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BACTERIOLOGICAL STUDIES

ON THE AVON RIVER,

CHRISTCHURCH, N.Z.

Presented in partial fulfilment
of the requirements for the Degree
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in the
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ABSTRACT

The Avon River rises from springs and flows 26 km through the city of Christchurch to an estuary. The river has many tributaries, some arising from springs, others draining lowlying swamp land. Many stormwater drains discharge into the Avon. Variations in river flow are determined largely by rainfall. For most of its length the Avon River flows through residential areas, and is used for both recreation and stormwater drainage.

The danger from polluted water is mainly from living organisms it may contain. Recent faecal contamination is the greatest danger as pathogens may be present, which, if ingested, may cause epidemics.

Previous work has shown high levels of bacterial pollution to be present but no information is available on the sources of the organisms.

This investigation was designed to establish baseline levels of contamination. It extended over a period of very low rainfall during which the contribution of various sources of pollution were examined. The effect of a period of heavy rain on the numbers of bacteria was assessed. The survival time of the bacteria in the water was also examined.

Numbers of indicator bacteria occurring in water samples from 10 sites were investigated on 10 occasions during periods of low and high rainfall. The samples were examined for numbers of faecal coliform bacteria and faecal streptococci. Sediment samples were taken from 5 of the sites from which water samples were obtained and numbers of faecal coliform bacteria and faecal streptococci in them were estimated. At most sites during low flow numbers of faecal coliform bacteria exceeded the 200/100ml specified for Class C waters. Because numbers were high at sites closest to the source of the river samples from 6 sites in the upper reaches were examined on 4 occasions.

The inputs per metre during dry weather were found to vary over the length of the river although the sources of pollution were not determined. The relative contributions of humans and animals to the contamination has been estimated and it appears that in dry weather the ducks are minor contributors to the pollution. During wet weather the numbers of faecal coliform bacteria and faecal streptococci rose markedly and at many sites exceeded the 2000

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2. INTRODUCTION

2.1 Plan and Scope of Present Work

In recent years water pollution has been recognised as a part of the critical problem of water resource management. To provide an adequate supply of water of the required quality has become one of our greatest natural resource problems.

By disposing of pollutants into bodies of natural water the potential uses of the water are reduced. The limits of the pollution load in streams and rivers are determined by a number of factors, such as the oxygen balance, which, if it is allowed to become depleted may lead to the death of the aerobic biota, including fish.

The city of Christchurch is drained by two rivers, both of which have considerable recreational and aesthetic potential. The Avon River flows through a largely residential area. It is used for recreation, including fishing, swimming, canoeing, rowing, whitebaiting, and also for storm-water drainage. The second, the Heathcote, passes through agricultural land close to the Port Hills, but because of industrial pollution in its lower reaches, it has been of less value for recreation since the use of a waterway for contact recreation requires water of a high quality.

In the preliminary classification (1974) of Natural Waters in the area administered by the North Canterbury Regional Water Board, the Avon River has been classified Class D, which is the lowest standard for water to be used for general recreation, agriculture and for general industrial water supplies. This is the minimum quality for all fresh waters. Possibly the river was given this low classification because it was considered that owing to pollution it could not be maintained at a

sufficiently high standard to allow for swimming and other forms of recreational use where the water may be swallowed. Unfortunately, the sources of this pollution have not yet been identified, and it may have been precipitate to classify the Avon at such a low standard before a full investigation of the quality of its water was made. Once evidence has been collected concerning sources of pollution, and the causes determined it may be possible to raise the quality of the water to such a standard that swimming in it can be recommended. In New Zealand the classification of waters is based upon their end use. Class A and B waters are intended for water supply purposes, for both public and industrial use. Class C water defines the minimum standard for primary contact recreational use, such as swimming, and this class of water is based upon a number of parameters, including pH, oxygen content, and bacterial numbers as well as colour and the absence of toxic compounds. Class B water is also defined by its pH and oxygen content, but the limits set are wider than in Class C water. However, for this class of water there is no bacteriological standard.

The danger of polluted water comes largely from living organisms, only infrequently from dead organic matter or toxic compounds. It is rare for ill-health from consumption of water to be due to an excess of an inorganic salt or a metal, such as lead, in the water.

Chemical analyses give valuable information on past or remote pollution but do not reveal all forms of recent pollution. Bacteriological examinations give less information on the remote history of the water but do show recent pollution with greater reliability than chemical analyses (Holden 1970).

Recent contamination by human or animal excrement can be the greatest danger associated with water that may be ingested by man. If faecal contamination has occurred the water may contain living pathogenic bacteria such as those that cause cholera, typhoid fever and dysentery. It is

possible to detect these pathogenic bacteria in water, but since they are often present in very low numbers and may only be present for short periods the laborious techniques and expensive equipment required for that isolation and identification makes their routine isolation impracticable.

However, pathogenic organisms in faeces or sewage are almost always outnumbered by the normal excremental organisms which are easier to detect in water and may be used to indicate faecal pollution. The organisms most commonly chosen as indicators of faecal pollution are the coliform group of bacteria.

The coliform group, however, includes a number of species of bacteria, some of which may have their origins in other than in faecal material. One member of the coliform group, Escherichia coli, is of undoubted faecal origin being restricted in its habitat to the gut of warm blooded animals. Its use as an indicator for detecting animal faeces in polluted waters gives a precise estimate of water quality (W.H.O., 1971).

The occurrence and density of coliform bacteria has been used both in New Zealand and overseas countries in determining standards of water quality and hence the use to which the water may be put. In New Zealand the water classification scheme sets a median value for coliform bacteria based on not fewer than 5 samples taken over not more than a 30-day period. But some American States include a criterion that not more than 10% of the samples shall exceed a specified upper limit.

One of the weaknesses of testing for coliform bacteria is that it is not possible to distinguish whether or not the sources of the isolates are human or non-human. Another group of bacteria used as indicators are the faecal streptococci which, like E. coli, are natural inhabitants of the gut and are present in large numbers in human and animal excreta. This bacterium has

also been chosen as an indicator because it too is not naturally found in water. nor does it normally multiply in water.

Recent work (Geldreich, 1967; Geldreich and Kenner, 1969) reported that there is a correlation between faecal coliform and faecal streptococcal numbers and this ratio gives a reliable guide for differentiating between human and non-human faecal contamination.

Their findings showed that faecal streptococci densities were significantly higher than faecal coliform densities in all warm-blooded animal faeces examined except in those of humans.

Previous work on the Avon River has shown high levels of bacterial pollution due to faecal contamination. From the results of chemical and bacteriological examination of water samples Hogan and Wilkinson (1959) compared the effects of season, tide and different river usage on the pollution and hydrography of both the Avon and the Heathcote Rivers.

Since 1959 there have been changes in the inputs into the river. Effluent from trade waste has almost completely disappeared but with the increased development of residential areas, particularly in the headwaters, there has been an increase in stormwater discharge.

The Christchurch Drainage Board, the North Canterbury Catchment Board, and the Christchurch City Council all carry out water sampling for bacteriological and other testing but there has been no regular monitoring of the river.

There is no data on the contribution of any source to the pollution of the water.

In this study the first experiments were designed to establish levels of contamination over the entire river for dry weather, when numbers would be lowest. Earlier work has not established baseline levels over a period of very low rainfall.

Following the collection of this baseline data the contributions of various sources to the faecal pollution would be examined.

During low flow the inputs of bacteria would be from wildlife, leaks in the sewage system which could contribute contamination from human sources, and from bacteria on the sediment moving into the water above.

From Hogan and Wilkinson's (1959) data it appears that numbers of bacteria increase rapidly a short distance from the source, and a study of the inputs and levels of bacteria in the upper reaches may provide information on sources of microbial pollution.

During periods of rainfall there would be additional inputs from surface runoff. (See Table 1) As there may be increased numbers of micro-organisms from disturbed sediment during high flows, therefore, it would be necessary to collect samples and enumerate bacteria present in the undisturbed stream sediment.

This would enable estimates to be made both of the contribution of bacteria from the sediment to the numbers of bacteria in the water during high flow, and the numbers of bacteria which may be lost from the water during low flow by adsorption into the sediment.

The use of faecal coliform bacteria as a parameter for monitoring recreational water quality must ultimately be related to the probable occurrence of waterborne pathogens. Salmonella is a pathogen which can be detected with the use of procedures adaptable to field studies, and many stream pollution studies include Salmonella detection as a supplementary bacteriological procedure as the presence of the pathogen implies a direct health hazard.

However the presence of pathogens, such as Salmonella, in water is highly variable and the inability to detect Salmonella in cases of faecal pollution when faecal coliform densities are above several hundred per 100ml does not imply poor correlation with the faecal coliform test.

The absence of Salmonella does not necessarily indicate the absence of other waterborne diseases as the presence or absence of other pathogens will not necessarily correlate with Salmonella occurrence.

Table 1. Quality and Characteristic of Contaminants
found on Street Surfaces (after Sartor et al, 1974)

Measured Constituents	Weighted means for all samples g/curb km
Total Solids	394,800
BOD ₅	3,807
COD	26,790
Volatile solids	28,200
Phosphates	310
Nitrates	26.5
Total N	620
Heavy Metals Zn	183.3
Cu	56.4
Pb	160.7
Ni	14.1
Hg	20.6
Cr	31.0
Pesticides -- p,p - DDD	0.019
p,p - DDT	0.017
Dieldrin	0.007
PCB	3.13
Total Coliform bacteria	6.2×10^{10}
Faecal Coliform bacteria	3.5×10^9

The survival time of the bacteria in the water could be estimated by field experiments which maintain samples of bacteria in the water isolated from other bacteria in the water but in a situation as close as possible to their natural habitat. By knowing the length of time bacteria spend in the river before they die, the inputs of bacteria at different points in the river could be established.

Using the results from these experiments together with information collected by the Christchurch Drainage Board on river flows and sediment loads, as well as numbers of ducks (Smith 1972), the contribution to the total pollution has been estimated. From this evaluation the feasibility of reducing numbers of bacteria in the river and the need for further work can be considered, with respect to the uses of the river.

2.2 DESCRIPTION OF THE AVON RIVER

Topography and Geography

The Avon River drains a catchment area of 83.7 km (Fig. 1).

It rises from springs in the Waimairi County and flows 26 km through the city of Christchurch to an estuary. Before European settlement the river had many small tributaries which drained the surrounding low-lying swamp land. Many of these swamps and their drainage streams have disappeared as the area has been developed, leaving dry watercourses which may be used as stormwater channels to cope with the increased stormwater runoff caused by close settlement.

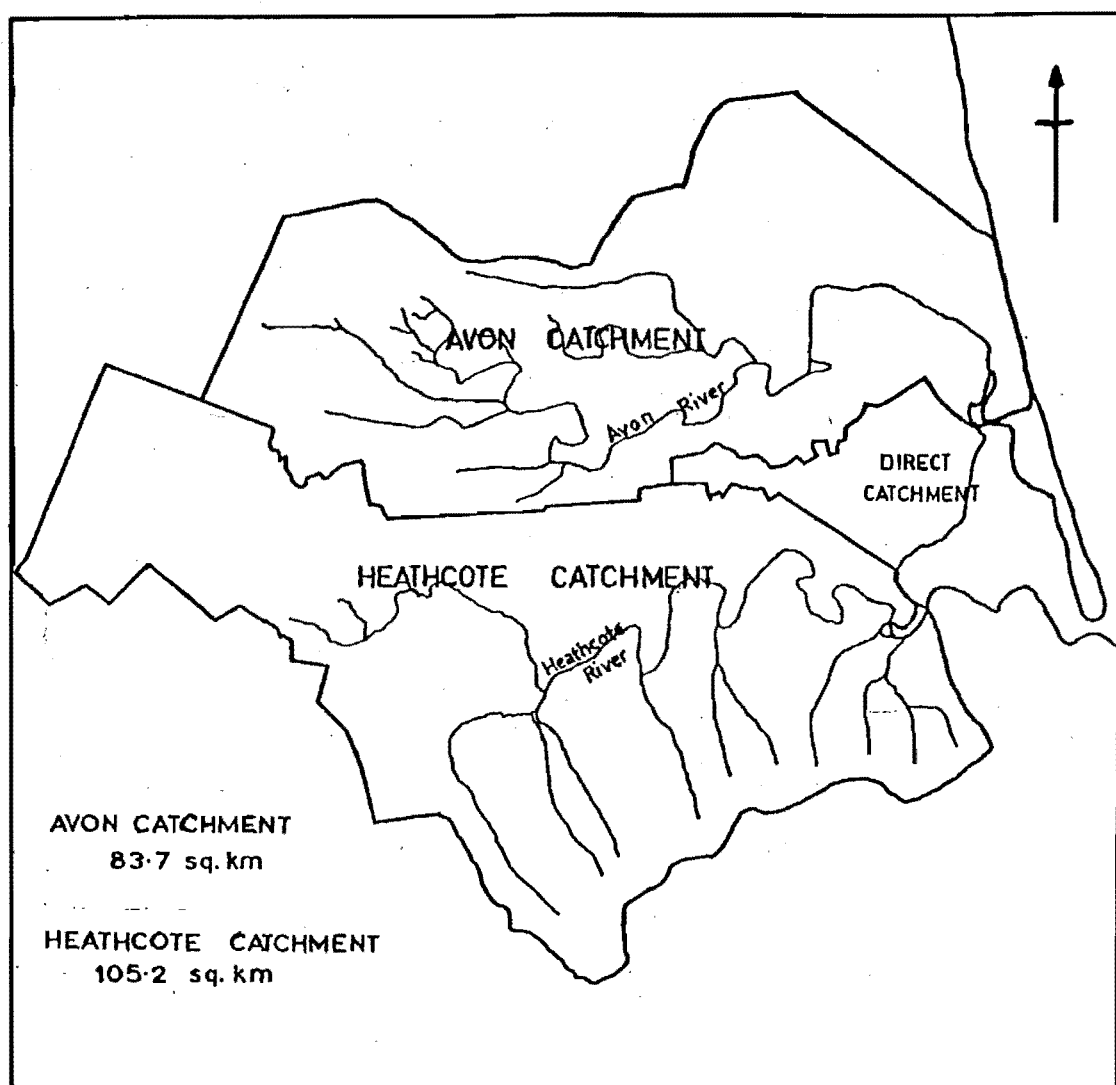
The headwaters of the Avon River now consist of 3 major tributaries. These are the Wairarapa Stream (6 km), the Waimairi Stream (5 km) and a southern stream known as the Avon River (South Branch) or the Avon tributary. Each of these streams arises from several springs which have a constant flow. The flows from the springs soon turn into small moderately fast streams with beds of mud or clay and patches of weed (Marshall, 1973). Woods quoted by Marshall (1973), suggested a common source from the aquifers in the fluvial Waimakariri shingles which when they come into contact with overlying and less permeable marine deposited clays and sands of the East Coast, may give rise to springs of the rheocrene and helocrene types.

These spring types are defined following Noel (1954):

Rheocrene: arise as torrents and flow rapidly away; they do not form pools.

Helocrene: arise from seepage in boggy or marshy areas; no well defined vent.

Fig. 1. Catchments of the Avon and Heathcote Rivers



The tributaries flow through areas which are mainly residential and converge a short distance upstream from the Fendalton Road bridge (Fig. 2).

From this confluence the Avon follows a meandering course round and through Hagley Park, the central city area and the north east and eastern residential suburbs of Christchurch. Tidal influence extends a considerable distance up the river. Flood tide estuarine (saline) and river (fresh) water mixing extends up the Avon River as far as the Wainoni Road Bridge (Fig. 2). This is 8 km from the estuary mouth. However, tidal effects extend beyond this point. According to Mawson (1972) the limit of tidal influence in the Avon River lies near the Barbadoes Street bridge.

Two drains, the Riccarton main drain and the Addington drain, enter the Avon a short distance upstream from the Antigua Boatsheds. Between Fitzgerald Avenue and Avondale Road, two other streams enter the Avon. Dudley Creek (10 km) which has its origin about Greers Road, near Harewood Road, passes through the Papanui industrial area and then through residential areas. The other stream is the outlet of Horse Shoe Lake, which joins the Avon just downstream from Kerrs Reach. A number of streams which drain the peat lands of Marshlands flow into Horse Shoe Lake. Below Bridge Street the Avon enters the northern corner of the Avon-Heathcote estuary, which is almost the shape of an equilateral triangle and 8 sq. km in area. The estuary is divided into two regions, an area of sandflats and an extensive area of mudflats. The wide mudflats are completely uncovered at low tide except for the main channels of the two rivers and a few areas of standing water. These are dissected by meandering tunnels

FIGURE 2:

SAMPLING SITES AND SITE NUMBERS ON THE AVON RIVER AND TRIBUTARIES

Site Number	Site
1	Avonhead Road bridge
2	Athol Terrace bridge
3	Waimairi Road bridge
4	Ilam Road bridge
5	Clyde Road bridge
6	Idris Road bridge
7	Straven Road bridge (near Royds Street)
8	Straven Road bridge (near Te Kura Place)
9	Fendalton Road bridge
10	Antigua Boatsheds bridge
11	Colombo Street bridge
12	Fitzgerald Avenue bridge
13	Avondale Road bridge
14	Pages Road bridge
15	Hereford Street bridge
16	Wainoni Road bridge

