Microbial studies of a high alpine water supply used for recreation
by Sidney Arthur Stuart

A thesis submitted in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in Microbiology
Montana State University

Abstract:
Selected waters from the high alpine zone within Grand Teton National Park, Wyoming, were studied during the past four summers to determine the influence of various factors on the quality of these waters. The water samples collected were analyzed for populations of indicator bacteria. Water that originated in remote areas contained some indicator bacteria and these populations increased as the water flowed toward the valley. In general, the magnitude of this increase was not significantly influenced by the presence or absence of human visitors but, rather, by the nature of the biological community through which the streams flowed. It was determined that it is possible for coliforms of the non-fecal type to grow and multiply in alpine streams using extracellular products excreted by algae but it was not determined to what extent (if any) this occurs in Grand Teton National Park. Once in the valley lakes, the indicator bacteria declined to very low levels. A minority of the coliforms that were recovered from all of the sites proved to be fecal coliforms. The fecal streptococci isolated were identified as the species that were found primarily in the fecal material of the native rodent and moose populations. It is concluded that management questions that relate to the carrying capacity of alpine areas should be approached with the aid of other biological parameters along with levels of indicator bacteria in the streams.

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SUPPLY USED FOR RECREATION

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SIDNEY ARTHUR STUART

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Approved:

Chairman, Examining Committee

Head, Major Department

Graduate Dean

Montana State University
Bozeman, Montana

December, 1975

ACKNOWLEDGMENTS

The author would like to thank Dr. Gordon A. McFeters for his guidance and assistance throughout the course of this study. Thanks are also due to Dr. David G. Stuart, Dr. John C. Wright and Mr. John E. Schillinger for their technical and editorial assistance. A sincere thanks to Susan Harrigan, Jim Hawkins and Sandra Dunkel for their field and laboratory assistance.

The help of Peter Hayden and Frank Betts, of the National Park Service, is gratefully acknowledged. The use of facilities at the Jackson Hole Biological Research Station under the direction of Dr. Oscar H. Paris is also appreciated.

This work was supported by the National Park Service (Contracts No. CX-12004B025, CX-6000-3-0087, 929-0-P20067 and PX-1200-50892), the United States Environmental Protection Agency (training fellowships No. U-910413-01 and U-910687-01-0) and the New York Zoological Society.

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ABSTRACT

Selected waters from the high alpine zone within Grand Teton National Park, Wyoming, were studied during the past four summers to determine the influence of various factors on the quality of these waters. The water samples collected were analyzed for populations of indicator bacteria. Water that originated in remote areas contained some indicator bacteria and these populations increased as the water flowed toward the valley. In general, the magnitude of this increase was not significantly influenced by the presence or absence of human visitors but, rather, by the nature of the biological community through which the streams flowed. It was determined that it is possible for coliforms of the non-fecal type to grow and multiply in alpine streams using extracellular products excreted by algae but it was not determined to what extent (if any) this occurs in Grand Teton National Park. Once in the valley lakes, the indicator bacteria declined to very low levels. A minority of the coliforms that were recovered from all of the sites proved to be fecal coliforms. The fecal streptococci isolated were identified as the species that were found primarily in the fecal material of the native rodent and moose populations. It is concluded that management questions that relate to the carrying capacity of alpine areas should be approached with the aid of other biological parameters along with levels of indicator bacteria in the streams.

INTRODUCTION

Water originating in the high apline zone contributes significantly to the water supply for much of the western part of the nation. In the past, this alpine zone has been visited by relatively few people. However, the quality of water in high-elevation mountain areas is becoming an increasingly important consideration to agencies such as the National Park Service and the U.S. Forest Service who have expressed concern both for the possible public health consequences of drinking untreated "pure mountain stream" water, and for ecological consequences of nutrient additions. This increased concern over the water quality of high-elevation mountain areas is partly due to the increasing numbers of people who visit these areas each year.

