

Direct River Disposal

This alternative cannot be given serious consideration under any circumstances. Certainly, any stream or river has a self-purification capacity. However, since the river is used as a drinking water supply, the risk of disease transmission is too great.

A simple calculation can be used to illustrate this point. A die-off of enteric microorganisms in water will occur because of the hostile environment and predation, and will do so at somewhat predictable rates. Let us assume that the contents of a portable toilet used by a party of twenty for one day is dumped into a river flow of 10,000 cfs. The total number of fecal coliform bacteria will be approximately 40 billion organisms. If it were to take three minutes to dump and rinse the toilet, and complete mixing were to occur, the fecal coliform level in the river would be about 80 FC/100 ml. The contents, however, would probably get mixed with only 20% of the river water, resulting in a density of about 400 FC/100 ml. Using Chick's Law for microbial death, and a death rate constant $k = 0.5$ for cold water, it would take approximately 0.6 days to reduce the numbers to the 200 FC/100 ml recommended maximum for primary contact recreation. Further assume a river velocity of 5 mph. Were another party camped one mile below to remove water for drinking or cooking, the levels of fecal coliforms would only have been reduced about 1%.

In 1968, the National Technical Advisory Committee on Water Quality Criteria made the following recommendations to the Secretary of the Interior [63]:

"<u>General Requirements</u>

I. All surface waters should be capable of supporting life forms of aesthetic value.

II. Surface Waters should be free of substances attributable to discharges or wastes as follows:

   a. Materials that will settle to form objectionable deposits.

   b. Floating debris, oil, scum, and other matter.

   c. Substances producing objectionable color, odor, taste, or turbidity.

   d. Materials, including radionuclides, in concentrations or combinations which are toxic or which produce undesirable physiological responses in human, fish, and other animal life and plants.

   e. Substances and conditions or combinations thereof in concentrations which produce undesirable aquatic life."

The Arizona Department of Health Services has adopted essentially the same basic requirements in their regulations, "Water Quality Standards for Surface Waters [64]." Designated primary beneficial uses of the Colorado River from Lake Powell to Topock are full and partial body contact, domestic and industrial water supply, cold and warm water fishery, agricultural, and aquatic life and wildlife. These regulations and designations prohibit the dumping of raw waste into the river.

CHAPTER 10

AREAS FOR FURTHER STUDY

Mechalas et al. have developed a methodology for numerically evaluating the risk of infection for aquatic recreationalists during primary contact activities, such as swimming and skindiving, in a specific body of polluted water [6]. The application of this methodology to the beaches of the Grand Canyon would be impossible or, useless.

To establish numerically a risk of disease transmission among river-runners due to the present method of human waste disposal practiced in the Canyon, three probability distributions would have to be generated. The first would measure the frequency of occurrence of specified concentrations of disease organisms on the surface of the beaches and side canyons along the Colorado due to inefficient body waste disposal. To be totally accurate, this curve would actually have to be a series of curves--one for each applicable disease. Salmonellae have been used in this context in similar studies for recreational water quality [65]. These data would also have to reflect changes of season and weather conditions. A second probability distribution would measure the frequency with which river-runners ingest specified numbers of disease organisms, either in association with fecal matter, sand, or contaminated food, or as a result of present methods of waste disposal. A third probability distribution, and the only one which can be determined in this case, is the measure of the probability of disease as a result of a person ingesting a given challenge dose of disease organisms. A dose response curve such as that developed by Hornick et al. [53] could be used.

In any case, even if the first probability distribution could be developed at great time and expense, the accuracy with which the second distribution could be quantified would prohibitively decrease its worth.

The only possible way to approach a quantitative analysis of the risk of illness incurred by people who run the Colorado River through the Grand Canyon would be to initiate an epidemiological investigation. River-runners, as they leave the Canyon, could be asked to report any enteric diseases diagnosed as such by doctors. Complicating the results of such a study is the fact that the high dissolved solids content of the river water (especially sulfates and magnesium salts) may have a diarrhetic effect on some river-runners. The frequency of cases of salmonella and shigella not requiring medical attention is also a factor. A more accurate method of carrying out this type of study would be to take stool samples (either individual or after the consolidation of the individual waste) at Lees Ferry or the first beach stop and at Diamond Creek for a number of river parties, and have the samples analyzed for pathogenic organisms. This method would still not separate the cases of illness resulting from drinking contaminated water from some of the tributaries and those spread through personal contact from the cases of disease resulting from inadequate disposal of human waste.