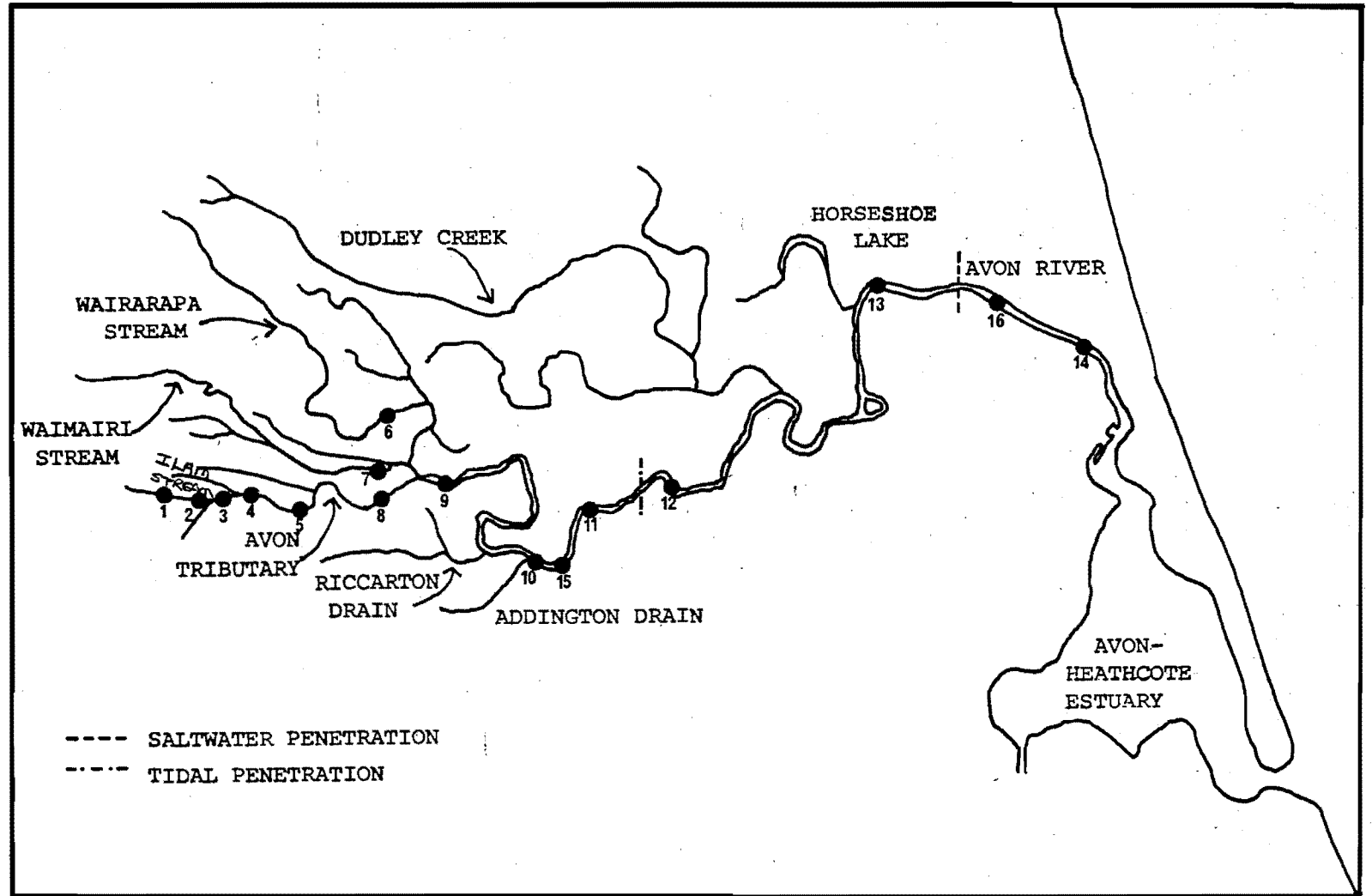
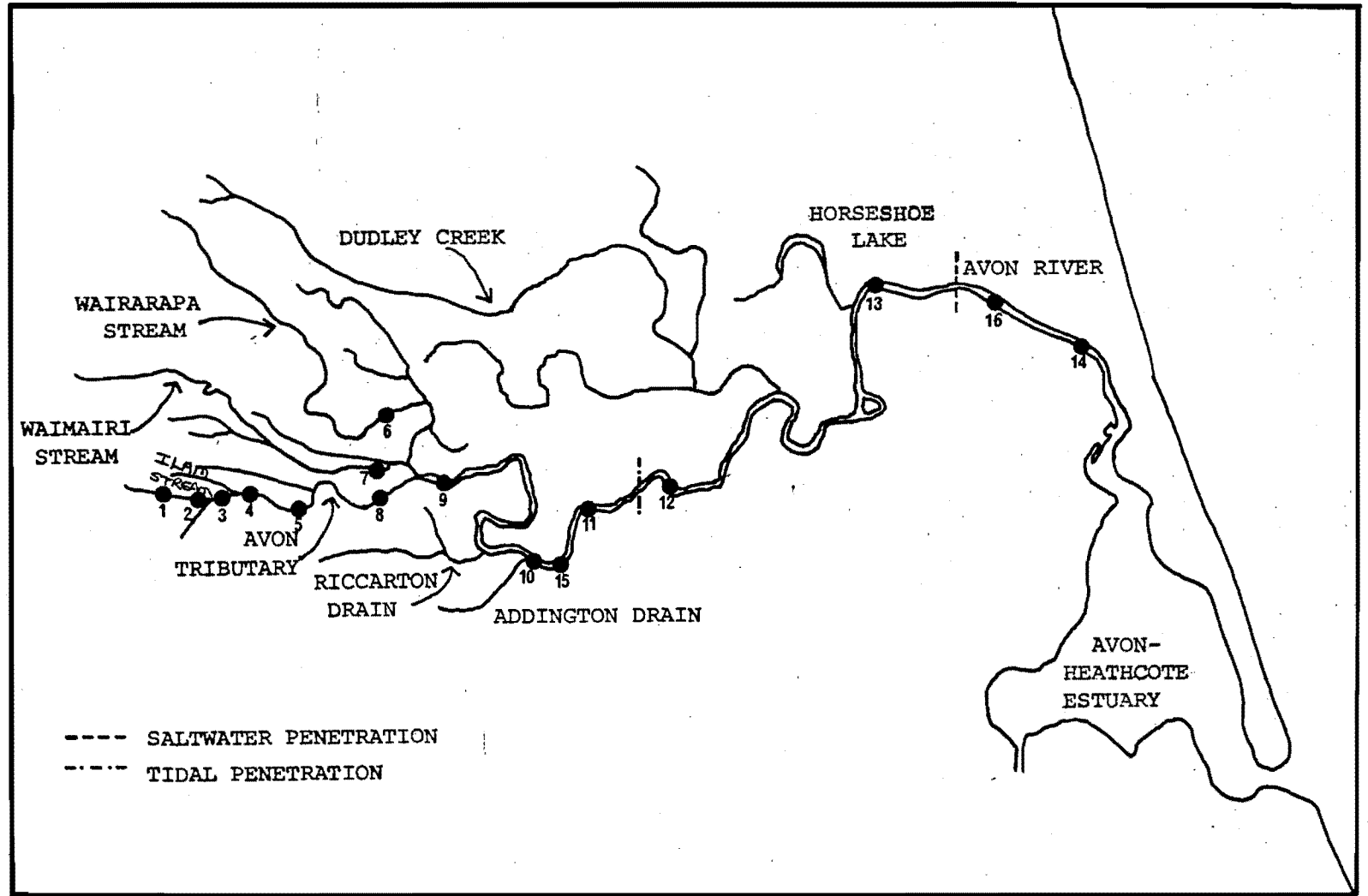


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10	Antigua Boatsheds bridge
11	Colombo Street bridge
12	Fitzgerald Avenue bridge
13	Avondale Road bridge
14	Pages Road bridge
15	Hereford Street bridge
16	Wainoni Road bridge



which generally lie at right angles to the main channels. (Knox and Kilner, 1973).

Hydrology

River Flow

Variations in river flow are determined largely by rainfall patterns and by the occurrence, frequency and intensity of "storms" accompanied by high rainfall. Christchurch is in a relatively low rainfall area, the average annual rainfall being 670 mm. This rainfall is fairly evenly distributed throughout the year but summer droughts may occur.

Knox (1973) reports that from data collected at gauging stations on the river and on a number of creeks and drains the average daily flow has been computed and is given by Mawson (1972) as $3.25 \text{ m}^3/\text{s}$ for the Avon River. For the Avon River, Greenland (1972) has estimated the base flow above the tidal reach as $1.83 \text{ m}^3/\text{s}$, and the total dry weather discharge from Horse Shoe Lake, Travis Swamp and Dudley Creek as $0.34 \text{ m}^3/\text{s}$. With an annual average rainfall of 670 mm for the catchment the rainfall runoff would give an annual average discharge of $3.19 \text{ m}^3/\text{s}$, close to Mawson's value of $3.26 \text{ m}^3/\text{s}$.

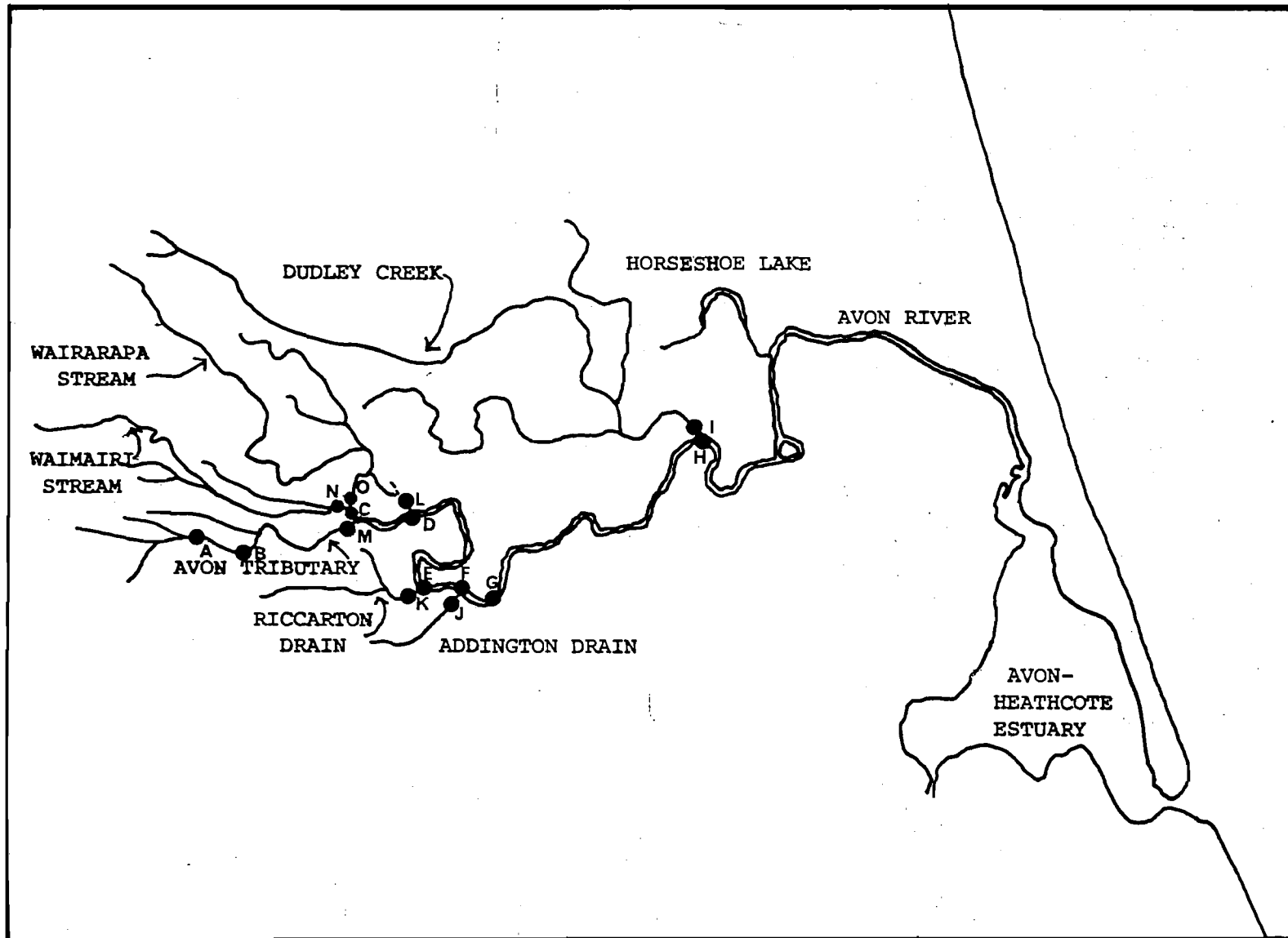
These average values can be greatly exceeded. There are few reliable measurements of past flood flows. However, in the 1968 flood a flow of $11.32 \text{ m}^3/\text{s}$ was recorded for the Avon. Using rainfall records, and recognised statistical methods of determining probable flood flows, Mawson had estimated (Anon., 1973a) a peak discharge at the river mouth of over $52.9 \text{ m}^3/\text{s}$ for a hypothetical 50 year storm.

Values collected by the Christchurch Drainage Board for dry weather flows in the river and tributaries are shown in Figure 3.

DRY WEATHER FLOWS IN THE TRIBUTARIES, DRAINS AND MAIN STREAM OF THE AVON RIVER.

DATA FROM CHRISTCHURCH DRAINAGE BOARD, INVESTIGATIONS DEPARTMENT, "DRY WEATHER FLOWS IN RIVERS AND DRAINS" (1 FEBRUARY 1973).

Site	Flow (m^3/s)
A	0.27
B	0.34
C	1.922
D	1.936
E	2.040
F	2.088
G	2.27
H	2.278
I	0.19
J	0.048
K	0.104
L	0.014
M	0.77
N	0.607
O	0.545



According to the Investigations Department, Christchurch Drainage Board (Anon., 1973b), the variation between winter low flow and summer low flow does not appear to be significant compared to total flow for most gauging sites.

Stormwater and Sewage Disposal

The swamps on which Christchurch is built have virtually gone, but there is no doubt that the site was a particularly difficult one from a drainage and sewerage viewpoint. Most of Christchurch is very flat; the elevation of Cathedral Square is less than 6.0 m above sea level.

There are many piped stormwater drains, some of them very long, discharging into the Avon. Their carrying capacity depends largely on the hydraulic gradients which are determined by the levels of their outlets. These, for greatest efficiency, should not be submerged. This is often not the case and water may back up in the drains causing local flooding of streets and properties.

The flat topography has resulted in a flat sewage reticulation system and forced continued reliance on many pumping stations to move sewage and trade wastes across the city to the sewage works at Bromley. These sewage works at Bromley now discharge treated effluent into the Estuary only on the ebb tide, in an attempt to prevent the transport of the effluent up the Avon and Heathcote Rivers.

DRY WEATHER FLOWS IN THE TRIBUTARIES, DRAINS AND MAIN STREAM OF THE AVON RIVER.

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L	0.014
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N	0.607
O	0.545

Each sewage pumping station has, if possible, at least one overflow, for use in case of emergency to prevent sewage being discharged directly from gully traps on to private property or from sewer manholes or vents onto public streets. Most of the overflows are to the Avon and Heathcote rivers, but some go to streams and open drains, and a few to stormwater sewers (Scott, 1963).

3. MATERIALS AND METHODS

3.1. Sampling Sites

3.1.1 Water Samples

Three series of samplings were carried out. Each series was designed to consist of not less than 5 samples taken within 30 days as required by the water classification regulations.

To enable comparisons with earlier work the sites chosen for water samples included the eight sampling points used by Hogan & Wilkinson (1959), and one other at the Colombo Street bridge (Fig. 2 and Table 2). This gave samples from the entire river with more sampling stations where the river flowed through the commercial area, as this was where the contamination could be expected to be highest.

For the second and third series the sites at Pages Road and Avondale Road bridges were not used, and new sites in the headwaters of the river were chosen as it became apparent that there was extensive faecal contamination even in the upper reaches.

The first series of samples were collected between 29 November 1974 and 22 December 1974. Over this period there was less than 20 mm of rain, and no samples were collected during or directly following rainfall. These samples are referred to as the "dry weather" samples.

The second series of samples was collected between 16 January 1975 and 29 January 1975 during a period when 94 mm of rain fell, with 54 mm falling on 16 January and 18 mm falling on the 17 January. The samples collected over this period are referred to as the "wet weather" samples.

Table 2. Sampling sites on the Avon River and tributaries.

Site Number	Sampling Site	River or Stream
1	Avonhead Road Bridge	Avon tributary
2	Athol Terrace Bridge	"
3	Waimairi Road Bridge	"
4	Ilam Road Bridge	"
5	Clyde Road Bridge	"
6	Idris Road Bridge	Wairarapa Stream
7	Straven Road Bridge (near Royds Street)	Waimairi Stream
8	Straven Road Bridge (near Te Kura Place)	Avon tributary
9	Fendalton Road Bridge	Avon River
10	Antigua Boatsheds Bridge	"
11	Colombo Street Bridge	"
12	Fitzgerald Avenue Bridge	"
13	Avondale Road Bridge	"
14	Pages Road Bridge	"
15	Hereford Street Bridge	"

The third series of samples was collected between 14 January 1975 and 29 January 1975. These samples were taken at sites on the Avon tributary to study the apparently high levels of contamination in the headwaters of the River, and are referred to as the "headwater" sampling.

3.1.2 Sediment Samples

Sediment samples were taken as close as practicable to five of the sites used for water sampling. The sites chosen were at Fitzgerald Avenue, Colombo Street, Hereford Street, Antigua Boat Sheds, and Fendalton Road. Sites for sediment sampling were limited by depth of water and the flow of the river, which is sufficiently fast to prevent build-up of sediment in many places.

3.2 Sampling Methods

3.2.1 Water Samples

Samples were collected in 300 ml glass bottles which had been wrapped and sterilized at 121°C for 30 minutes. The bottles were fastened to a line and lowered into the river. The full sample bottles were placed in an insulated container containing ice and transported to the laboratory for analysis. Analyses were made within 8 hours of collection. In the tidal zone, samples were collected at low water to reduce the possibility of collecting water forced up from the estuary on the rising tide.

3.2.2 Sediment Samples

To collect sediment samples of known surface area probes were devised. These were glass tubes 1 m long with an internal diameter of 21 mm. Each tube was fitted with a rubber bung at one end. The tubes were wrapped in

aluminium foil and sterilized by autoclaving at 121°C for 30 minutes. Ten probes were made and two used at each site. Each probe was used to collect a number of samples (between 2 and 8) at various distances from the bank of the river. For each site there were, therefore, duplicate samples, each consisting of sediment from a series of probings across the river.

3.3 Bacteriological Procedures

Enumeration of Micro-organisms

There are numerous difficulties in the enumeration of micro-organisms and a number of different methods have been devised to cope with these problems.

The first cause of error lies in the size of the sample taken. In sampling the river 300 mls of water was collected to represent a flow which could have been many litres in volume. This small sample may or may not have been representative of the whole.

The next problem is how to evaluate the amount of pollution from the numbers and types of micro-organisms present.

3.3.1 Water Samples

There are two basic methods for the detection and enumeration of coliform bacteria and streptococci in water. One is the multiple tube method ("The Bacteriological Examination of Water Supplies", Anon, 1969, Standard Methods, A.P.H.A. 1971; W.H.O. 1971) in which measured volumes of water are first added to a suitable liquid medium and incubated. After the appropriate incubation time at a specified temperature the inoculated tubes are examined for the standard reaction given by the organism being tested for. Because false positive reactions may occur due to the presence

of some other organisms this type of test is called presumptive. A presumptive test may be followed by confirmatory tests for coliform organisms and subsequently by further tests E. coli may be identified. By the inoculation of suitable volumes of water into a number of tubes an estimate of the number of presumptive coliform organisms present in a given volume of water can be obtained from statistical tables. This is called the "multiple tube" or "most probable number" (MPN) method for enumerating bacteria, and the result is expressed as numbers per 100 ml of the original water sample.

Faecal streptococci determination by the multiple tube method involves inoculation of glucose azide broth with aliquots of the water to be tested.

Tubes which give a positive result for this test may be subcultured into further tubes of the same broth and incubated at an elevated temperature. Tubes which are positive after the second incubation confirmed the presence of faecal streptococci.

The alternative method of counting coliform organisms in water is the membrane filtration method ("The Bacteriological Examination of Water Supplies", Anon 1969; Standard Methods, 1971; W.H.O. 1971.). This method which is recognised in England, U.S.A. and many other countries is widely used because of its simplicity and speed. It does not give results strictly comparable to the multiple tube method, and water samples with a high degree of particulate matter such as clay, colloidal iron or algae, cannot be examined by the membrane filter technique, because the membranes become blocked before the water has filtered through.

The membrane filter method is carried out by drawing a measured volume of the sample through a membrane composed of cellulose esters or certain other substances. All bacteria present in the water sample are retained on the surface of the membrane, and by incubating the membrane face-upward on suitable medium at an appropriate temperature, visible bacterial colonies are able to grow on the surface of the membrane. These colonies are counted and give the presumptive coliform count, or the so-called direct "E. coli" count, depending on the enrichment medium used. Direct counts of faecal streptococci may be made using a membrane filter and a suitable medium.

Counts on membranes are, however, subject to statistical variations and replicate counts of the same sample may not show the same number of organisms. The confidence levels for this type of test are given in "The Bacteriological Examination of Water Supplies" (Anon, 1969). The membrane filter method has been found to have a high degree of reproducibility and large sample volumes may be examined. Because of this, and because results are rapidly obtained, generally within 24 hours (Geldreich, 1966), this method was chosen to enumerate faecal coliforms and faecal streptococci in the water samples taken in this study.

Filtration Apparatus and Outline of Technique

The filtration apparatus used consisted of a porous carbon or sintered glass disc supported in silicone rubber gaskets fitted in a base to which a cylindrical funnel graduated at 50 ml and 100 ml was clamped. The membrane filter is supported on the porous disc. For filtration, the filter holding assembly is mounted on a flask with a slide arm which can be connected to a suction pump. After a measured volume of water has been filtered through the membrane under suction, it is removed and placed, face upwards, on either a suitable solid medium or a pad soaked in a liquid medium. The

filter-holding assembly is sterilized before use. Two cylindrical funnels are used with each filter holder; after each sample the funnel is rinsed with sterile water and placed in boiling water while the other funnel is used for the next sample. (W.H.O. 1971).

In this study the measured volumes of the sample used were 10.0 ml, 1.0 ml and 0.1 ml. (For volumes less than 10.0 ml the sample was diluted with sterile water so that a minimum of 10.0 ml was filtered). These samples were filtered through sterilized membrane filters (Oxoid, 5 cm, 0.45 μ m pore diameter.)

Enumeration of Faecal Coliforms

Membrane filters obtained from the membrane-filter technique (outlined above) were incubated on filter pads (Whatman Grade 17) which had been soaked with 2.0 ml M-FC broth (Difco) and incubated at $44.5 \pm 0.2^{\circ}\text{C}$ for 24 hours (Standard Methods, 1971).

Enumeration of Faecal Streptococci

Faecal streptococci were counted on M-enterococcus agar (Difco) and incubated at 37°C for four hours, then $44-45^{\circ}\text{C}$ for 44 hours ("The Bacteriological Examination of Water Supplies", Anon, 1969).