Significant increases in wilderness-related outdoor recreation have taken place and continue to do so at a rapid rate. Barbaro et al. (4) report a seven percent increase per year over the last decade. In effect, the movement of large numbers of people each summer from metropolitan centers to remote regions in the western United States represents periodic, seasonal urbanization of many National Parks and forests. Grand Teton National Park is such an area where in 1975, 2,549,030 people visited the park with approximately 32,045 of these people traveling to the alpine back-country within the park boundaries. With this substantial use of park facilities there is growing concern over the impact of visitor use on the aesthetic and wilderness values, campsite deterioration, and particularly the lowering of water quality. Any of these adverse impacts could irrevocably damage the unique qualities of such areas.

The drinking water used by these park visitors originates in the back-county where summer visitor use is heavy and sanitary facilities are limited or non-existent. Visitors also contribute to the natural nutrient budget of the lakes and streams by addition of sanitary wastes, outboard motor wastes, detergents and solid wastes. In 1969, for example, solid wastes left behind by recreational users of the Boundary Waters Canoe Area in Minnesota, totaled an estimated 163,440 Kg (360,000 lb) (44). According to Barton (5), this is equivalent to the addition of 1 ton of phosphates and 13 tons of nitrogen to these waters. In addition, heavy campsite use may accelerate soil erosion through deterioration of vegetation and compaction, which will further increase nutrient contributions to the waters. These observations point to the importance of understanding man's impact on the natural quality of waters within wilderness areas and their saftey for human use both in the remote back country and the more populated adjoining land.

Grand Teton National Park provides a unique opportunity to study the impact of various recreational uses on water quality, since the water from the high mountain elevations can be followed as it is exposed to a succession of recreational influences, as shown below.

1. Mountain climbing. Many of the highest and most remote alpine areas in the park are used primarily by mountain climbers for both day and over night activities. This form of recreation influences the quality of the water as it originates as snowmelt in the uppermost areas of the alpine country.

2. Backpacking and day hiking. As the streams flow toward the valley they become more accessible by trails and as a consequence are more heavily used than the more remote streams. The use of the streams in these areas vary from consumption as drinking water to fishing and bathing by some hikers and back packers. In the present investigation, the impact of these forms of recreation on water quality was studied.

3. <u>Horse-back riding</u>. Trails along the lower reaches of some streams within the park are heavily used by horses. Data obtained from streams above and below this activity will reflect the influence of this use on the quality of water.
4. <u>Swimming, fishing and boating</u>. The lakes and streams located in the valley floor of Grand Teton National Park, which collect water from the higher, more remote areas, are used for such activities as fishing, swimming and recreational boating including both motorized and non-motorized crafts. The valley floor is also the location of housing for seasonal and permanent park employees and concessionaires who operate climbing schools, boat tours, horse rides and general stores. Data obtained from waters in these areas will reflect the influence of such activities on the quality of the water within the valley floor.

Another desirable and unique quality provided by Grand Teton National Park is that the high alpine zone is a relatively simple ecosystem when compared with those of the lower wooded areas. Of particular importance in the high alpine zone is the lack of the topsoil component which normally serves as a biological and physical filter to reduce or eliminate the quantity of fecal pollutants which reach the water course. This observation suggests that the high alpine zone is more fragile and can, therefore, successfully withstand less human visitation before water quality deteriorates and become a public health hazard. This simplified ecosystem also makes it easier to study the effects of the surrounding ecosystem on the types and occurrence patterns of bacterial flora of sanitary importance found in high alpine waters.

Because of the need to study the quality of water originating in the high alpine zone and because of the unique and desirable study situations provided by Grand Teton National Park the present study was carried out on streams and lakes within that particular park. Drainages were investigated that allowed the comparison of high alpine areas exposed to intensive recreational use with those having little or no human activity. This study also provided the opportunity to follow waterborne bacterial microflora from the source of the streams in the high alpine zone and observe changes in this population as the waters progressed toward the valley floor passing through biological communities of increasing complexity.

The objectives of this study were:

(1) To determine the types and occurrence patterns of the bacterial flora of sanitary importance in a high alpine water supply.

(2) To evaluate the impact of various recreational activities on the indicator bacterial flora of a high alpine water supply.

(3) To determine the effects of the surrounding ecosystem on the bacteriological water quality.

(4) To provide a basis relative to water quality and usefulness of indicator bacteria, for the management of various areas within Grand Teton National Park.