A second investigation which might be worthwhile would be to analyze the death-rate of enteric organisms in dry-hole dumps. Because of the daily variations and extremes in temperature and moisture, the die-off on the beaches should be at least slightly greater than that in the laboratory simulated cores. If the die-off is significantly greater, i.e., near the rates for holding-tank dumps, then dry hole burials should be designated by regulation as the method of consolidation and disposal to be used.

Such a study would probably involve a researcher traveling with a party which practices dry-hole burials for at least one night or until an appropriate sand beach could be reached. The researcher could aid in digging and marking a hole, and, in the morning, homogenizing the waste in the hole--seeing that it is evenly spread over the bottom of the hole before the hole is filled. He would take an initial core and then be picked up by helicopter, so that the initial number of fecal coliforms could be accurately determined in the lab by the MPN multiple tube method. The coring and analysis should be repeated after three or four days and then at intervals appropriate to the die-off observed over the first period. Although it would be interesting to repeat the burial and analyses procedures in a cooler, wetter month such as September and gather temperature and moisture data, this more involved analysis would not be absolutely necessary. If the burial were made in June, assuming temperature and moisture conditions are similar, the results could be directly compared to the chemical holding-toilet field study results.

BIBLIOGRAPHY

1. Smith, B., Grand Canyon National Park, National Park Service, Personal communication (1976).

2. Inner Canyon Unit Office, Grand Canyon National Park, National Park Service, cited by Knudson [4].

3. National Park Service, Private River Trip Permit Criteria, (1975).

4. Knudson, A. B., "A Bacteriological Analysis of Portable Toilet Effluent at Selected Beaches Along the Colorado River, Grand Canyon National Park, Arizona," unpublished Colorado River Research Report, Grand Canyon National Park, Arizona (1976).

5. Pelczar, M. J., and R. D. Reid, Microbiology, McGraw-Hill, New York (1972).

6. Mechalas, B. J., et al., "An Investigation into Recreational Water Quality," Water Quality Data Book, Vol. 4, EPA Water Poll. Control Res. Series 18040 DAZ 04/72 (1972).

7. "Morbidity and Mortality Weekly Report," Annual Summary, 1975, Center for Disease Control, 24 (1976).

8. Rudolfs, W., et al., "Literature Review on the Occurrence and Survival of Enteric, Pathogenic, and Relative Organisms in Soil, Water, Sewage, and Sludges, and on Vegetation," Sewage and Industrial Wastes, 22, 1261 (1950).

9. Beard, P. J., "Longevity of Eberthella typhosus in Various Soils," Amer. Jour. Public Health, 30, 1077 (1940).

10. Pound, C. E., and R. W. Rites, "Wastewater Treatment and Reuse by Land Applications," Vol. II, EPA Technol. Series, EPA660/2-73-0056, (1973).

11. Granger, J., and E. Deschamps, Arch. de Med. Exper. et D'Anat. Path., I, 33 (1889), cited in Beard [9].

12. Karlinski, J., Cent. f. Bakt. (Ger.), 65 (1889); Arch. f. Hyg., 13, 302 (1891), cited in Rudolfs et al. [8].

13. Dempster, R., Brit. Med. Jour., 2, 936 (1902).

14. Pfuhl, cited by Beard [9].

15. Robertson, *Brit. Med. Jour.*, 1, 69 (1898), cited by Rudolfs et al. [8].

16. Martin, S., "Annual Reports of the Medical Officer of the Local Government Board," (1896-1900), cited by Rudolfs et al. [8].

17. Rullman, W., *Centralbl. f. Bakt.* (Ger.), 30, 321 (1901), cited by Beard [9].

18. Firth, R. H., and W. H. Horrocks, "An Inquiry into the Influence of Soil, Fabrics, and Flies in the Dissemination of Enteric Infection," *Brit. Med. Jour.*, 2, 936 (1902).

19. Sedgwick, W. T., and C. E. A. Winslow, *Mem. Amer. Acad. Arts and Sci.*, 12, 508 (1902), cited by Rudolfs et al. [8].