3.3.2 Sediment Samples

The numbers of coliform bacteria and faecal streptococci were assessed by the M.P.N. Method. ("The Bacteriological Examination of Water Supplies", Anon, 1969), because the fine particles in the sediment quickly blocked the membrane filters.

Preparation of Media

Coliforms:

(i) Minerals modified glutamate medium (MMG). Dehydrated powder of MMG (Oxoid) was added to 1 l of water containing 2.5 g ammonium chloride powder. This was dispensed into test-tubes and sterilized at 116°C for 10 mins.

(ii) Brilliant Green Bile broth (BGBB). Forty g of BGBB (BBL) was added to 1 l of water, dispensed in tubes with Durham tubes, and auto-claved at 121°C for 15 mins.

(iii) Peptone water for indole reaction

Tryptone (Oxoid)	20 g
NaCl (Analar)	5 g
Water	1000 ml

Sterilized in 5 ml aliquots at 121°C for 15 mins.

(iv) Kovacs Reagent

Paradimethyl amino-benzaldehyde (Analar)	5 g
Amyl alcohol	75 ml
Concentrated hydrochloric acid (Analar)	25 ml

Streptococci:

(i) Glucose azide medium (single strength).

Polypeptone (BBL)	10 g
NaCl (Analar)	5 g
Potassium phosphate K_2HPO_4 (M & B)	5 g
Potassium dihydrogen phosphate KH_2PO_4	2 g

Glucose (Analar)	5 g
Yeast extract (Oxoid)	3 g
Sodium azide (Hopkins & Williams Ltd)	0.25 g
Bromocresol purple 1.6% solution in 2 ml of absolute alcohol	

Above ingredients were dissolved in 1 l water, dispensed and autoclaved at 121°C for 15 mins. Double strength medium was prepared by doubling the weight of ingredients per litre of water.

Preparation of dilutions:

Dilutions were prepared according to standard bacteriological techniques. The jars containing the samples were shaken vigorously to mix the sample. From the liquid over the sediment sample 3x1 ml aliquots were pipetted into 99 ml blanks of sterile water.

These were then distributed as follows:

Coliforms

5 tubes containing 10 ml of double strength MMG inoculated with 10 ml	
" " " 5 ml " single " " " " 1 ml	
" " " 5 ml " " " " " 0.1 ml	

Streptococci

5 tubes containing 10 ml of double strength GAB inoculated with 10 ml	
" " " 5 ml " single " " " " 1 ml	
" " " 5 ml " " " " " 0.1 ml	

Incubation and Further Inoculation (Confirmed Test)

Coliform Bacteria

Tubes of MMG were incubated at 37°C for 48 hr but were not checked at the end of 24 h as recommended in British Ministry of Health and Local Government (1969), because insufficient time was available.

Tubes giving a positive reaction (yellow colour and gas formation) were inoculated into 5 ml BGGB and incubated at 37°C for another 48 h. Because of a clay layer in the bottom of some of the MMG tubes gas production was not always able to be detected in the Durham tube and so any tube which produced acid was inoculated into BGGB. Positive tubes of BGGB were labelled as "Confirmed Coliforms" and were reinoculated into 5 ml amounts of BGGB and tryptone and incubated for 24 h at 44.5°C. Tubes containing tryptone were tested for indole production with Kovac's reagent and the BGGB tubes were observed for gas production.

Streptococci

Tubes were incubated at 37°C for 72 h and those which showed a yellowing of the medium were considered positive. These were inoculated into 5 ml aliquots of single strength GAB and incubated at 45°C for 48 h. Again tubes showing a yellow colour were taken as positive.

3.3.3 Rates of die-off of Bacteria in the River

To measure the survival of bacteria in the river field experiments were set up. Sections of dialysis tubing, tubular cellophane material about 30 mm in diameter and 250 mm long, were tied at one end and filled with river water, the other end being closed with a rubber band to make what

can best be described as a "water sausage". The dialysis membrane was made of regenerated cellulose by the Visking process. This type of membrane will retain the bacteria but permits the passage of water and small molecules such as sugars and salts.

The filled tubes were suspended in the river a few centimetres below the surface so that the bacteria contained in them were in almost the same ecological condition as they would have been if they had been left in the stream. Ten ml samples were removed from the tubes at regular intervals and numbers of faecal coliforms and faecal streptococci enumerated by the membrane filter technique.

3.3.4 Salmonella Detection

The sites chosen were nine of those used for water sampling (sites 6,7,8,9,10,11,12,13,14).

There are many different techniques for isolating Salmonella. However one method which has proven to be successful for isolating Salmonella from river water is one which uses cotton swabs. (Spino, 1966). Swabs were prepared using the method of Spino (1966). Gauze cloth 23cm wide was folded into 5 layers of 36cm lengths. This pad (23x36 cm) was then cut across the folds at one end leaving a pad measuring 23x33cm. The pad was then cut lengthwise from the cut end into 5 strips of 4.5cm to within 10cm of one end. The uncut end was then securely wrapped with 16 gauge wire and placed just below the surface of the water. The swabs were attached under the bridges in the flow of the water, and after seven days in the water the swabs were retrieved from all sites except 13 and placed in plastic bags in an insulated container containing ice.

Two sets of enrichment media (Tetrathionate Enrichment Broth (BBL) and Selenite-F Broth (BBL)) were prepared with 300mls of broth in 500ml wide-necked flasks. Strips were cut from the swabs using scissors that were dipped in ethyl alcohol and flamed, and added to the flasks. Half the swab was used for each medium. The flasks were then incubated in a dry-air waterjacket incubator at 37⁰ C for 24 hours (Edwards and Ewing, 1972).

After incubation of the enrichment broth for 24 hours, plates of various differential agars were streaked from the enrichment media. The media chosen were two moderately selective ones (Salmonella Shigella (SS) (BBL), and Hektoen Enteric (HE) (Difco)) and two highly selective media (Bismuth Sulphite Agar (BS) (Oxoid), and Brilliant Green Agar (BG) (Difco)).

Following the method described by Edwards and Ewing (1972) primary Differentiation of bacteria from selected colonies on the agar plates was carried out. Tubes of Triple Sugar Iron Agar (TSI) (Oxoid) and Lysine

an acid butt and an alkaline slant were planted immediately on the urea agar of Christensen (Edwards and Ewing, 1972). The cultures were examined after 2-4 hours, and the negative tubes reincubated. Cultures of Proteus produce marked alkalinity in Christensen's urea agar after a short period of incubation but Salmonella show negative or weak positive alkalinity after 24 hours incubation.

4. RESULTS AND DISCUSSION

4.1 NUMBERS OF BACTERIA IN AVON RIVER DURING CONDITIONS OF LOW FLOW

4.1.1 Observed Numbers

Mean values of the numbers of faecal coliform and streptococcal bacteria have been calculated and are presented in tables 3 and 4. Since there was variability due to factors such as river flow, time of day, oxygen status of the water and temperature, the geometric mean and standard deviation of the \log_{10} numbers for each site are presented in Appendix Figs. 1 and 2.

During low flows, with the exception of sites at Avondale Road Bridge and Pages Road Bridge, the mean numbers of faecal coliforms exceeded the limit of 200 faecal coliforms/100 ml required for Class C waters. However numbers do not exceed the 2000/100 ml specified for Class B waters.

Table 3. The Range of Values and the Means of Numbers of Faecal Coliform Bacteria per 100 ml for the Dry Weather Condition Sampling.

Site No.	Sampling Station	Range	Mean
6	Wairarapa Stream	120 - 1045	531
7	Waimairi Stream	140 - 800	351
8	Avon tributary	225 - 485	341
9	Fendalton Road	215 - 985	501
10	Antigua Boatsheds	430 - 1220	540
11	Colombo Street	100 - 4350	1571
12	Fitzgerald Avenue	400 - 1745	940
13	Avondale Road	0 - 195	167
14	Pages Road	85 - 220	176

Table 4. The range of Values and the Means of Numbers of Faecal Streptococci per 100 ml for the Dry Weather Condition Sampling.

Site No.	Sampling Station	Range	Mean
6	Wairarapa Stream	175 - 305	249
7	Waimairi Stream	150 - 375	230
8	Avon tributary	180 - 295	230
9	Fendalton Road	295 - 605	453
10	Antigua Boatsheds	135 - 1360	258
11	Colombo Street	210 - 290	236
12	Fitzgerald Avenue	210 - 455	291
13	Avondale Road	10 - 45	26
14	Pages Road	50 - 110	71

Because the concentration of faecal coliform bacteria and faecal streptococci in the water at the Straven Road and Idris Road bridges which are close to the origin of the river was found to be high, sampling was carried out from near the source of the river on four occasions. The results which are presented in Tables 5 and 6 (and Appendix Figs 3 and 4) show that the average number of faecal coliform bacteria are greater than 200/100 ml for all sites except at Avonhead Road, closest to the source. At four of the six sites however, there was at least one occasion on when there were fewer than 200 faecal coliforms per 100 ml.

Numbers of faecal streptococci were also high. The numbers obtained are not strictly comparable with the dry weather sampling period as they were collected at a later stage when there was rainfall (Fig. 7).

In Table 7 the number of faecal coliforms per 100 ml for the dry and wet weather samplings of the present survey are compared with numbers obtained in previous studies.

Table 5. The Range of Values and the Means for Numbers of Faecal Coliforms per 100 ml for Samples taken in the Avon Tributary.

Site No.	Sampling Station	Range	Mean
1	Avonhead Road	20 - 53	44
2	Athol Terrace	120 - 9200	418
3	Waimairi Road	177 - 11400	628
4	Ilam Road	180 - 11000	721
5	Clyde Road	259 - 11200	441
8	Straven Road	220 - 9000	719

Table 6. The Range of Values and the Means for Numbers of Faecal Streptococci per 100 ml for Samples taken in the Avon Tributary.

Site No.	Sampling Station	Range	Mean
1	Avonhead Road	100 - 1250	259
2	Athol Terrace	88 - 295	154
3	Waimairi Road	181 - 365	226
4	Ilam Road	172 - 460	299
5	Clyde Road	220 - 577	321
8	Straven Road	215 - 666	322

Table 7. Comparison of results from present study during dry and wet periods with values from Hogan & Wilkinson (1959) and results of a Catchment Board Sampling (23 January 1973). Hogan & Wilkinson (1959) and Catchment Board results are expressed as MPN per 100 ml. The results of the present study were obtained by the membrane filter method.

Site No.	Sampling Site	Faecal Coliform Organisms per 100 ml				Total Coliform Organisms per 100 ml	
		Hogan and Wilkinson	Catchment Board	Present Study Dry	Present Study Wet	Hogan and Wilkinson	Catchment Board
6	Wairarapa Stream at Idris Road	≈ 300	250	531	2,131	≈ 5,000	250
7	Waimairi Stream at Straven Road	≈ 100	700	351	1,623	≈ 5,000	700
8	Avon tributary at Straven Road	≈ 400	2,500	341	1,163	≈ 5,000	7,000
10	Antigua Boatsheds	≈ 100	700	540	1,060	≈ 10,000	>110,000
12	Fitzgerald Avenue	≈ 1,000	> 110,000	940	2,103	≈ 9,000	25,000
13	Avondale Road	≈ 200	25,000	167	-	≈ 8,000	7,000
14	Pages Road	≈ 800	7,000	176	-	≈ 9,000	-

During the dry weather period numbers of faecal coliform organisms were lower than those for the corresponding period in 1972, when they fluctuated between 500 and 1000/100 ml (Report, 1972). It must be noted however that the method used was the multiple tube procedure which may not be strictly comparable to the membrane filter technique. Some of the observed differences may be accounted for because of the dry December of 1974 when only 20 mm of rain fell. Because water is now released from Bromley Sewage works only on the ebb tide, reducing effluent flow up the river, this may have influenced the low values obtained in the present study for the Avondale and Pages Road sites. (It was noted (Report, 1973) that after one tidal flushing without discharge from the sewage treatment works numbers dropped to 140 faecal coliforms/100 ml at Avondale Road bridge.)

Numbers obtained in this survey are also much lower than those obtained by the Government analyst (Report, 1973a), who analysed samples taken approximately 3 hrs after high tide on 23 January 1973 by the Catchment Board.

Differences at the Pages Road and Avondale Road bridges may be explained by the differences in tide at times of collection, but the reasons for differences at the Fitzgerald Avenue bridge are more obscure. The similarity between E. coli and total coliform counts on the 23 January 1973 suggest very recent sewage pollution for the total length of the river. This is in contrast to the survey of the Estuary and work of Hogan (1959) (Table 7), where total coliform numbers were much in excess of E. coli numbers.

Comparison of Hogan & Wilkinson's 1959 data (Table 7) shows that although numbers along most of the river were higher in 1959 than in the present sampling, the counts were made throughout the year and would probably have included some storm water discharge. The comparison suggests that pollution has not increased since that date.

4.1.2 Rates of Purification in the Avon during Low Flows

Tables 3 and 4 show that a reduction in numbers of faecal coliforms and streptococci occurs between some sampling stations. The reduction could be due to dilution from the inflow of water or death or incorporation of the bacteria into the sediment. The actual decline in numbers observed between any two stations may be greater than that shown by the results, as there is no reason to believe that contamination from external sources does not take place all along the river. The rate of observed decline will therefore be minimal because contamination by more bacteria will probably be occurring at the same time.

If it is assumed that the disappearance of bacteria from the water between any station and the next can be described by an exponential function then

$$N_f = N_i e^{-\frac{t}{b}} \quad \text{Equation 1}$$

where N_f = final number of bacteria remaining after time t

N_i = initial number at time t_0

e = natural exponential

t = time (seconds) for river to flow between two sampling points at 1.645 s/m

b = time constant, i.e. the time taken for the number to decrease to 63% of the initial value

$$\therefore \frac{N_f}{N_i} = e^{\frac{-t}{b}}$$

$$\therefore \log_e \frac{N_f}{N_i} = \frac{-t}{b}$$

$$\therefore b = \frac{-t}{\log_e \frac{N_f}{N_i}}$$

Equation 2

To calculate b, the time constant, experiments were conducted to estimate the rate of die-off of bacteria from the water at various sites.

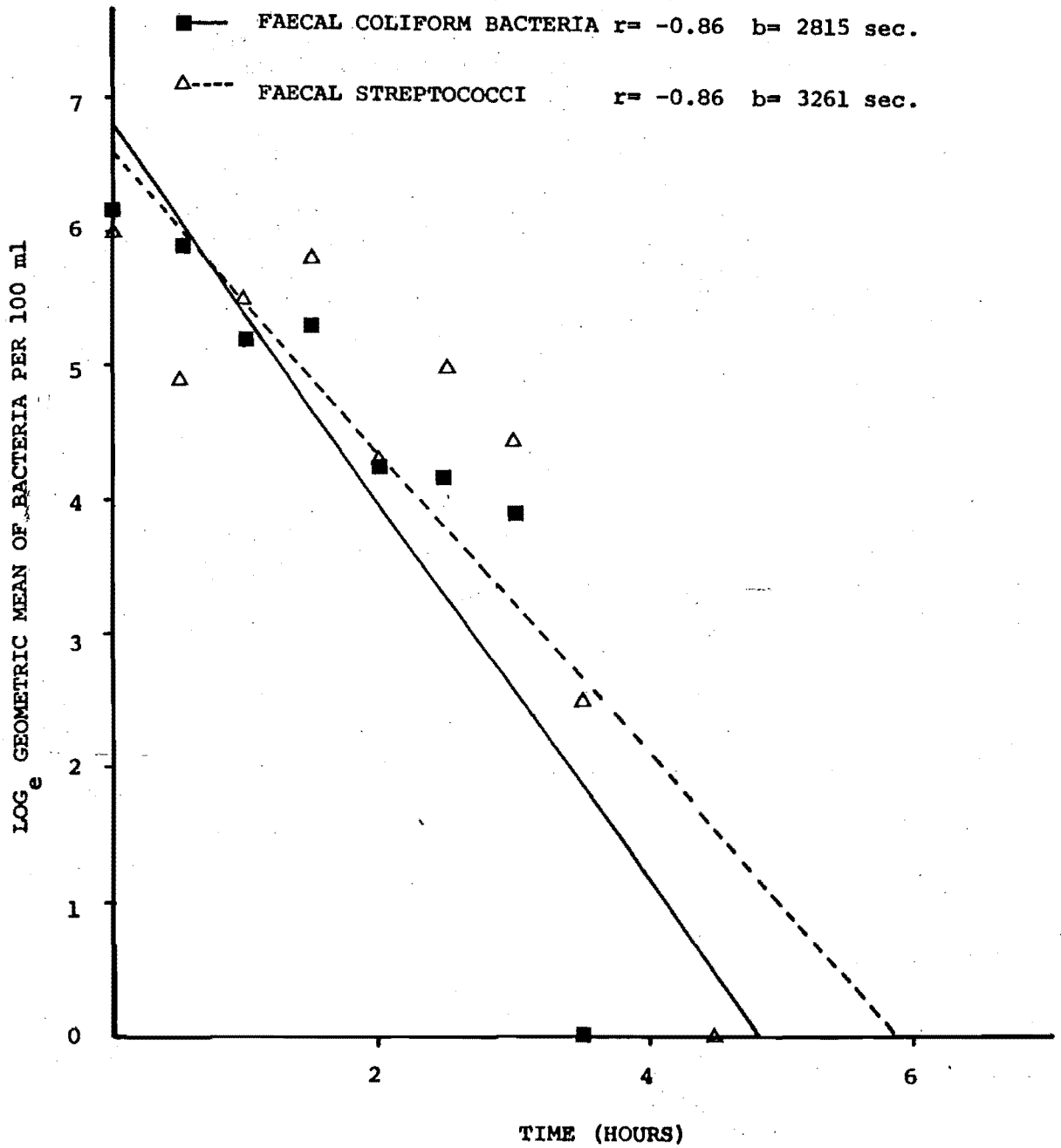
From regression analyses (see Fig. 4) of the data obtained in these experiments values of t, N_f , and N_i were obtained and were used to calculate b, according to equation 2. The values of b obtained are shown in Table 8. The data obtained from the analysis of the results of this experiment are "best fit" values.

The derived values of b were used in equation 1 to calculate the concentration to which the bacteria would decline during the time they took to pass from one station to the next. The difference between the numbers of bacteria observed at the second station and the number of those observed at the first station still remaining at the second station (N_f) must have been added to the system between the two stations. The value obtained will be a minimum because some of the bacteria added will have entered the water near the first station, and would have disappeared before reaching the second station.

FIGURE 4:

REGRESSION ANALYSES OF REDUCTION IN NUMBERS OF FAECAL COLIFORM BACTERIA
AND FAECAL STREPTOCOCCI CONTAINED IN DIALYSIS TUBING PLACED IN THE AVON
RIVER.