LITERATURE REVIEW

Effects of Recreational Use on Water Quality

In recent years a number of studies have been made to determine the nature and extent of water quality changes in mountain streams resulting from recreational use. While water quality deterioration has been observed, the results of most studies indicate that there is little correlation between the quantity of pollution and the extent of recreational use. Benedetti (10), in an investigation of the effects of recreational use on municipal watersheds in the Pacific Northwest, determined that there was very little change in the water quality with recreation. The most significant effect of recreation was found to be the disruption of soil and vegetation around the water causing heavier contamination during times of runoff. Carswell et al. (14) found that there was little or no deterioration of water quality in five separate watersheds as a result of recreation permitted on streams and reservoirs of municipal watersheds. It was concluded that the effects of recreation on aesthetics, damage to facilities, complications in administration of public property, and political and sociological impacts were of greater significance than the effects of recreation on water quality.

In an extensive study of the chemical and bacteriological analysis of three mountain watersheds in Oregon and Washington, by Lee et al. (48), it was shown that there was no significant differences between the effects of a heavily used recreational area on water quality as compared with the effects of lightly used and closed watersheds. It was concluded that recreational use of an area is not always followed by high coliform counts. Also, it was determined that wild animal populations were responsible for most of the indicator organism densities, particularly at low levels of stream flow (48,61). In a similar study carried out in the Bozeman Creek Watershed, which was closed to public entry from 1917 to 1970, while the adjacent Hyalite Watershed was developed during this period by the Forest Service for public use, a significant difference in the quality of the water was found between the two areas (11,60 65). In this study, the open watershed consistently produced water of a higher quality than did the closed watershed over a six-year period. Even the highest, most undisturbed streams in the closed watershed had coliform counts exceeding state criteria for "A-Closed" municipal water supplies (30). It was also believed that the large population of big game animals in the closed area contributed to this observed lower water quality (60). Van Nierop (64) has shown that, with proper practices being observed, public uses of reservoirs and municipal watersheds such as logging, fishing, boating, etc., can be permitted without detrimental effects on water quality.

In studying the Boundary Waters Canoe Area of Minnesota, Barton (5) observed that there were some serious water quality problems attributed to recreational use. It was estimated that recreational visitors contributed approximately 9 tons NaCl, 1 ton phosphorus and 13 tons nitrogen to the waters of this remote area in one year. King and Mace (44) also conducted investigations into the effects of recreation on water quality of lakes in the Boundary Waters Canoe Area. Of the parameters measured, total coliform populations and available phosphate concentrations seemed to be affected by recreational use of selected campsites. It was concluded that the leaching of effluent from pit privies near the campsites was probably responsible for water quality changes. Shoreline activities such as swimming, washing dishes, cleaning fish, and boat launching were also listed as probable causes of contaminations. Skinner et al. (57,58), in studies of alpine and non-alpine regions in Wyoming, noted that bacteriological water quality appeared to be adversely influenced by recreational use of the land and stream flow. In a study done by the California Water Resources Center (17) on the Bishop Creek Wilderness Area high fecal coliform and high fecal streptococci counts were closely associated with human intervention. Studies done by Johnson and Middlebrooks (41) on the South Fork of the Ogden River indicate that there is insufficient contamination from present land uses along the river to exceed stream standards or to create a health hazard. However, within the recreational area, water quality data could be used to indicate differences in use at particular recreational sites.