20. Melick, C. O., "The Possibility of Typhoid Infections Through Vegetables," *Jour. Infect. Dis.*, 21, 28 (1917).

21. Murillo, F., "Cholera, Typhoid, and Vegetables," *Plus Ultra* (Madrid), 2, 115 (1919), cited by Rudolfs et al. [8].

22. Kligler, I. J., "Investigations of Soil Pollution and the Relation of the Various Types of Privies to the Spread of Intestinal Infections," International Health Board, Monograph 15, Rockefeller Inst. of Med. Res., p. 1 (October 10, 1921), cited by Rudolfs et al. [8].

23. Levy and Kayser, *Centralbl. f. Bakt.*, 33, 489 (1903), cited by Rudolfs et al. [8].

24. Galvagne and Calderini, *Zeit. f. Hyg.*, 61, 188 (1908), cited by Rudolfs et al. [8].

25. Ruchhoft, C. C., "Studies on the Longevity of *B. typhosus* in Sewage Sludge," *Sewage Works Jour.*, 6, 1954 (1934).

26. Williams, R. S., and W. A. Hoy, *Jour. Hyg.*, 30, 413 (1930), cited by Rudolfs et al. [8].

27. Maddock, E. C. G., "Studies on the Survival Time of Bovine Tuberele Bacillus in Soil, Soil and Dung, in Dung, and on Grass," *Jour. Hyg.*, 33, 103 (1933).

28. Mallman, W. L., and W. Litsky, "Survival of Selected Enteric Organisms in Various Types of Soil," *Amer. Jour. of Pub. Health*, 41, 38 (1951).

29. Dazzo, F., et al., "The Influence of Manure Slurry Irrigation on the Survival of Fecal Organisms in Seranton Fine Sand," *Jour. Environ. Qual.*, 2, 470 (1973).

30. Rhines, C., "The Persistence of Avian Tubercle Bacilli in Soil and in Association with Soil Microorganisms," *Jour. Bact.*, 29, 299 (1935).

31. Mirzoev, G. G., "Survival Time of Shigella Bacteria at Low Temperatures and Questions of the Self Purification of the Soil and Water in the Far North," *Gig. Sanit.* (USSR), 33, 106 (1968), Microb. Abs., 128754 (1968).

32. Bagdasarian, G. A., "Survival of Viruses of the Enterovirus Group (Poliomyelitis, ECHO, Coxsaskie) in Soil and on Vegetables," *Jour. Hyg. Epidemiol. Microbiol. Immunol.*, (Prague), 8, 497 (1964).

33. Duboise, S. M., et al., "Poliovirus Survival and Movement in Sandy Forest Soil," *Appl. and Environ. Microbiol.*, 31, 536 (1976).

34. Glotzbecker, R. A., and A. L. Novello, "Poliovirus and Bacterial Indicators of Fecal Pollution in Landfill Leachates," News of Environmental Research in Cincinnati, USEPA (January 31, 1975).

35. Lefler, E., and Y. Kott, "Virus Retention and Survival in Sand," in "Virus Surval in Water and Wastewater Systems," J. F. Malina, Jr., and B. P. Sagik (Ed.), Proceedings of the Center for Research in Water Research Symposium, Number Seven, Univ. of Texas, Austin (1975).

36. Sobsey, M. D., et al., "Studies of the Survival and Fate of Enteroviruses in an Experimental Model of a Municipal Solid Waste Landfill and Leachate," *Appl. Microbiol.*, 30, 565 (1975).

37. Geldreich, E. E., "Sanitary Sigificance of Fecal Coliforms in the Environment," Fed. Water Qual. Cont. Admin., Publication No. WP-20-3 (1966).

38. Cuthbert, W. A., et al., "Survival of *Bacterium coli* and *Streptococcus faecalis* in Soil," *Jour. Appl. Bacteriol.*, 18, 408 (1955).

39. Ostrolenk, M., et al., "Comparative Studies of Enterococci and *Escherichia coli* as Indicators of Pollution." *Jour. Bacteriol.*, 53, 197 (1947).

40. Young, C. C., and M. Greenfield, "Observations on the Viability of the *Bacterium coli* Group Under Natural and Artificial Conditions," *Amer. Jour. Public Health*, 13, 270 (1923).

41. Van Donsel, D. J., et al., "Seasonal Variations in Survival of Indicator Bacteria in Soil and Their Contribution to Stormwater Pollution," Appl. Microbiol., 15, 1362 (1967).

42. Geldreich, E. E., et al., "Technical Considerations in Applying the Membrane Filter Procedure," Health Laboratory Science, 4, 113 (1967).