REDUCTION OF NUMBERS AT IDRIS ROAD



REDUCTION OF NUMBERS AT PALMERS ROAD

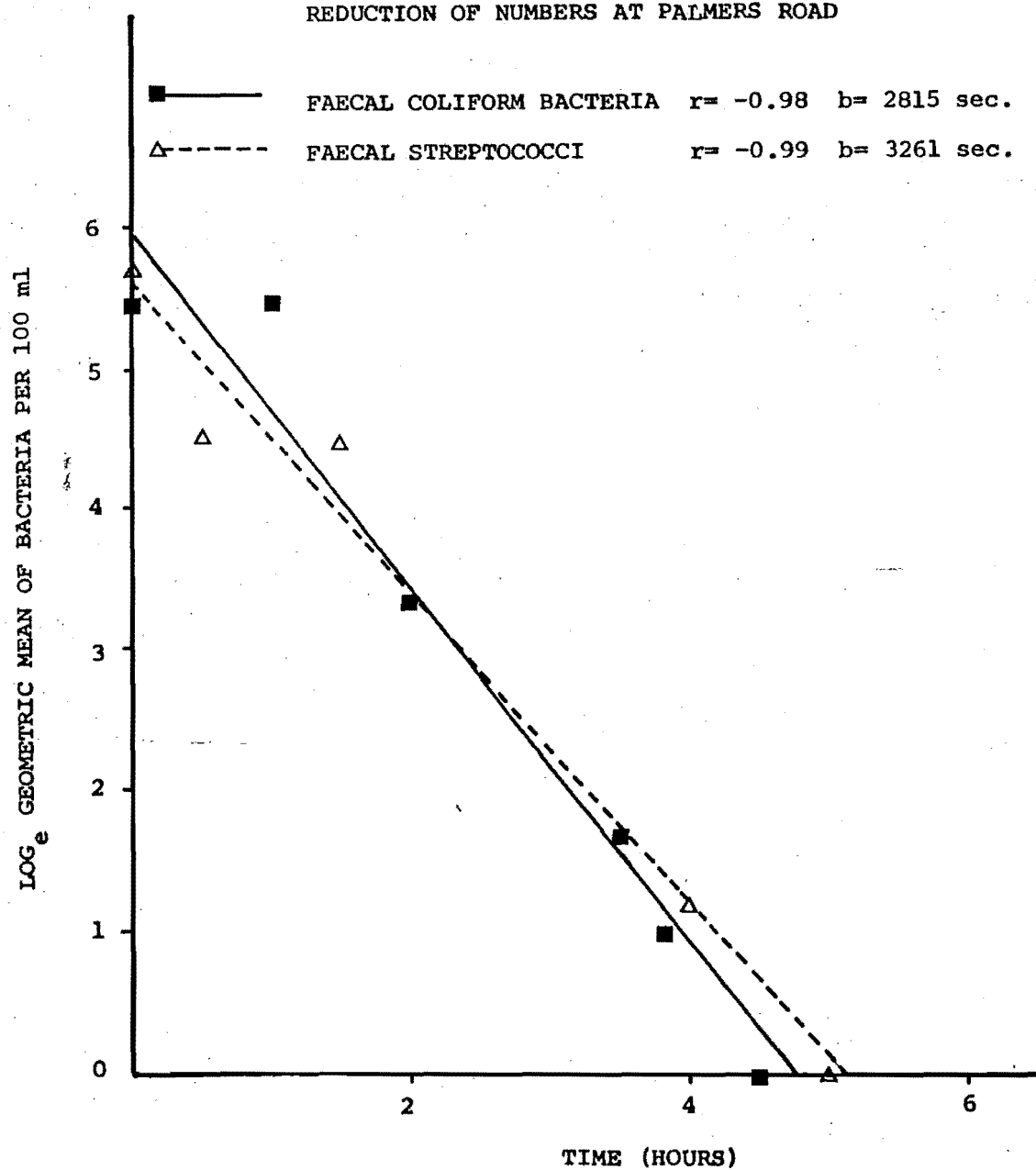


Table 8. Values of b, time constant, i.e. time taken for the number to decrease decrease to 63% of the initial value, for different sites on the Avon River.

Site	Indicator	b(secs)
Wairarapa Stream	{ faecal coliform bacteria	2630
at Idris Road	{ faecal streptococci	3231
Avon River at	{ faecal coliform bacteria	2815
Palmers Road (tidal)	{ faecal streptococci	3261

Table 8a. Flow (100ml/s) at sampling sites . (Flow data from Table 3.)

* estimated value.

Sampling Station	Flow (100ml/s)
1	1400 *
2	1500 *
3	2000 *
4	2700
5	3400
6	5450
8	7700
9	19,360
10	20,880
11	23,000 *
12	26,000 *
13	32,600
14	32,600

By assuming that the input is evenly distributed between the two sampling points and using the values of b calculated for the death rate, it is possible to calculate the total input of bacteria over a distance, D .

$$N_x = N_o - N_f$$

=(number observed at the second station) -

=(number of N_i remaining after some have died off during travel of time t seconds)

N_a = number of bacteria added over the time the river takes to flow between two stations. These die off and result in N_x bacteria by the time the river reaches the second station.

If q bacteria enter the stream per metre then over distance dx ($=dt$) the number entering is $N_a = q dx$. However these will die off at the above rate so will contribute $N_a e^{-\frac{x}{b}}$ bacteria at the second station. Hence over the total distance the total number added will be :

$$N_x = \int_0^D q e^{-\frac{x}{b}} dx$$

$$= -qb \left[e^{-\frac{x}{b}} \right]_0^D$$

$$= -qb \left[e^{-\frac{D}{b}} - 1 \right]$$

$$q = \frac{N_x}{b} \cdot \frac{1}{\left(e^{-\frac{D}{b}} - 1 \right)}$$

The numbers of faecal coliform bacteria and faecal streptococci entering the river per metre, and the numbers entering over each stretch have been calculated, and are shown in Figures 5 and 6. These figures give an idea of the amount of pollution occurring at each part of the river.

In the tributaries inputs of faecal coliforms and faecal streptococci were both high. One stretch did show an overall loss of bacteria but this could be due to insufficient sampling. The results showed that in dry weather the highest input per metre of faecal coliform bacteria occurred between Antigua Boatsheds and Colombo Street. Over this section the input of faecal coliform bacteria was over 10 times greater than the input of faecal streptococci. For this period of dry weather the highest input per metre of faecal streptococci was between Colombo Street and Fitzgerald Avenue. The input of faecal coliform bacteria, in both dry and wet weather, was considerably higher over the stretch of river between Antigua Boatsheds and Fitzgerald Avenue, than elsewhere in the river; whereas the numbers of faecal streptococci were high over a greater distance between Straven Road and Fitzgerald Avenue.

For the wet weather period most inputs per metre of both faecal coliform and faecal streptococci were at least twice those during dry weather. However as there is no data for flow rates during wet weather comparison can only be made using figures which assume that the flow in the river is uniform at all times in the entire river.

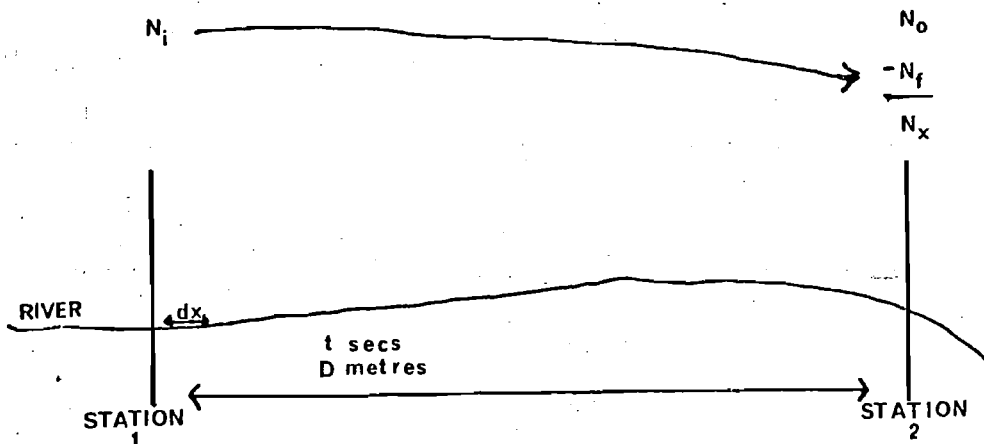
Using data for the average contribution of faecal coliform bacteria per capita from Geldreich (1966) it is estimated that one duck contributes 127,000 faecal coliform bacteria per second. From the figures for inputs of faecal coliform bacteria to the river in dry weather even at the highest level (36,394/m/s) this would be only one duck at each four metres of bank. From Smith (1972) it has been estimated that numbers of ducks near Hagley Park averaged about $2/100\text{m}^2$. Assuming the river to have an average width of 10m then this would give an estimated one duck for each five metres of bank. From this it would seem that this estimated number of ducks could cause most of the faecal coliform input in this section of the river. In other sections of the river where inputs are lower only a proportion of

duck faeces would need to be contributed to the input of faecal coliform bacteria to reach the calculated levels.

Geldreich (1966) also estimated that ducks contribute 203,330 faecal streptococci per second. The highest level of faecal streptococci recorded for dry weather was 4513/m/s which could be contributed by one duck in every 46 metres of bank. However it has been shown that possibly not all types of faecal streptococci found in ducks may grow on the medium used in this study as Middaugh et al (1971) have found that some types are inhibited.

SCHEMATIC REPRESENTATION OF RIVER WITH SYMBOLS USED IN CALCULATIONS

IN SECTION 4.1.2



4.1.3 Sources of Bacteria entering the River during Low Flows

Some idea of the source of the microbial pollution can be obtained by a consideration of the ratio of faecal coliform bacteria (FC) to faecal streptococci (FS).

KEY TO FIGURES 5 AND 6

N_f = number of bacteria remaining from N_i bacteria after time t .

N_i = initial number of bacteria at time t_o .

$$N_x = N_o - N_f$$

= (number of bacteria observed at the second station)

- (number of N_i bacteria remaining after some have died off during travel of time t (seconds)).

t = time for river to flow between two sampling points at 1.645 s/m.

q = number of bacteria entering the stream per metre per second using flow data from Table 8a.

q_a = number of bacteria entering the stream per metre per second into each 100mls of water assuming uniform flow over the whole river.

FIGURE 5:

NUMBERS OF FAECAL COLIFORM BACTERIA ENTERING THE RIVER PER METRE AND NUMBERS ENTERING OVER EACH STRETCH DURING DRY WEATHER SAMPLING PERIOD.

NOTE: Samples in the Avon tributary were not taken on the same occasions as samples in the Avon River.

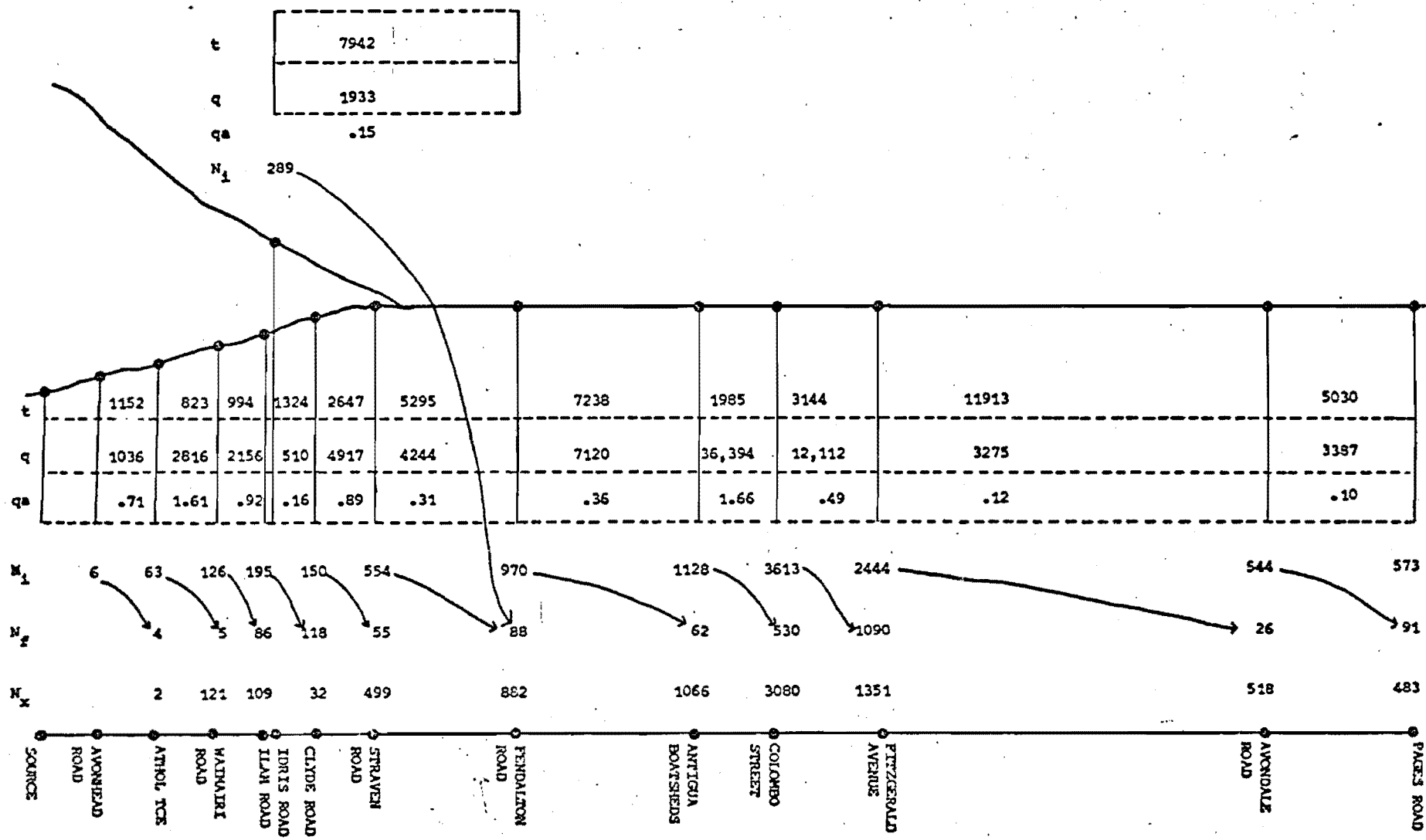


FIGURE 5a:

NUMBERS OF FAECAL COLIFORM BACTERIA ENTERING THE RIVER PER METRE AND NUMBERS ENTERING OVER EACH STRETCH FOR WET WEATHER SAMPLING PERIOD.

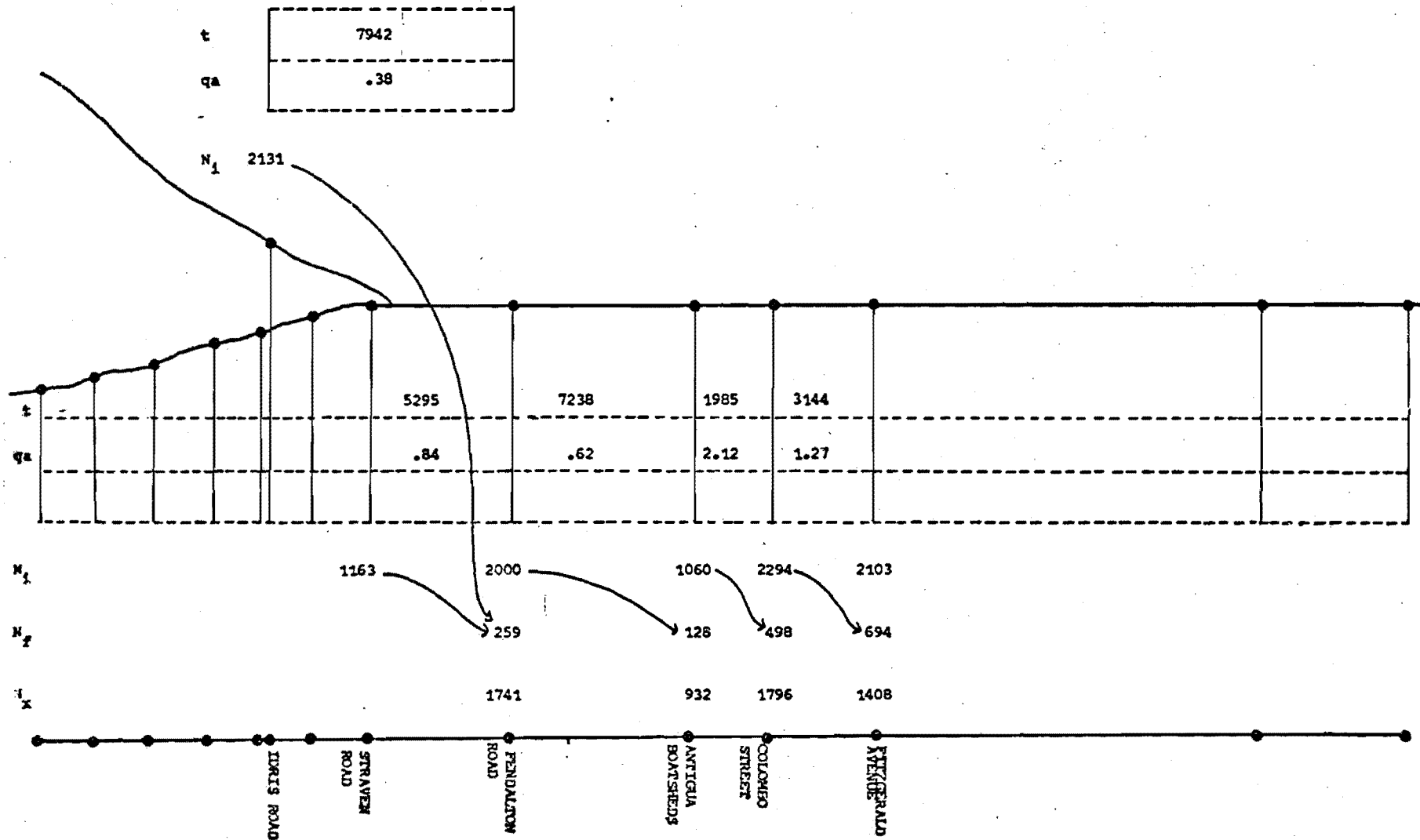


FIGURE 6:

NUMBERS OF FAECAL STREPTOCOCCI ENTERING THE RIVER PER METRE AND NUMBERS ENTERING OVER EACH STRETCH DURING DRY WEATHER SAMPLING PERIOD.

NOTE: Samples in the Avon tributary were not taken on the same occasions as samples in the Avon River.

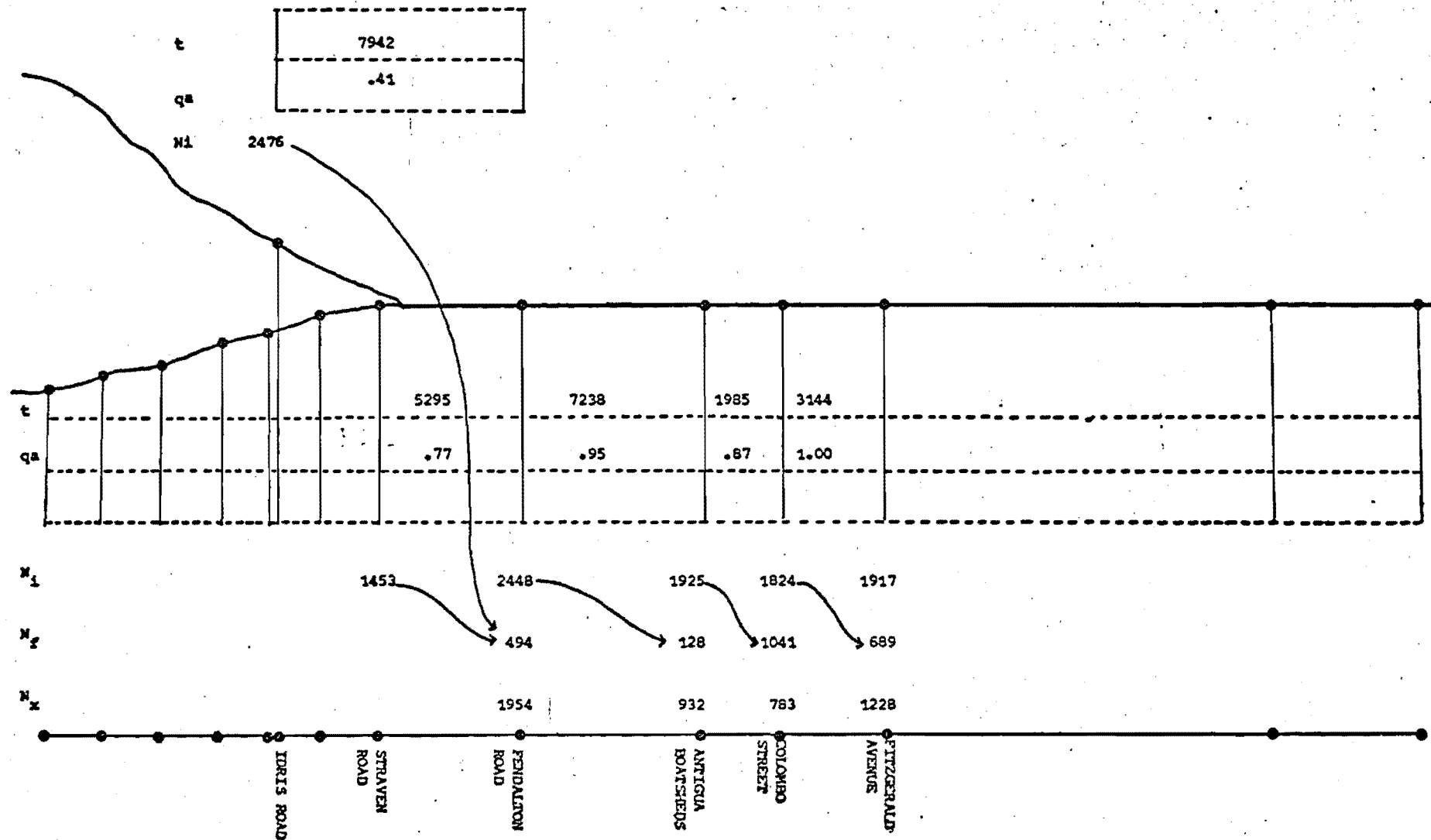


FIGURE 6a:

NUMBERS OF FAECAL STREPTOCOCCI ENTERING THE RIVER PER METRE AND NUMBERS ENTERING OVER EACH
STRETCH FOR WET WEATHER SAMPLING PERIOD.