Methods of Studying Water Quality in High Alpine Regions

Probably the most useful water quality parameter in correlating recreational use with water pollutions is the indicator bacterial population. Strong technical foundations have been laid for this type of work. Both qualitative and quantitative standard methods have been developed for the determination of bacterial indicators of pollution (1,22). Fair and Morrison (50) and Kunkle and Meiman (46,47) have provided information on proper sampling and handling of data concerning high quality water from mountainous regions. Studies on Flathead Lake in Montana demonstrated the effectiveness of using bacterial indicators of pollution to assess water quality in a relative pristine lake (6). Peterson and Boring (52) used coliform counts to assess the impacts from recreation and grazing on two isolated mountain streams. Although it was found that these uses had an influence on coliform counts, it was not determined how much of an influence came from specific land uses and how much was naturally occurring. Kabler and Clark (43) determined that fecal coliform counts are more reliable indicators of fecal contaminations than total coliform counts. Geldreich (26) states that the fecal coliform test is the most accurate determination for detecting water pollution by the feces of warm-blooded animals. In a study done on the Buffalo Lake Recreation Area, Geldreich (28) again offers a rationale for the fecal coliform concept with respect to; sanitary significance, density relationships with fecal streptococci and use of fecal coliforms as a bacteriological approach to a study of the recreational area. Geldreich has also suggested that fecal streptococci counts can be extremely useful in determining the source of pollution (29). Although fecal coliform counts have an excellent correlation with fecal contamination from warm-blooded animals, they do not differentiate between contamination from humans and animals. Geldreich found that *Streptococcus bovis*, *S. equinus* and *S. faecium* are found in very large numbers in warm-blooded animals other than man. On the other hand *S. faecalis* is rather unique to the human intestinal tract. Geldreich and Kenner (27) used a ratio of fecal coliforms to fecal streptococci (FC/FS) to determine the exact origin of contamination. A high ratio (4:1 or greater) indicates human origin, while a low ratio (1:0.7 or less) indicates animal origin. A modification of the FC/FS ratio was proposed by the Ohio River Valley Sanitary Commission (51). This approach uses the fecal coliform total coliform ratio (FC/TC) to measure the effects of human contamination on a river or stream. However, use of the FC/TC ratio was suggested more as a measure of the seriousness and extent of human fecal contamination rather than to differentiate between sources of pollutions.

One of the purposes for measuring coliform densities is to get an indication of the probable presence of pathogens. Fair and Morrison (21), however, have isolated salmonellae from unpolluted Colorado streams containing only 30 coliforms per 100 ml. Gallagher and Spino (25) point out that low total and fecal coliform counts do not by themselves indicate the absence of pathogens. Dutka (19) attacks the use of the total coliform count as an indicator based on studies showing that coliforms do not fulfill any of the criteria for a true indicator organism. Geldreich (29) has suggested that fecal coliform counts are more sensitive as indicators of pathogens. It was found that salmonella species occurred 85 percent of the time when fecal coliform counts were between 200 and 2000 per 100 ml. Smith and Twedt (59) observed that no salmonellae were isolated at concentrations of less than 200 fecal coliforms per 100 ml, but that in almost every case for densities higher than this, these pathogens were found.

Effect of Surrounding Ecosystem (other than man) on Water Quality

In a study of mountain streams by Morrison and Fair (50), it was found that runoff was the most important factor in influencing bacteriological counts. Coliform counts, as well as chemical concentrations, were found to be the highest during times of overland flow. In a similar study by Kunkle and Meiman (46), it was found that the highest total and fecal coliform counts occurred during the spring runoff and the lowest counts occurred during the winter. In a continuation of their mountain stream studies Kunkle and Meiman (47) determined that total coliform counts varied on a diurnal basis as well as seasonally and with land uses.

Kittrell and Furfari (45) determined that coliform counts were not only related to levels of organic loading, but also determined by temperature, rainfall, runoff, stream characteristics, pH and turbidity. It was also observed that total coliform counts actually increased under conditions of suitable stream temperature and pH. Hendricks (36), using a chemostat, found maximal specific growth rates for various enteric bacteria including pathogenic strains occurred at 30C in autoclaved river water taken 750 m below a wastewater outfall. Culture generation times ranged between 33.3 and 116 hr. Little or no growth occurred in the water at incubation temperatures of 20 and 5C and neither the stock cultures nor the aquatic strains were capable of growth in autoclaved river water taken above the wastewater outfall at the three different temperatures tested. Survival of bacterial indicators in subarctic Alaskan river under total ice cover and water temperature at 0C was studied by Gordon (31) who reported that after 7 days flow time, total coliforms were reduced to 3.2 to