43. Standard Methods for the Examination of Water and Wastewater, 14th Ed., Amer. Pub. Health Assoc., Washington, D. C., (1976).

44. Bronfenbrener, J., et al., "On Method of Isolation of the Members of the Colon-Typhoid Group of Bacteria," Jour. Bacteriol., 5, 79 (1920).

45. Frost, W. H., and H. W. Streeter, "A Study of the Pollution and Natural Purification of the Ohio River. Bacteriolgical Studies," Public Health Bulletin, 143, USPHS, Washington, D. C. (1924).

46. Smith, R. J., "Relationships of Indicator and Pathogenic Bacteria in Stream Water," Jour. Water Poll. Control Fed., 45 (1973).

47. Geldreich, E. E., and N. A. Clark, "Bacterial Pollution Indicators in the Intestinal Tract of Freshwater Fish," Appl. Microbiol., 14, 429 (1966).

48. Jordan, E. O., "The Changes in Bacterial Content of Stored Normal and Typhoid Feces," Jour. Infect. Dis., 38, 306 (1926), cited in Rudolfs et al. [8].

49. Geldreich, E. E., et al., "Occurrence of Coliforms, Fecal Coliforms, and Streptococci on Vegetation and Insects," Appl. Microbiol., 12, 63 (1964).

50. Thomson, S., "The Number of Pathogenic Bacilli in Feces in Intestinal Diseases," Jour. Hygiene, 53, 217 (1955).

51. Thomson, S., "The Number of Bacilli Harboured by Enteric Carriers," Jour. Hygiene, 52, 67 (1954).

52. Hornick, R. P., et al., "Typhoid Fever: Pathogenesis and Immunologic Control," Part 1, New England Jour. Med., 283, 686 (1970).

53. Hornick, R. B., et al., "Typhoid Fever: Pathogenesis and Immunologic Control, Part 2, New England Jour. Med., 283, 739 (1970).

54. McCullough, N. B., and C. W. Eisele, "Experimental Human Salmonellosis. I. Pathogenicity of Strains of *Salmonella melegaridis* and *Salmonella anatum* Obtained from Spray-Dried Whole Eggs," *Jour. Inf. Dis.*, 88, 278 (1950).

55. McCullough, N. B., and C. W. Eisele, "Experimental Human Salmonellosis. III. Pathogencity of Strains of *Salmonella newport*, *Salmonella derby*, and *Salmonella bareilly* Obtained from Spray-Dried Whole Eggs," *Jour. Inf. Dis.*, 89, 209 (1951).

56. McCullough, N. B., and C. W. Eisele, "Experimental Human Salmonellosis. IV. Pathogenicity of Strains of *Salmonella pullorum* Obtained from Spray-Dried Whole Eggs," *Jour. Inf. Dis.*, 89, 259 (1951).

57. "Salmonella Surveillance," Annual Summary, 1973, USPHS, Center for Disease Control, Report No. 121 (1974).

58. Malherbe, H. H., and M. M. Strickland-Cholmley, "Quantitative Studies on Viral Survival in Sewage Purification Processes," in *Transmission of Viruses by the Water Route*, G. Berg (Ed.), John Wiley & Sons, New York (1967).

59. Plotkin, S. A. and M. Katz, "Minimal Infective Doses of Viruses for Man by the Oral Route," in *Transmission of Viruses by the Water Route*, G. Berg (Ed.), John Wiley & Sons, New York (1967).

60. Sabin, A. B., "Behavior of Chimpanzee-Avirulent Poliomyelitis Viruses in Experimentally Infected Human Volunteers," *Amer. Jour. Med. Sci.*, 230, 1 (1955).

61. Moseley, J. W., "Transmission of Viral Diseases by Drinking Water," in *Transmission of Viruses by the Water Route*, G. Berg (Ed.), John Wiley & Sons, New York (1967).

62. Salvatto, J. A., Jr., *Environmental Engineering and Sanitation*, 2nd Ed., Wiley-Interscience, New York (1972).

63. "Water Quality Criteria," National Technical Advisory Committee to the Secretary of the Interior, Fed. Water Poll. Control Adm., Washington, D. C. (1968).

64. "Water Quality Standards for Surface Waters," Arizona Dept. of Health Services, R9-21.

65. Dudley, R. H., et al., "A Scientific Basis for Determining Recreational Water Quality Criteria," *Jour. Water Poll. Control Fed.*, 48, 2761 (1976).