From their studies on the relationship between the FC/FS ratio and the source of pollution, Geldreich (1967) and Geldreich & Kenner (1969) determined that ratios greater than four indicated a pollution source of mainly human origin, such as domestic waste water, and ratios less than 0.7 suggested that the pollution source was waste from warm-blooded animals other than humans, e.g. livestock and poultry wastes. Lin et al (1974) summarized the relationship of a range of faecal coliform to faecal streptococci (FC/FS) ratio values and their respective probable origins. These are shown in Table 9.

The ratio FC/FS was calculated for the stations on the Avon River for the period of dry weather, and is shown in Table 10. These values are the average values of the ratios of FC/FS calculated over a number of samplings. As numbers of bacteria may vary at different sampling times due to the difficulties in obtaining a representative sample because of the many variables affecting the river, a mean value of a number of samples should give a more representative value.

Table 9. The relationship between FC/FS values and pollution sources (from Lin et al (1974)).

FC/FS = x Range	Indicating Source of Pollution
$x \geq 4$	Human wastes
$4 > x \geq 2$	Predominance of human wastes in mixed pollution
$2 \geq x \geq 1$	Uncertain in interpretation
$1 > x \geq 0.7$	Predominance of animal wastes in mixed pollution
$0.7 \geq x$	Livestock or poultry wastes

Table 10. Ratio of faecal coliform bacteria to faecal streptococci for sites on the Avon River and tributaries. Samples collected during a period of dry weather.

Site No.	Sampling Station	Ratio FC/FS
6	Wairarapa Stream	1.8
7	Waimairi Stream	2.0
8	Avon tributary (Straven Road)	1.5
9	Fendalton Road	1.6
10	Antigua Boatsheds	2.7
11	Colombo Street	5.2
12	Fitzgerald Avenue	3.4
13	Avondale Road	6.4
14	Pages Road	2.3

The data for dry weather suggest that according to the table of Lin et al (1974) (Table 10) as far as the Antigua Boatsheds the pollution is uncertain of interpretation and past this point, mainly of human origin.

However, when the numbers taken from Geldreich (1966) (Table 11) for ducks and humans are added together it can be seen that a ratio of 1.5 (Avon tributary) represents one duck to 15 humans (Table 12) and the ratio of 4.4 more than 1000 humans to one duck. This would suggest that the ducks are minor contributors to the pollution. However, it remains to be seen if the ducks common on the Avon have similar numbers of coliforms and streptococci to those reported in the North American studies.

FIGURE 7:

SITE NUMBER 6: WAIRARAPA STREAM AT IDRIS ROAD BRIDGE:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIOS OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

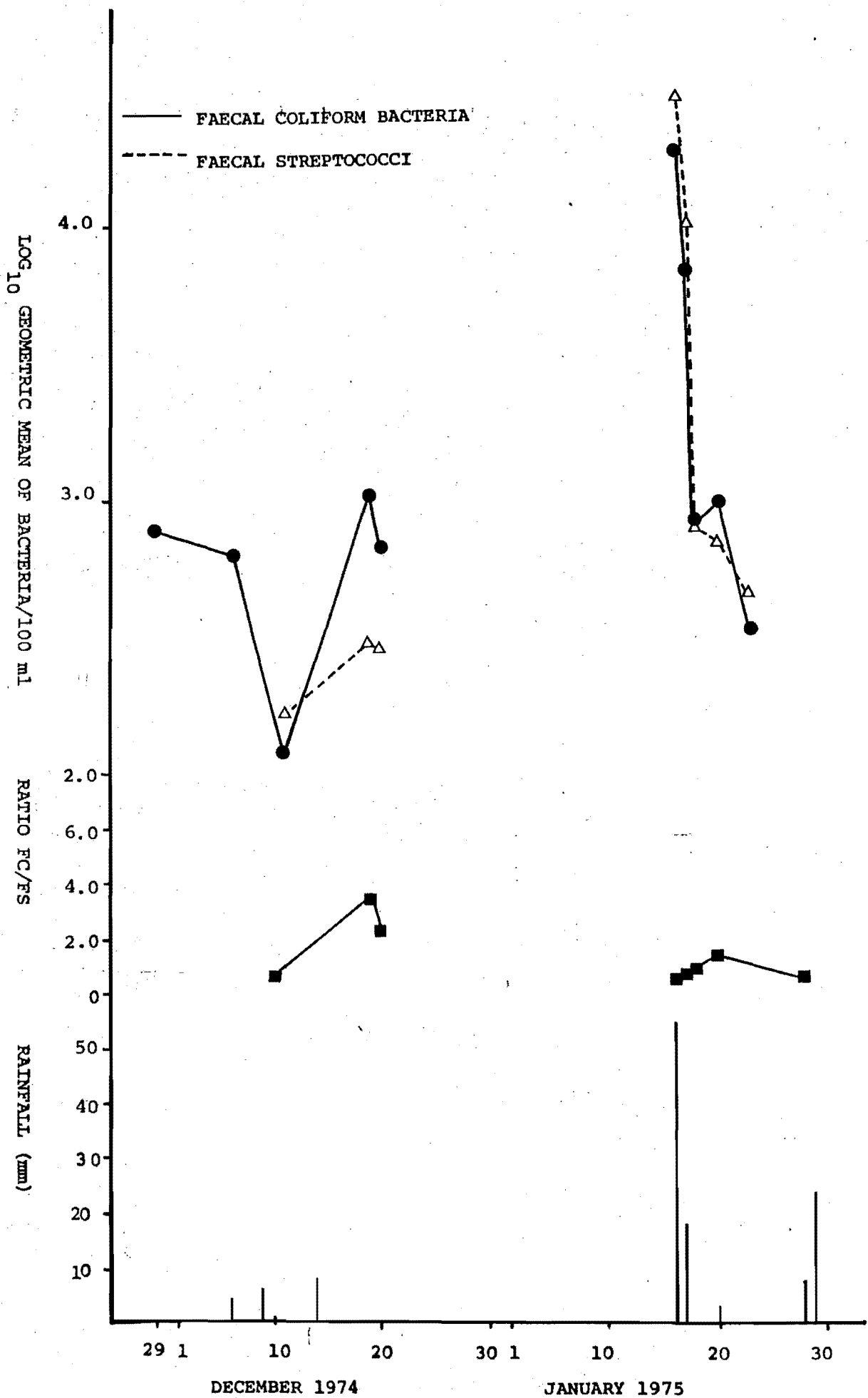


FIGURE 8:

SITE NUMBER 7: WAIMAIRI STREAM AT STRAVEN ROAD BRIDGE:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIOS OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

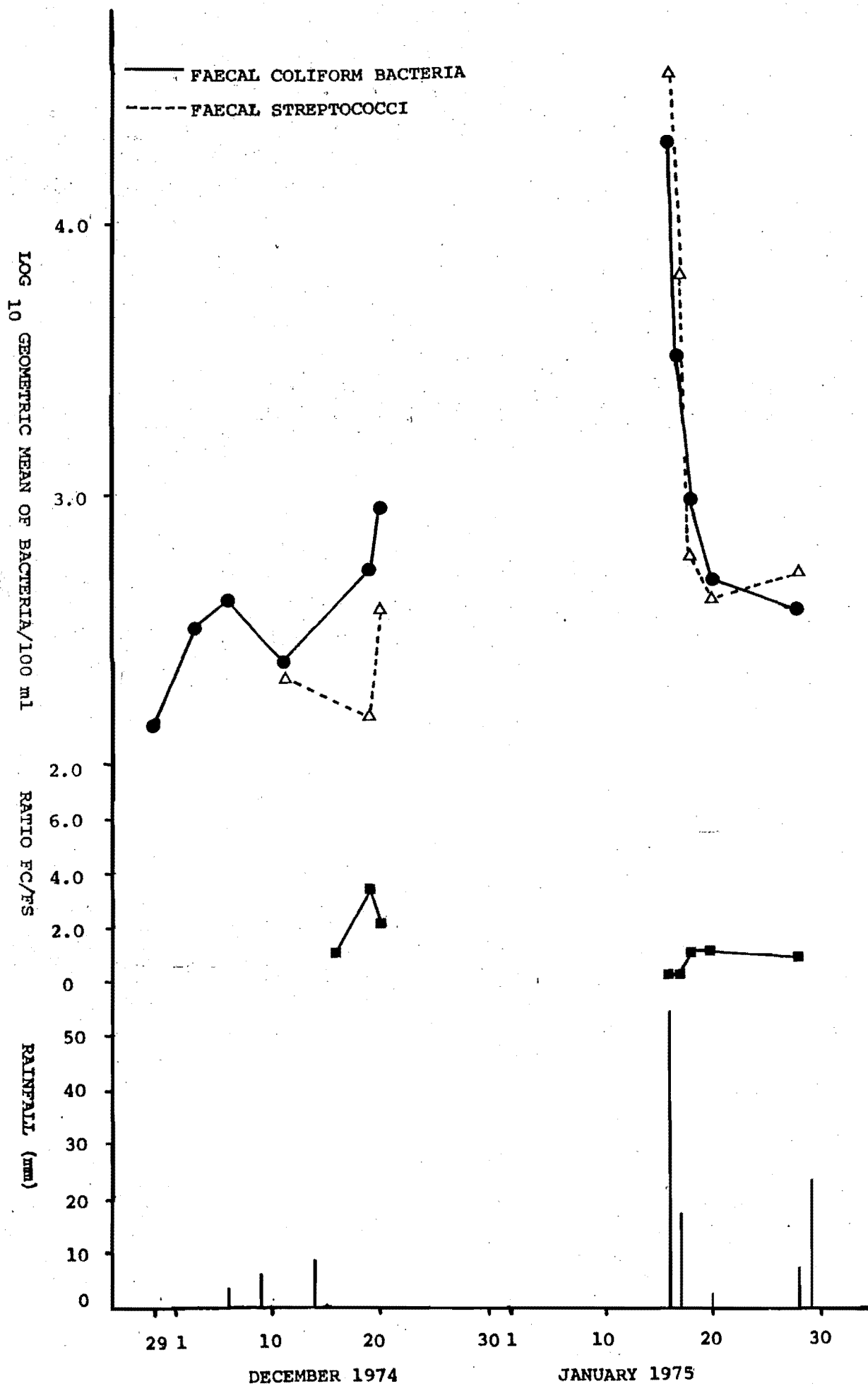


FIGURE 9:

SITE NUMBER 8: AVON TRIBUTARY AT STRAVEN ROAD BRIDGE:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIO OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

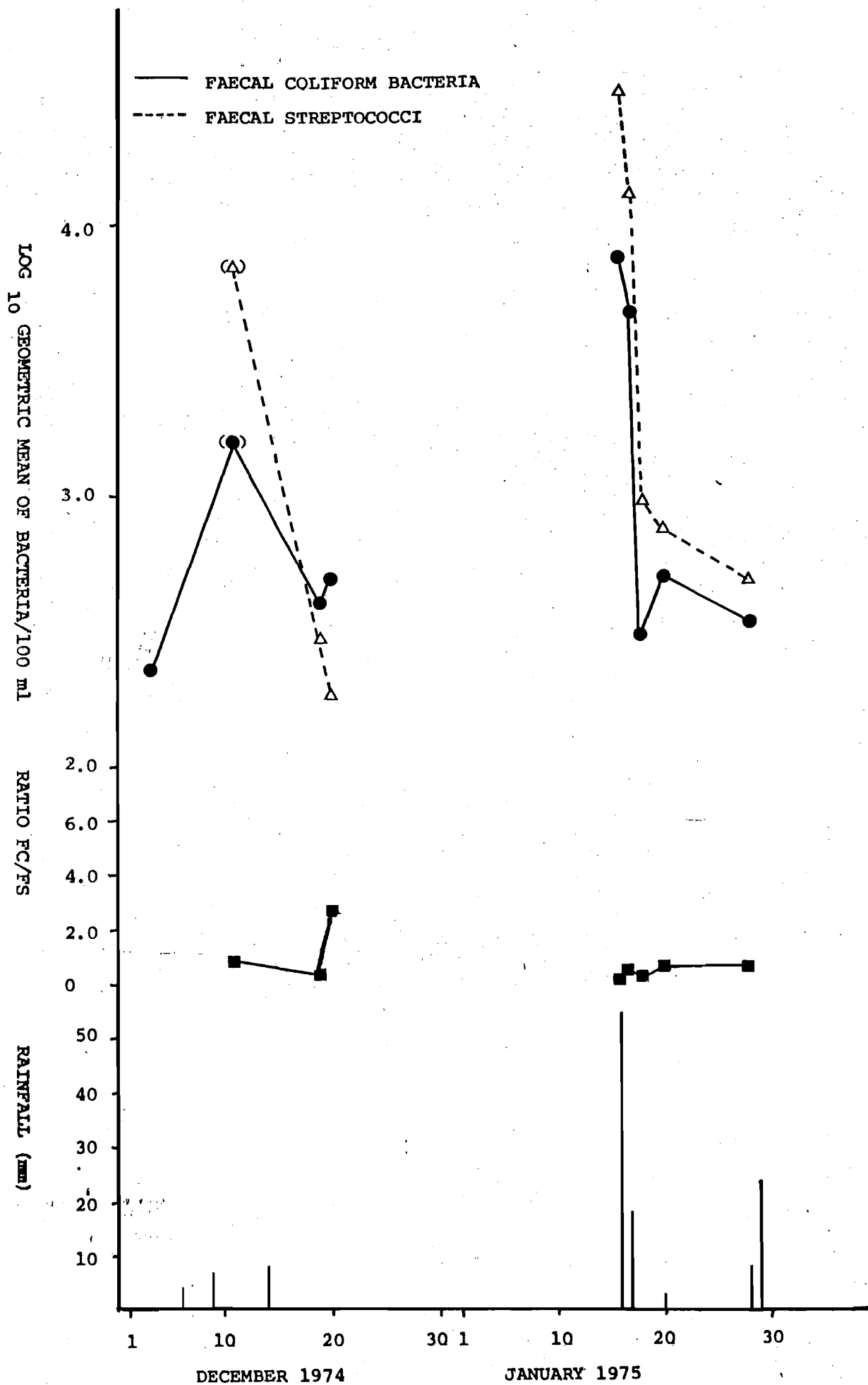


FIGURE 10:

SITE NUMBER 9: AVON RIVER AT FENDALTON ROAD BRIDGE:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIOS OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

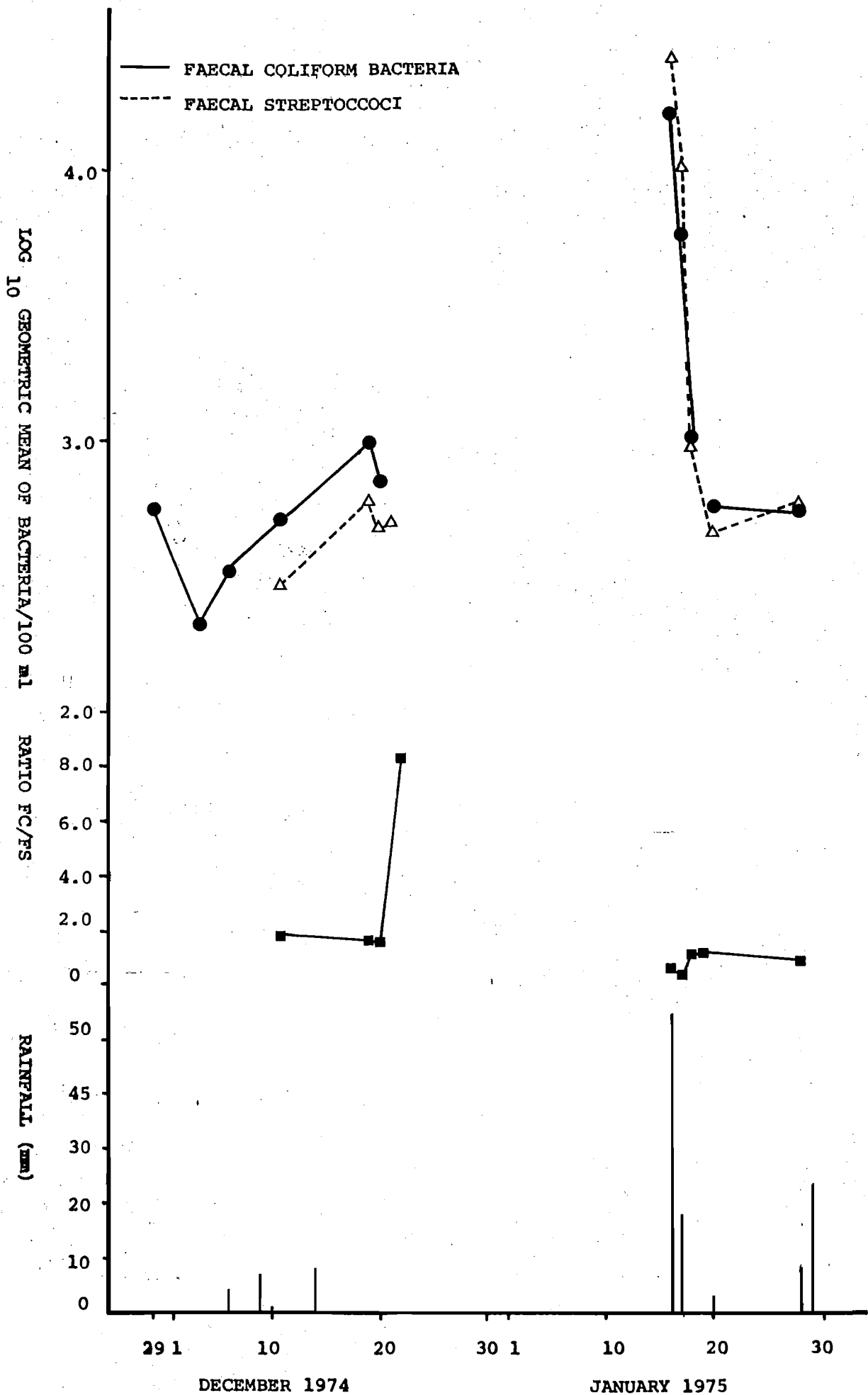


FIGURE 11:

SITE NUMBER 10: AVON RIVER AT ANTIGUA BOATSHEDS:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIOS OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