6.5 percent of the initial count, fecal coliforms 2.1 to 4.2 percent and fecal streptococci 18.1 to 37.3 percent. Another study done by Hendricks (34) on growth of selected enteric bacteria in clear mountain stream water indicated that the aquatic environment associated with a clear mountain stream not only can maintain populations of enteric bacteria but also can supply sufficient nutrients to initiate multiplications and de novo protein synthesis. Jannasch (40) calculated generation times of 20 to 200 hr for aquatic bacteria in natural waters as a result of significant differences between dilution rates and washout rates in a chemostat. The measured growth rates were affected by the treatment of the water samples (type of sterilization) and by competition with the natural microflora for the unknown growth-limiting substrate. Bott (12) using direct measurement of bacterial growth rates in a natural stream showed that the unicellular and filamentous populations had doubling times of 42 to 51 hr when the water temperature was 0 to 5C, 8.4 to 10.8 hr when the water temperature was predominantly 11 to 16C, and 2.8 to 6.0 hr when the water temperature was 16.5 to 21.0C. Hendricks (37) in studying sorption of heterotrophic and enteric bacteria to glass surfaces, showed an initial rate of attachment equivalent to a doubling time of about 24 hr. After 24 hr both the sorbed and suspended populations stabilized with a mass doubling time approximating 100 hr. Hendricks (35) also demonstrated that bottom sediments of streams have a high adsorptive capacity for basic nutrients derived from the flowing water, retaining them in a form readily usable by various enteric bacteria including pathogenic strains. Sedimentation and adsorption of bacteria also occur in stream bottom sediments resulting in higher microbial populations than that observed in overlying waters.

The survival of intestinal bacteria in water may also be influenced by the antagonistic action of organisms in the indigenous flora, particularly by available predators. Johnstone and Kubinski (42) found that a few species of ciliated and flagellated protozoans were a major mechanism responsible for the removal of fecal bacteria in high quality waters. However, initial predatory responses required a lengthy lag period of 4 to 5 days.

Indicator bacteria must have certain quantities of organic carbon present for their survival and multiplication. Studies by Butterfield (13) and more recently by McGrew and Mallette (49), have shown that intestinal bacteria, including *Escherichia coli* can survive and later multiply in media that contained less than 5 μg/ml glucose. Hendricks and Morrison (34) also point out the fact that enteric bacteria can grow and reproduce in extremely dilute nutrient concentrations.

There is good evidence that healthy, actively growing phytoplankton species release a considerable proportion of their photoassimilated carbon into the aquatic environment (2,15,23,24,33,38,55,56,62). Hellebust (33) reported that some phytoplankton are capable of excreting up to 25 percent of their photoassimilated carbon during their log growth phase. Therefore, when large populations of algae are present, adequate supplies of carbon should be present for the survival of some heterotrophic water borne bacteria. Ward and Moyer (66) reported that organics excreted by algae during growth could serve as bacterial nutrient sources. Bell et al. (9) observed that dominant bacterial populations associated with algal blooms are a result of both stimulation and inhibition mediated by the release of extracellular products. Using C^{14}-labeled glycollate, Wright (69) measured uptake by the natural planktonic microorganisms in a lake in eastern Massachusetts. Bacteria able to grow on glycollate exhibited the same uptake pattern as seen in the lake. Bauld (7,8) in studying benthic algal-bacterial mats in alkaline hot springs, demonstrated that C^{14}-labeled organic compounds excreted during algal photosynthesis could be subsequently assimilated by natural populations of the bacteria present in the mat.

MATERIALS AND METHODS

The present research was carried out at the Jackson Hole Biological Research Station, Montana State University and at various sampling sites within Grand Teton National Park.

Travel to Sampling Sites

Travel to the sampling sites from the lab headquarters at the research station was by car and on foot. The trip to the trail heads by car was about 12 miles and the hike to the farthest sampling site was about 8 miles on foot with the other sites along the way.

Location of Study Sites

The main study areas, Figure 1, were the streams and lakes in Leigh and Cascade Canyons as well as the lakes in the valley into which these drainages empty (Leigh, String and Jenny Lakes) in addition to the streams in Glacier Gulch and Garnet Canyon. One phase of the overall study centered around a microbiological evaluation of the heavily used Cascade Canyon with Lake Solitude (elevation 9035 ft) at its upper end. Site C-1 (Lake Solitude) was located above timberline in a glaciated cirque. Sites C-2 through C-8 were located below timberline with Sites C-3 through C-8 being

Figure 1. Location of sample sites within Grand Teton National Park