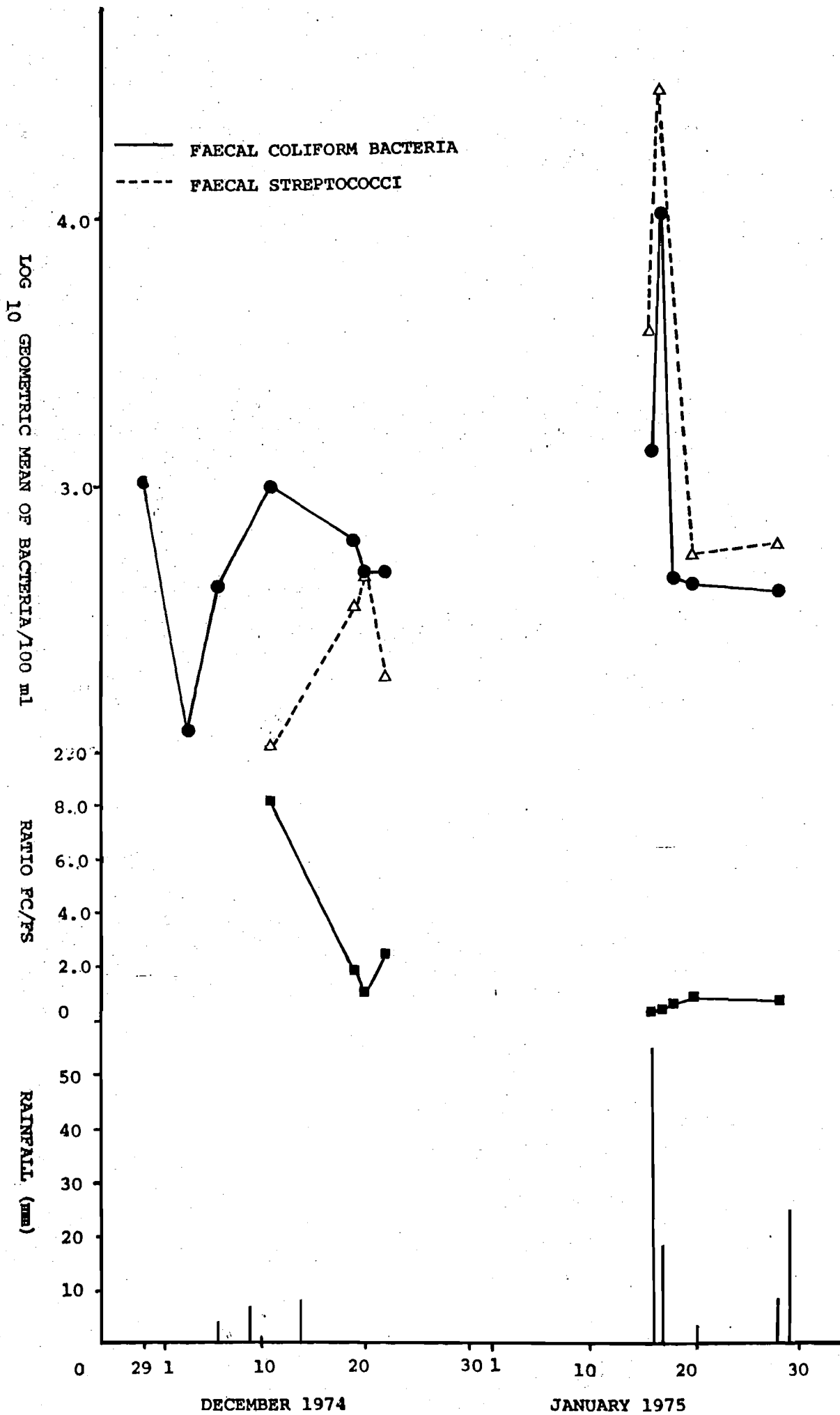


FIGURE 12:

SITE NUMBER 11: AVON RIVER AT COLOMBO STREET BRIDGE:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIOS OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

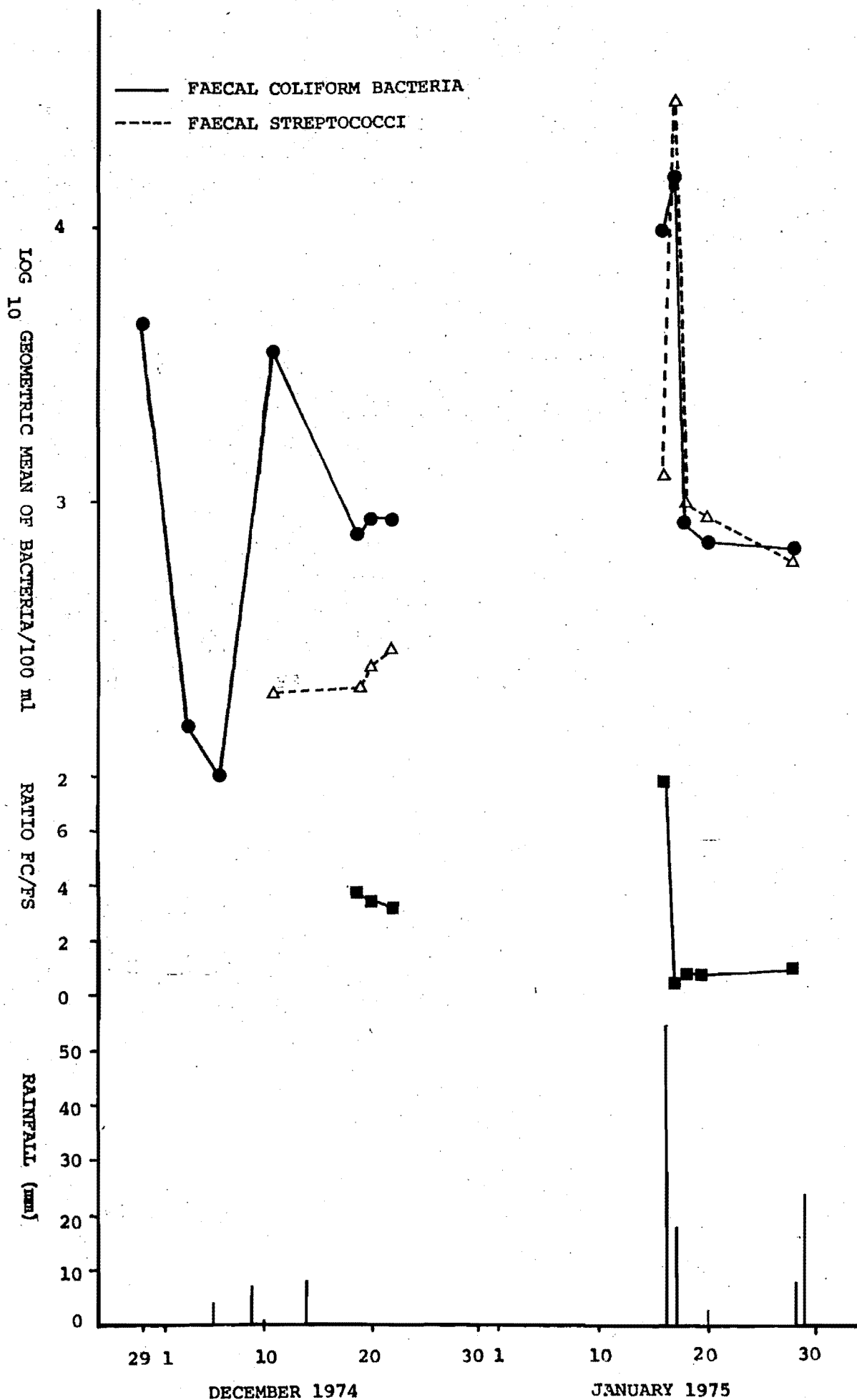


FIGURE 13:

SITE NUMBER 12: AVON RIVER AT FITZGERALD AVENUE BRIDGE:

NUMBERS OF FAECAL COLIFORM BACTERIA AND FAECAL STREPTOCOCCI PER 100 ml
ENUMERATED DURING DECEMBER 1974 AND JANUARY 1975. RATIOS OF FAECAL COLIFORM
BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) AND RAINFALL DATA FOR THE
PERIOD ARE ALSO SHOWN.

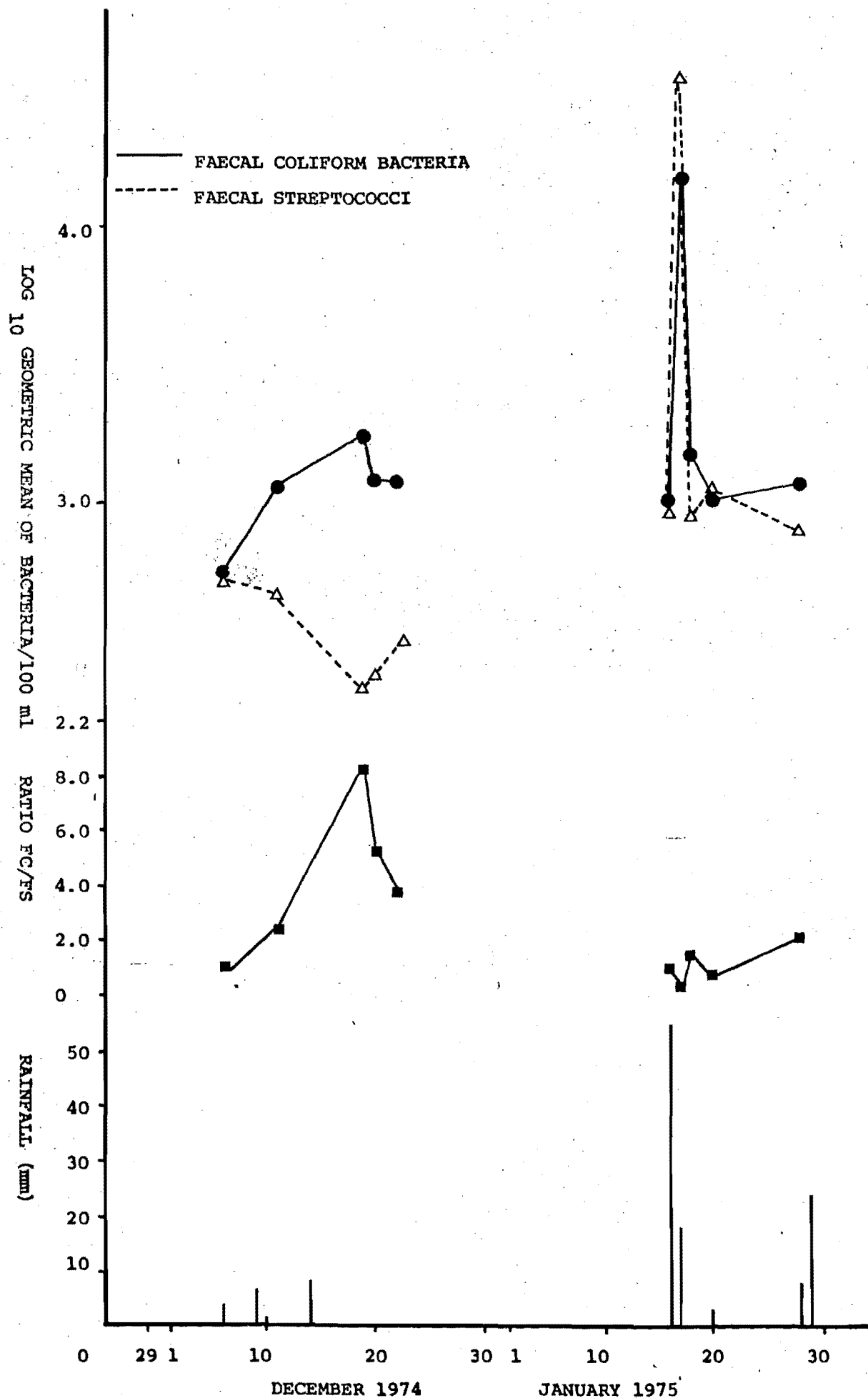


Table 11. Ratios of FC/FS for man and other animals based on contributions per capita per 24 hours of faecal coliform bacteria (FC) and faecal streptococci (FS). (Geldreich, (1966)).

ANIMALS	AVERAGE CONTRIBUTION PER CAPITA PER 24 HR		RATIO FC/FS
	Faecal Coliform (million)	Faecal Streptococci (million)	
Man	2,000	450	4.4
Duck	11,000	18,000	0.6
Sheep	18,000	43,000	0.4

Table 12. Numbers of faecal coliforms and faecal streptococci added to show varying ratios for different contributions.

	Faecal Coliform Bacteria per capita per 24 hours (million)	Faecal Streptococci per capita per 24 hours (million)	RATIO FC/FS
1 man	2,000	450	4.4
1 duck	11,000	18,000	0.6
1 man + 1 duck	13,000	18,450	0.70
2 men + 1 duck	15,000	18,900	0.79
3 men + 1 duck	17,000	19,350	0.88
4 men + 1 duck	19,000	19,800	0.96
5 men + 1 duck	21,000	20,250	1.04
7 men + 1 duck	25,000	21,150	1.18
10 men + 1 duck	31,000	22,500	1.38
20 men + 1 duck	51,000	27,000	1.89
40 men + 1 duck	131,000	45,000	2.91
100 men + 1 duck	211,000	63,000	3.35
200 men + 1 duck	411,000	108,000	3.81
500 men + 1 duck	1,011,000	243,000	4.16
1000 men + 1 duck	2,011,009	468,000	4.30

4.1.4 Numbers of Animals Causing Pollution

Using flow data collected by Christchurch Drainage Board (1973) for the Avon River and for drains and tributaries entering it (Fig. 3) and the average numbers for the dry weather sampling of faecal coliforms and faecal streptococci per 100 ml water (Table 3 and 4) for different points on the river calculations of numbers of bacteria passing that point per day have been made. These are shown in Table 13.

Using Geldreich's numbers for the average contributions of faecal coliforms and faecal streptococci per capita per 24 hours the numbers of people or ducks whose total output could cause this level of contamination were calculated (Table 13).

Duck numbers near Hagley Park have been assessed by Smith (1972) who found that numbers averaged about $2/100 \text{ m}^2$. If numbers over the whole length of the river were $1/100 \text{ m}^2$ then some 3,000 ducks would be present. Using Geldreich's figures for the contribution per 24 hours for ducks (Table 11), 3,000 ducks would contribute 330×10^{11} faecal coliform organisms. Taking the approximate number of faecal coliform organisms passing Antigua Boatsheds during dry weather, which is 10×10^{11} , then each duck would need to deposit only 3% of its faeces in the water to account for the numbers obtained. Further observation would be needed to determine the amount of faeces which is deposited in the river by ducks.

Table 13. Numbers of faecal coliforms (FC) and faecal streptococci (FS) passing different points on the river per day, and numbers of men and ducks whose total output would give this contamination.

Site No.	Sampling Station	FC/ 24 hr (x10 ⁸)	FS/ 24 hr (x10 ⁸)	Approx numbers of men & ducks whose total output would give this contamination.			
				FC		FS	
				Man	Duck	Man	Duck
4	Ilam Road	1,682	698	84	15	155	4
5	Clyde Road	1,295	943	65	12	209	5
6	Wairarapa Stream	1,436	673	72	13	150	4
7	Waimairi Stream	1,031	676	52	9	150	4
8	Avon tributary (Straven Rd)	1,002	676	50	9	150	4
9	Fendalton Road	8,320	7,523	416	76	1672	42
10	Antigua Boatsheds	9,742	4,654	487	89	1030	26
11	Colombo Street	30,812	4,629	154	280	1029	26
12	Fitzgerald Avenue	18,436	5,107	922	168	1268	32
13	Avondale Road	3,287	512	164	30	111	3
14	Pages Road	3,464	787	173	32	175	4

4.1.5 Sediment

It is possible that bacteria adsorbed onto particles in the sediment are contributing to the numbers found in the water. This is unlikely to be of much significance since it was found that numbers in the sediment were low (see Figs 14 & 15).

FIGURE 14:

NUMBERS OF FAECAL COLIFORM ORGANISMS AND FAECAL STREPTOCOCCI PER g OVEN DRY WEIGHT OF SEDIMENT FOR SITES ON THE AVON RIVER.

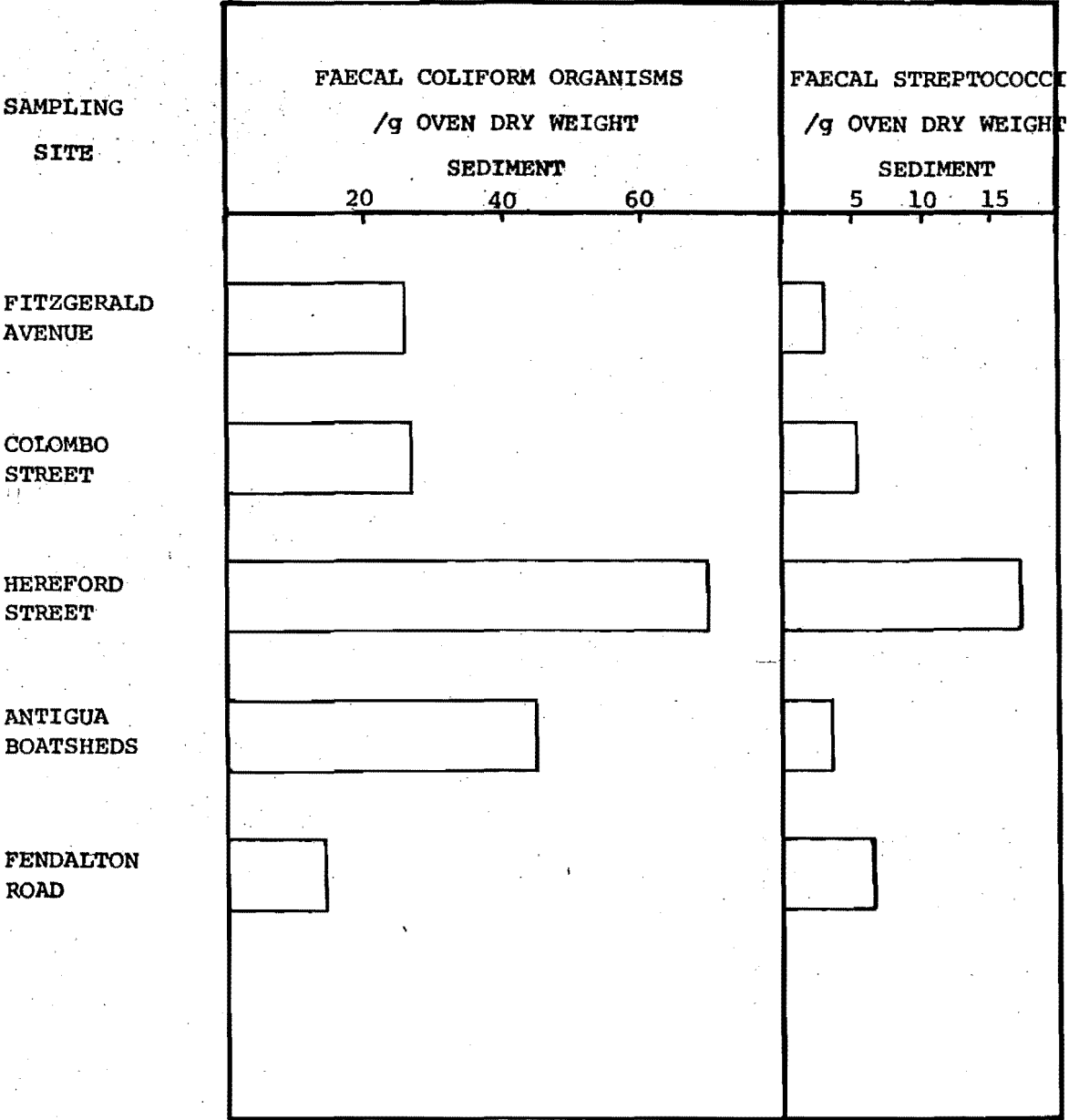
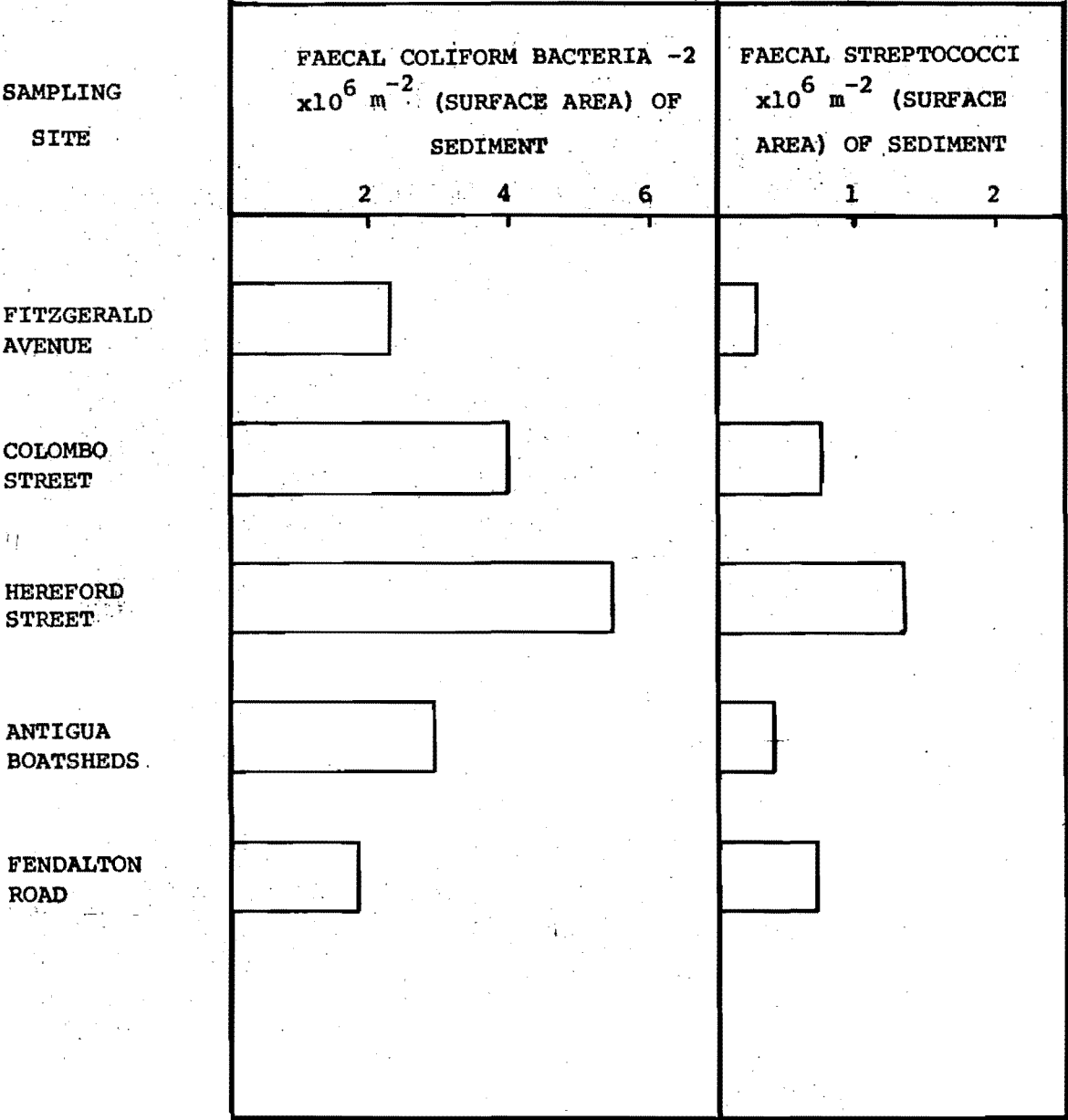


FIGURE 15:

NUMBERS OF FAECAL COLIFORM ORGANISMS AND FAECAL STREPTOCOCCI PER m^2 OF SEDIMENT FOR SITES ON THE AVON RIVER.



4.2 MICROBIAL POLLUTION DURING PERIODS OF RAINFALL

4.2.1 Observed Numbers

During wet weather the number of faecal coliform bacteria and faecal streptococci rose markedly and numbers at many stations exceeded the 2000 faecal coliforms/100 ml specified for Class B waters (Tables 14 & 15 and Appendix Figs 5 & 6). Numbers were high along the entire length of the river with a maximum at Colombo Street bridge.

Table 14. The range of values and the means for numbers of faecal coliforms per 100 ml for the wet weather sampling.

Site No.	Sampling Station	Range	Mean
6	Wairarapa Stream	350 - 14,245	2,131
7	Waimairi Stream	390 - 20,580	1,623
8	Avon tributary	310 - 7,797	1,163
9	Fendalton Road	575 - 16,352	2,000
10	Antigua Boatsheds	410 - 1,390	1,060
11	Colombo Street	680 - 15,500	2,294
12	Fitzgerald Avenue	1,025 - 15,000	2,103

Table 15. The range of values and the means for numbers of faecal streptococci per 100 ml for the wet weather sampling.

Site No.	Sampling Station	Range	Mean
6	Wairarapa Stream	470 - 30,460	2,476
7	Waimairi Stream	414 - 37,460	2,006
8	Avon tributary	500 - 30,700	1,453
9	Fendalton Road	590 - 25,820	2,448
10	Antigua Boatsheds	529 - 29,461	1,925
11	Colombo Street	620 - 29,154	1,824
12	Fitzgerald Avenue	790 - 33,496	1,917

Over the period during and after rainfall the numbers of faecal coliform bacteria and faecal streptococci rose to a peak and then dropped back rapidly to the previous level. These fluctuations in numbers of bacteria with rainfall are shown on graphs of the sampling sites (Figs 7-13). The peak level was similar at all sampling stations, but the time taken to reach this maximum varied with the distance downstream from the source of the river. In the tributaries the peak of contamination was on the day of the rainfall, showing the immediate effect the runoff had on the river where the dilution factor was low. In the Avon River, itself, from Antigua Boatsheds downstream the peak in bacterial levels was not reached until the day following the original high rainfall (see Figs 7-13). This could have been due to the increased dilution factor downstream or to slower runoff. Because of the rate of flow of the water the increase in bacterial levels from the tributaries and drains would not be found in the main river for some time after the runoff occurred.

When these results are compared with those from earlier studies (Table 7) the numbers of faecal coliform bacteria found during and after rain were the highest numbers recorded for almost all sites.

4.2.2 Additional Sources of Microbial Pollution Occuring during Rainfall

Sediment

During periods of rapid river flow resulting from rainfall, bacteria on the sediment both washed from roads and stirred up from the bottom of streams may contribute to the increased numbers of bacteria found during rainfall.

It was found that the numbers of faecal coliform bacteria and faecal streptococci in the sediment of the stream at low flow were low (Figs 14 and 15), compared with the numbers in the water. Using data on the amount of suspended sediment at high flows (Wallingford Report, 1970) and numbers of bacteria/g of sediment, the numbers of faecal coliforms which this sediment would contribute to the water was calculated (Table 16). These results show that the sediment is a minor source of these bacteria in the river, even where the numbers of bacteria in the sediment are highest.

Table 16. No. of faecal coliform bacteria contributed to the water from suspended sediment loads of 100 g m^{-3} and 140 g m^{-3} , occurring at high flow, assuming the entire sediment load is from the bottom. The upper and lower limits of the sediment load at high flow were used in the calculations.

Sampling Station	Faecal Coliform Bacteria/100 ml	
	Lower Limit Sediment Load 100 g m^{-3}	Upper Limit Sediment Load 140 g m^{-3}
Fitzgerald Avenue	0.26	0.36
Colombo Street	0.27	0.38
Hereford Street	0.69	0.97
Antigua Boatsheds	0.45	0.62
Fendalton Road	0.14	0.20

4.2.3 Urban Runoff

Since the disturbed sediment was shown to have a negligible effect in explaining the increased numbers observed during periods of rainfall runoff from streets and properties or overflow from the sewage system must be responsible for a significant part of an increase in numbers.

If it is assumed that flows at the Antigua Street Bridge during the period of the storm beginning on 16.1.75 were

24 hrs of 5 m³/s with 1,390 faecal coliforms/100 ml

6 hrs of 5 " " 12,090 " " "

18 hrs of 5 " " 1,390 " " "

24 hrs of 3 " " 455 " " "

then the total number of faecal coliforms which would flow past this point in 72 h would be 2.5×10^{13} . If the numbers which normally pass during conditions of low flow are subtracted (i.e. 455 faecal coliforms/100 ml and a flow of 2.08 m³/sec.), then the extra numbers amount to 2.2×10^{13} during the three days.

The length of roads in the catchment of the Avon up to the Antigua Street bridge is approximately 184 km, giving a curb length of 368 km. Hence the number of faecal coliforms washed from the roads and adjacent properties can be calculated as 5.9×10^{10} /km curb.

This result is 10-fold higher than the 3.5×10^9 faecal coliforms per km found by Sartor et al (1974).

The difference could be explained by the long interval of dry weather which had occurred prior to the rainfall on the 16th February, which allowed a large number of faecal coliforms to accumulate on the streets. Alternatively, the estimates of river flow used in the present study were insufficiently accurate and further work would be needed to verify the results.

4.2.4 Origin of the Microbial Pollution

For the wet weather period ratios of faecal coliform bacteria to faecal streptococci are considerably lower and most sites therefore show a predominance of animal wastes in the pollution (Table 17). In the Avon tributary and at the Antigua boatsheds the ratios indicate that pollution is from livestock or poultry wastes.

4.3 SALMONELLA DETECTION

When the colonies from the differential agars which produced an acid butt and an alkaline slant on TSI were transferred to Christensen's urea agar all except four cultures produced marked alkalinity indicating that they were not Salmonella cultures. The remaining cultures were reinoculated on differential agars and the colonies produced were not typical of Salmonella species.

Although the experiment did not detect the presence of Salmonella this does not necessarily ensure that all known Salmonella strains were absent. Isolation of Salmonella from streams has only recently been considered practical. Increased interest in the problems of faecal contamination of natural waters has increased the need for suitable methods for routine isolation of Salmonella.

The swab method of sampling is a satisfactory way of exposing a large surface area to the flow of water. It has been suggested (Spino, 1966) that Salmonella organisms from streams are detected more readily using an elevated temperature technique for incubation of the enrichment broths. The method we used was that of Edwards and Ewing (1972), however Spino (1966) has found that in some cases an elevated temperature enhances the recovery of Salmonella organisms from streams. Further work should be carried out to consider this possibility.

These results agree with the findings of Geldreich (1966), who found that in storm water from urban, residential and rural areas the ratio of faecal coliforms to faecal streptococci was less than 0.7. This could be explained if it was assumed that ducks normally deposit most of their faeces on the banks of the river. During rain the land runoff would wash the duck faeces into the river, markedly increasing the proportion of animal pollution in the river. The runoff from faeces deposited by animals and birds on ^{tr}~~streets~~^s and the land around houses would also contribute to the predominance of animal wastes.

Between Colombo Street and Fitzgerald Avenue in the wet weather there was a greater increase in the numbers of faecal coliforms than in the numbers of faecal streptococci. This would suggest that there was an increase in human contamination over this stretch during rainfall.

Table 17. Ratio of faecal coliform bacteria to faecal streptococci for sites on the Avon River and tributaries. Samples collected during a period of wet weather.

Site No.	Sampling Station	Ratio
2	Athol Terrace	0.7
3	Waimairi Road	0.8
4	Ilam Road	0.8
5	Clyde Road	0.8
6	Wairarapa Stream	0.9
7	Waimairi Stream	0.9
8	Avon tributary (Straven Rd)	0.4
9	Fendalton Road	0.8
10	Antigua Boatsheds	0.6
11	Colombo Street	0.8
12	Fitzgerald Avenue	1.1

5. CONCLUSIONS

During a low flow period the numbers of faecal coliform bacteria in the Avon River were above the 200/100 ml specified for Class C water, but below the 2000/100 ml for Class B water. These numbers were, in general, lower than those reported by previous investigators, but the previous results were obtained during periods of varying flow and rainfall. With a period of heavy rain after the dry weather, the numbers of faecal coliforms rose rapidly to over 1000/100 ml. The counts rose to a peak in the upper reaches earlier than further downstream, due possibly to a high dilution factor downstream, coupled with a later build-up of contamination from upstream. The numbers of faecal coliform bacteria dropped back rapidly to their previous levels when the rain ceased.

The numbers of faecal coliform bacteria in the tidal zone were found to be less than 200/100 ml. In this area numbers were lower than anywhere else on the river. These results differ from previous work in which high numbers were recorded in this area. The decrease in numbers could be due to a new practice adopted at the Sewage Works of discharging into the Estuary only on the ebb tide, thus reducing the amount of contaminated water being pushed back up the river.

The initial results led to further sampling sites being investigated in the upper reaches of the river. High levels of contamination were found at these sites. It was surprising to find such a high degree of contamination in an area where the river flows through mainly garden properties. Sewage overflows are unlikely to be a source of bacteria here, and, indeed, this investigation shows that the pollution appears to be mainly of animal origin. Estimates of the origin of the pollution (either animal or human) are based on ratios of faecal coliform bacteria and faecal streptococci resulting from North American studies. It has been assumed for the purposes of the present

study that the ducks found in the Avon River have the same ratio of faecal coliforms bacteria to faecal streptococci as the ducks in the North American study, but to verify these results the ratio of these bacteria will need to be enumerated for ducks in the Avon River.

Ducks are well known to be carriers of pathogenic bacteria. Some species of Salmonella may even be present in the eggs. In this study the organisms which have been isolated are only indicators of pathogens. From the public health aspect it will be necessary to confirm the presence of the pathogens themselves in Avon ducks.

To ascertain the amount of pollution occurring at different parts of the river, results of experiments estimating the length of time the bacteria survived in the water and the change in numbers of bacteria between sampling stations were used in a standard formula from which the numbers of bacteria dying between the stations could be worked out, and thus the number entering the river. This number was highest between Antigua Boatsheds and Colombo Street for dry weather flow and appeared to be a combination of animal and human wastes, thus a combination of ducks, dogs and sewage overflows may have been the cause. Further investigations are required to sort out the actual cause. In wet weather the inputs of animal waste between Fendalton Road and Antigua Boatsheds increased markedly. This can be explained if it is assumed ducks normally deposit their faeces on the land, so when it rains land runoff will increase the contribution from ducks, but again this hypothesis needs testing.

It was found that the sediment contained only a few indicator organisms so the increase in numbers of bacteria during rainfall must have come from a source other than the sediment. Urban runoff or sewage overflows must have contributed to the very marked increase. The numbers of faecal coliform bacteria per km of curb were estimated to be 10 times the numbers found in a study by

Sartor et al. (1974). This may be explained either by the unusually long dry period prior to the rainfall which allowed a large build-up of faecal coliforms, or by the fact that the estimates of river flow used in the study were insufficiently accurate. Further work is needed to verify the results.

The length of time the bacteria survive in the water is a critical factor in the study of bacterial pollution. In this project an artificial system was designed to simulate the natural environment as closely as possible by suspending dialysis tubing containing the bacteria in the river flow; however the bacteria may not have been in exactly the same conditions as those in the river. A more sophisticated experiment which simulated the natural conditions more closely will need to be designed to check the accuracy of the results obtained here. Bacteria which disappear from the water may not necessarily die. Although examination of the sediment in this study did not reveal their presence, other isolation techniques may do so. This whole problem of survival of bacteria is a complex one but it is of immense practical importance.

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8. APPENDIX

FIGURE 1:

GEOMETRIC MEANS OF FAECAL COLIFORM BACTERIA PER 100 ml WATER FOR SITES ON THE AVON RIVER SAMPLED DURING A DRY WEATHER PERIOD. STANDARD DEVIATION OF THE LOG_{10} OBSERVED NUMBERS AND THE RATIO OF FAECAL COLIFORM BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) ARE SHOWN FOR EACH SITE.

<u>Site No.</u>	<u>Sampling Site</u>
6	Wairarapa Stream (at Idris Road)
7	Waimairi Stream (at Straven Road)
8	Avon Tributary (at Straven Road)
9	Fendalton Road Bridge
10	Antigua Boatsheds
11	Colombo Street
12	Fitzgerald Avenue
13	Avondale Road
14	Pages Road

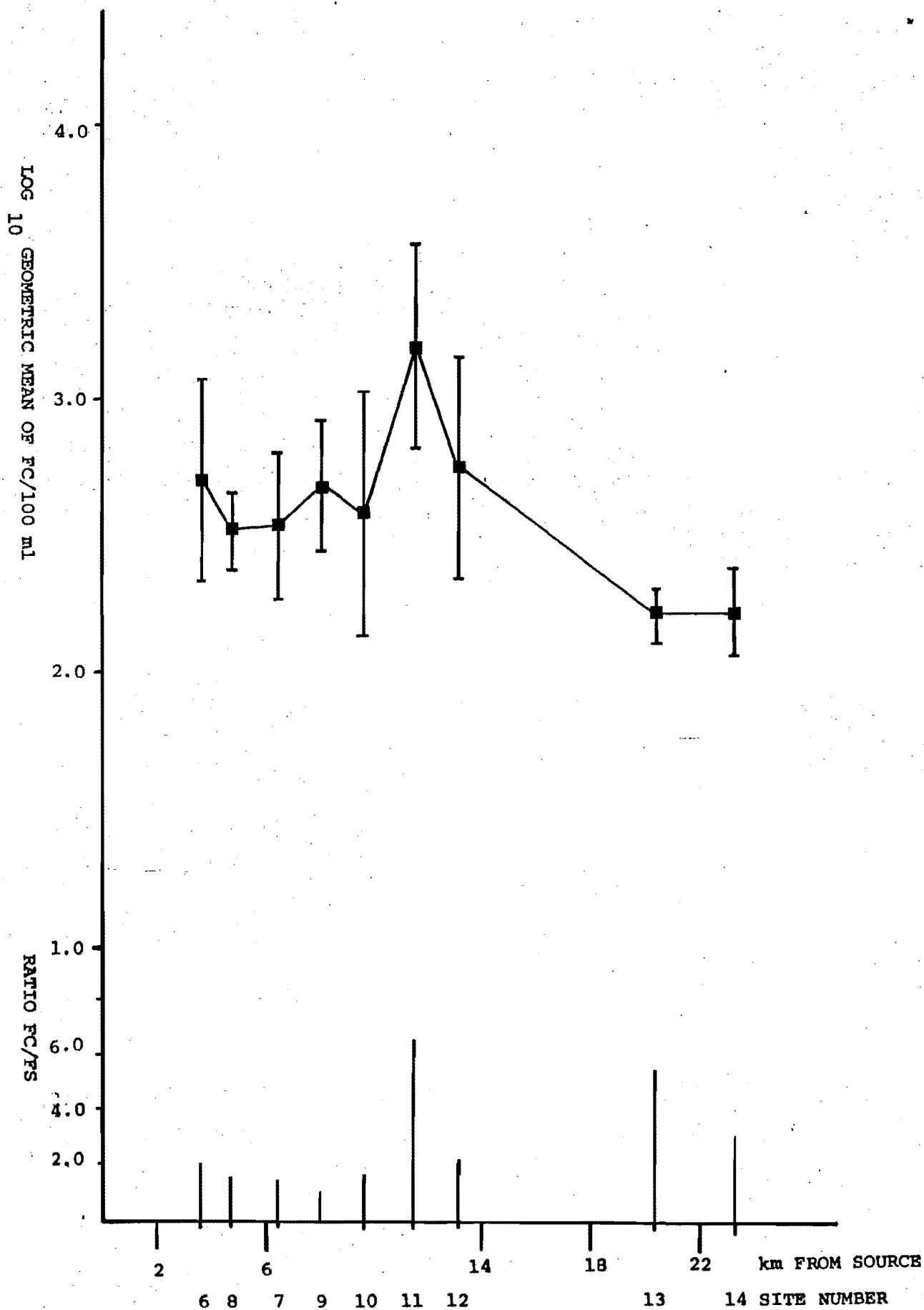


FIGURE 2:

GEOMETRIC MEANS OF FAECAL STREPTOCOCCI PER 100 ml WATER FOR SITES ON THE AVON RIVER SAMPLED DURING A DRY WEATHER PERIOD. THE RATIO OF FAECAL COLIFORM BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) ARE SHOWN AND STANDARD DEVIATION OF THE \log_{10} OBSERVED NUMBERS ARE SHOWN FOR EACH SITE.

<u>Site No.</u>	<u>Sampling Site</u>
6	Wairarapa Stream (at Idris Road)
7	Waimairi Stream (at Straven Road)
8	Avon Tributary (at Straven Road)
9	Fendalton Road Bridge
10	Antigua Boatsheds
11	Colombo Street
12	Fitzgerald Avenue
13	Avondale Road
14	Pages Road

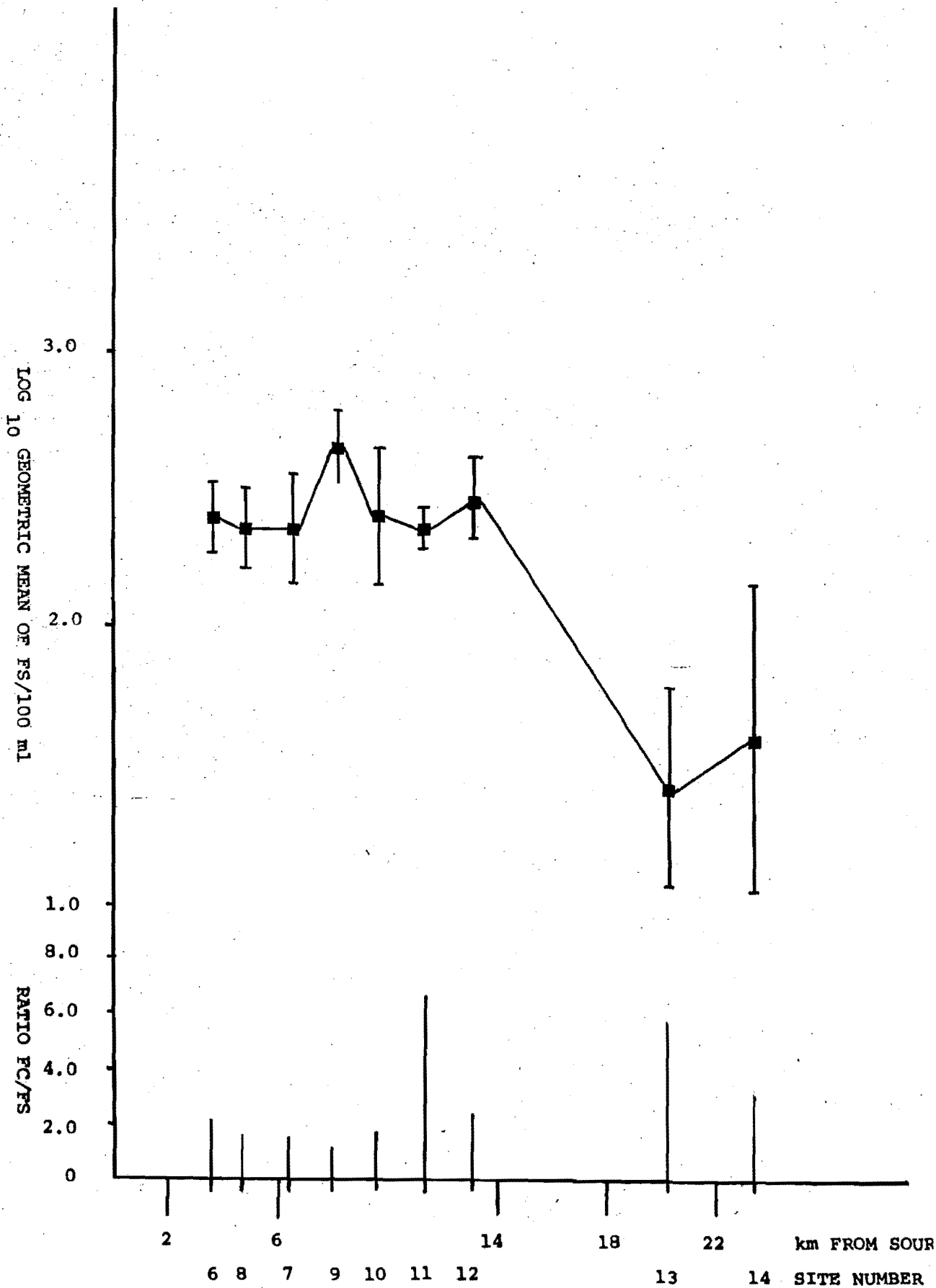


FIGURE 3:

GEOMETRIC MEANS OF FAECAL COLIFORM BACTERIA PER 100 ml WATER
FOR SITES ON THE AVON TRIBUTARY. THE STANDARD DEVIATION OR
LOG₁₀ OBSERVED NUMBERS AND THE RATIO OF FAECAL COLIFORM BACTERIA
(FC) TO FAECAL STREPTOCOCCI (FS) ARE SHOWN FOR EACH SITE.

<u>Site No.</u>	<u>Sampling Site</u>
1	Avonhead Road Bridge
2	Athol Terrace Bridge
3	Waimairi Road Bridge
4	Ilam Road Bridge
5	Clyde Road Bridge
8	Straven Road Bridge

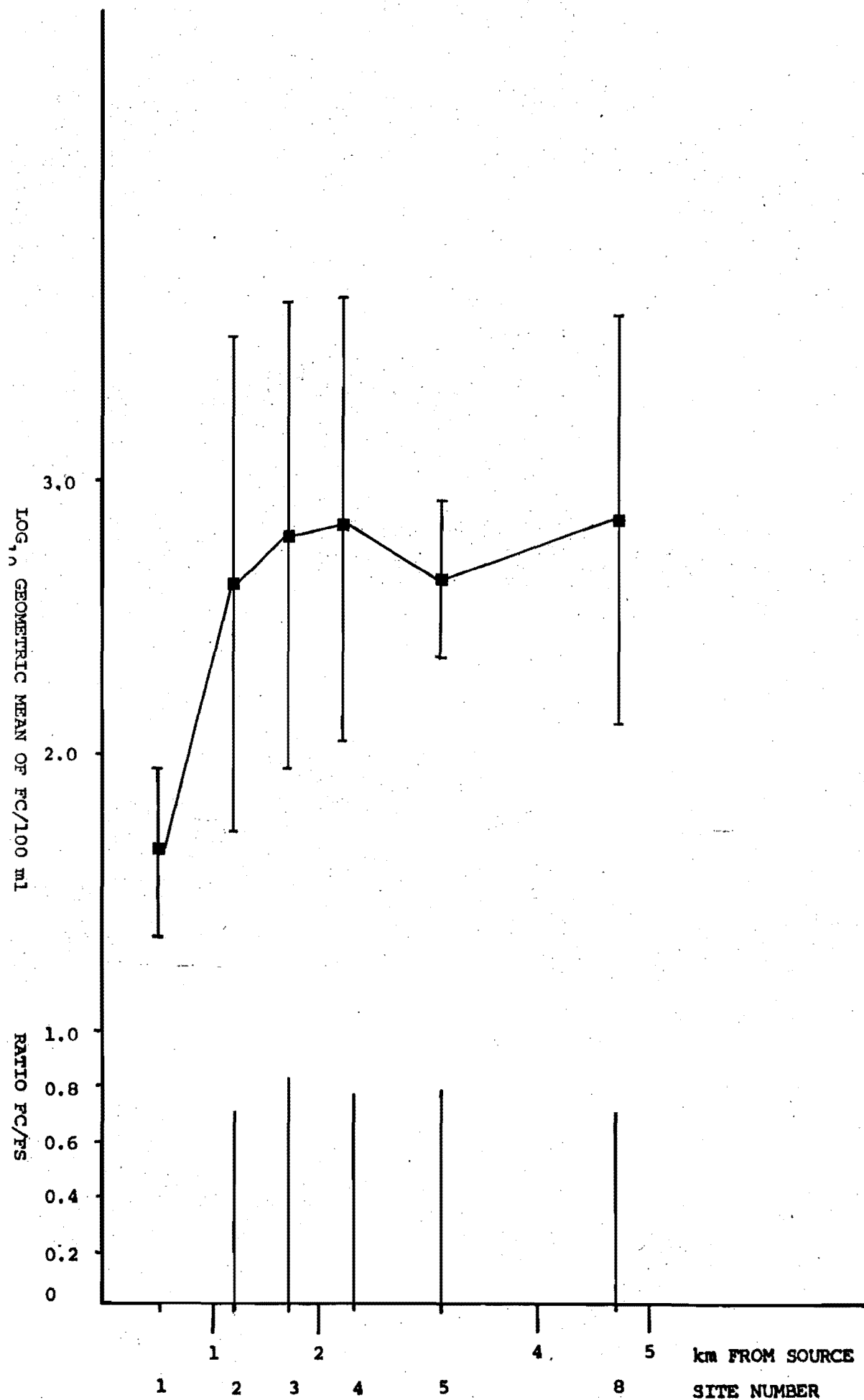


FIGURE 4:

GEOMETRIC MEANS OF NUMBERS OF FAECAL STREPTOCOCCI PER 100 ml WATER FOR SITES ON THE AVON TRIBUTARY. STANDARD DEVIATION OF LOG_{10} OBSERVED NUMBERS AND THE RATIO OF FAECAL COLIFORM BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) ARE SHOWN FOR EACH SITE.

<u>Site No.</u>	<u>Sampling Site</u>
1	Avonhead Road Bridge
2	Athol Terrace Bridge
3	Waimairi Road Bridge
4	Ilam Road Bridge
5	Clyde Road Bridge
8	Straven Road Bridge

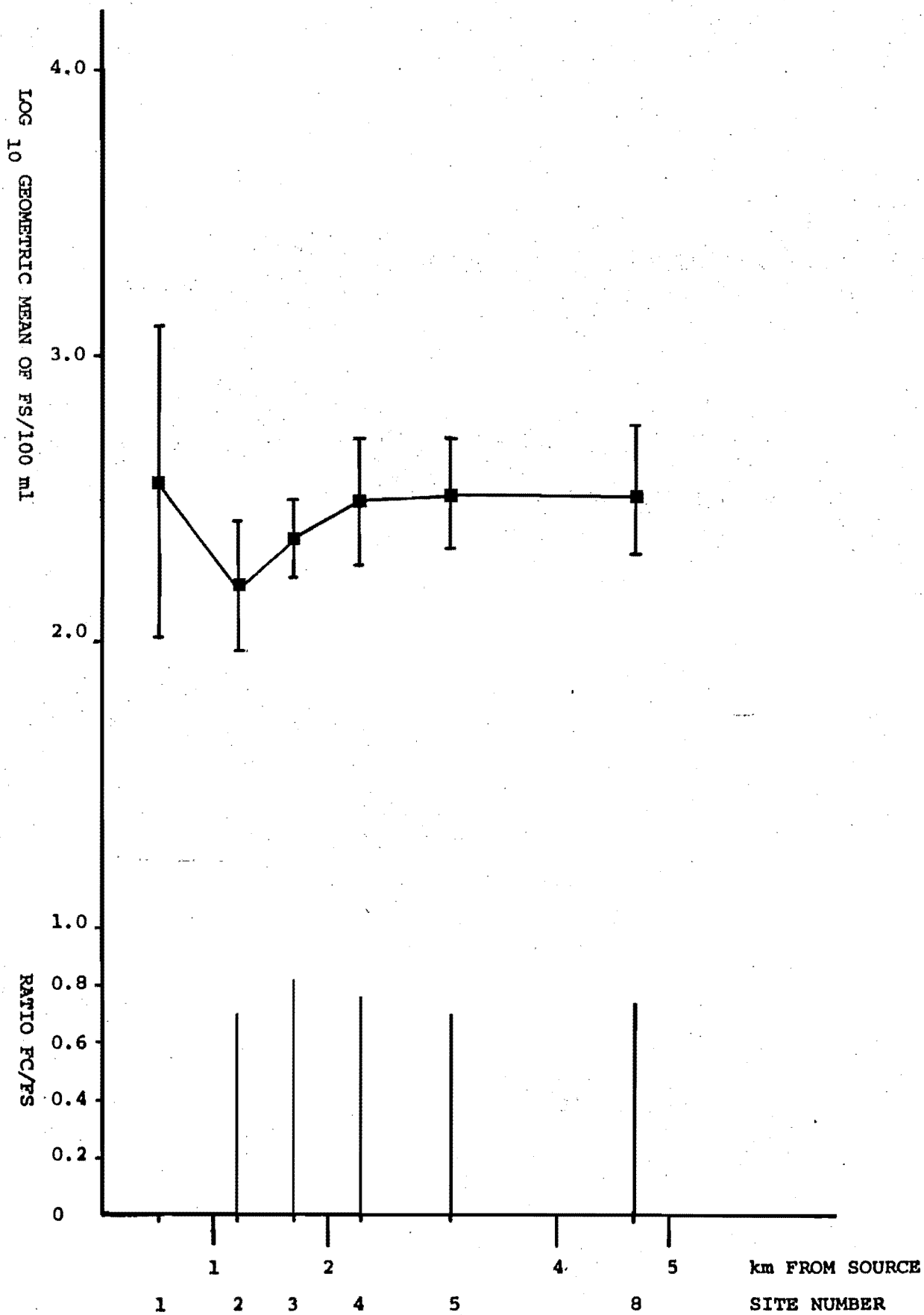


FIGURE 5:

GEOMETRIC MEAN OF FAECAL COLIFORM BACTERIA PER 100 ml FOR SITES ON THE AVON RIVER SAMPLED DURING WET WEATHER. STANDARD DEVIATION OF THE LOG₁₀ OBSERVED NUMBERS ARE SHOWN FOR EACH SITE AND RATIO OF FAECAL COLIFORM BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS).

<u>Site No.</u>	<u>Sampling Site</u>
6	Wairarapa Stream
7	Waimairi Stream (Straven Road)
8	Avon Tributary (Straven Road)
9	Fendalton Road Bridge
10	Antigua Boatsheds
11	Colombo Street Bridge
12	Fitzgerald Avenue Bridge

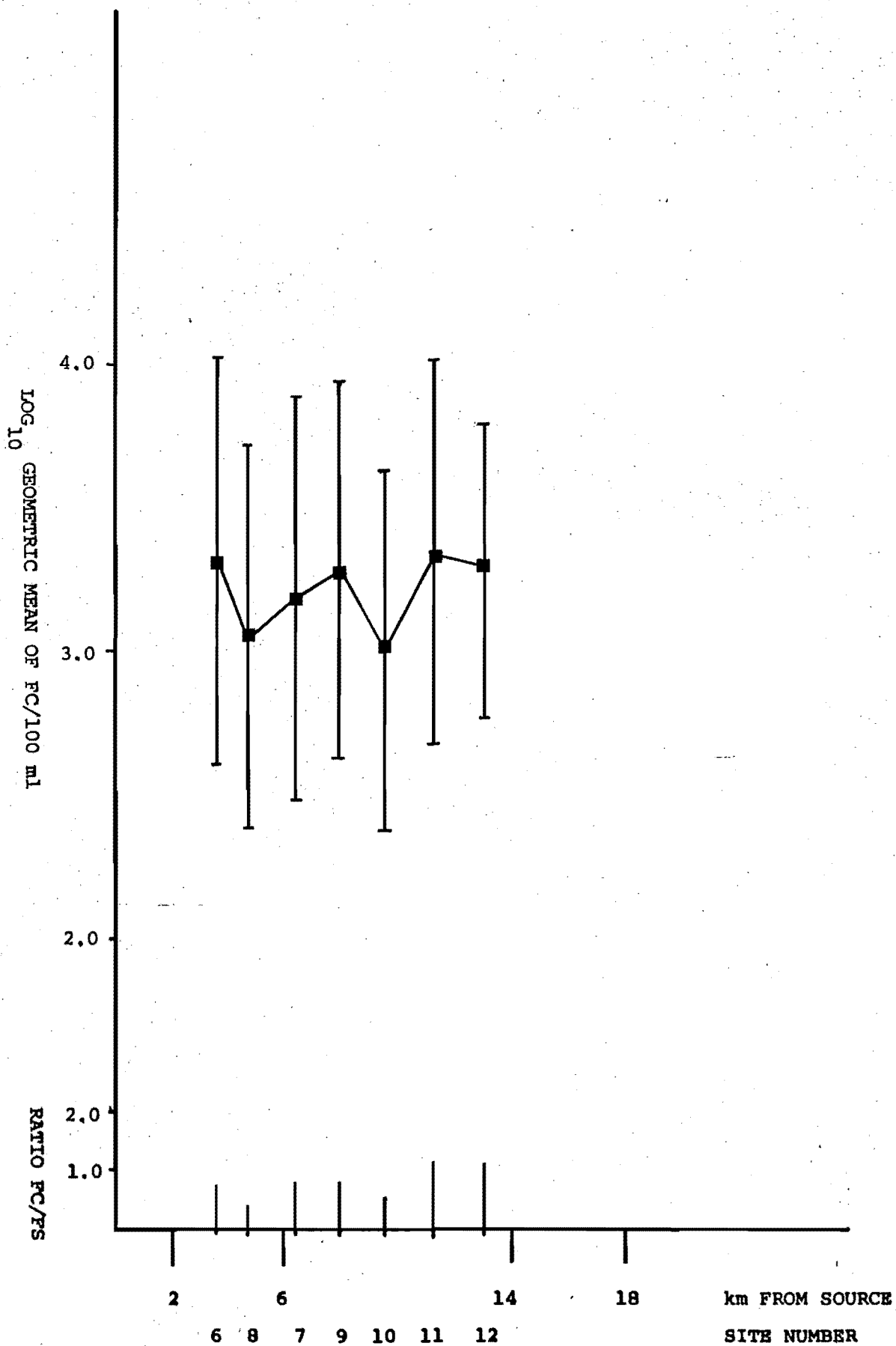
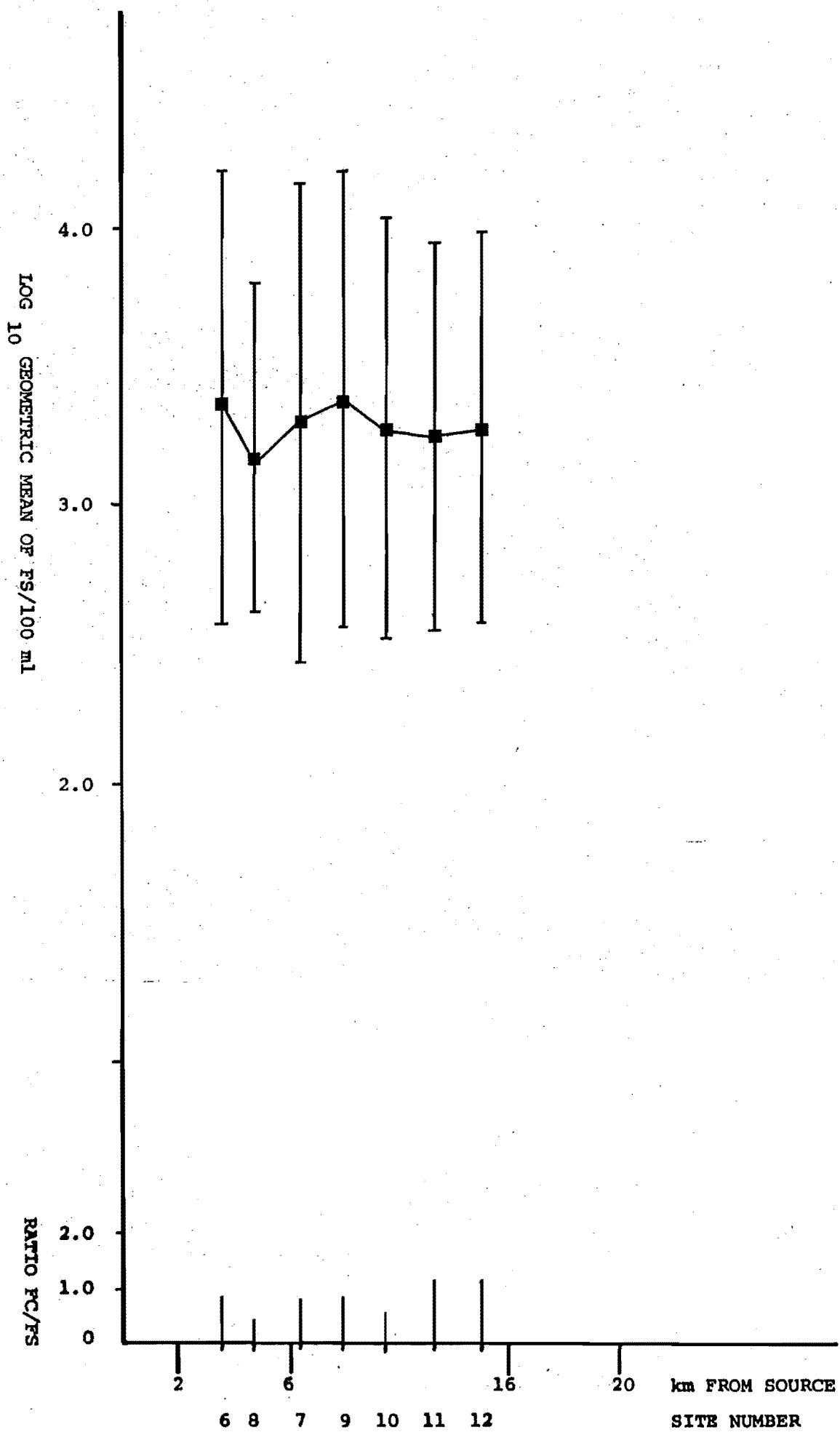


FIGURE 6:

GEOMETRIC MEANS OF FAECAL STREPTOCOCCI PER 100 ml WATER FOR SITES ON THE AVON RIVER SAMPLED DURING A WET WEATHER PERIOD. STANDARD DEVIATION OF \log_{10} OBSERVED NUMBERS AND THE RATIO OF FAECAL COLIFORM BACTERIA (FC) TO FAECAL STREPTOCOCCI (FS) ARE SHOWN FOR EACH SITE.

<u>Site No.</u>	<u>Sampling Site</u>
6	Wairarapa Stream (at Idris Road)
7	Waimairi Stream (at Straven Road)
8	Avon Tributary (at Straven Road)
9	Fendalton Road Bridge
10	Antigua Boatsheds
11	Colombo Street
12	Fitzgerald Avenue



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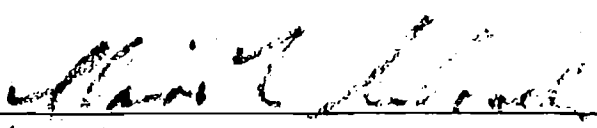
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