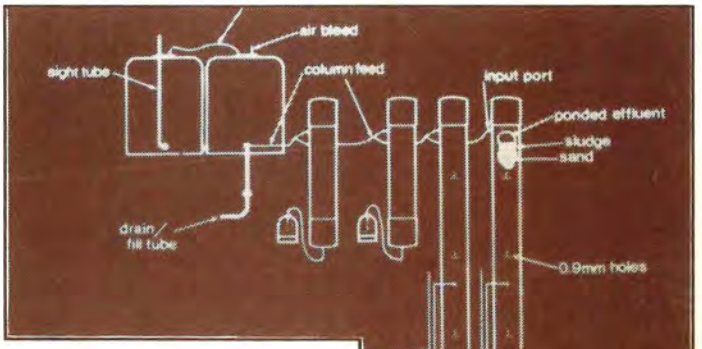
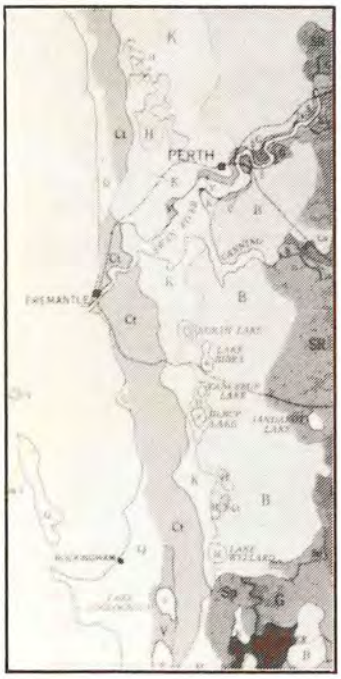
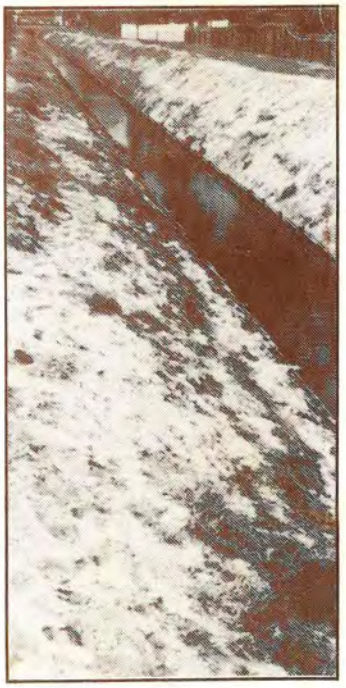
MICROBIAL ASPECTS OF SEPTIC TANK EFFLUENT DISPOSAL







Department of
Conservation and Environment
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May 1983

MICROBIAL ASPECTS
OF SEPTIC TANK EFFLUENT DISPOSAL
INTO COARSE SANDS
IN THE PERTH METROPOLITAN AREA

by

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FOREWORD

This bulletin has been prepared following the writing of a doctoral thesis in 1983 by the author who was the recipient of a Departmental Environmental Studies Fellowship from 1977 to 1981.

The Department of Conservation and Environment Fellowships aim to promote scientific research and to develop knowledge of man's effects upon the environment in areas not normally studied by Government Departments.

It is hoped that the publication of Dr Parker's research in this form will add to the community's knowledge and understanding of the microbial aspects of septic tank effluent disposal.

It should be understood that views stated or implied by the author are not necessarily those held by the Department of Conservation and Environment or the Government.

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ACKNOWLEDGEMENTS

This study was funded by the Department of Conservation and Environment, Western Australia. The work was carried out in the Department of Microbiology, University of Western Australia, in co-operation with a group studying other aspects of septic tank operation in the Division of Land Resources Management, Commonwealth Scientific and Industrial Research Organization (CSIRO)*. Numerous people and organisations have contributed to the study, both practically and scientifically.

I would like to thank Professor Neville Stanley, Associate Professor John Mackenzie, Dr Brian Mee, all of the Department of Microbiology, University of W.A., Dr Warren Grubb, Western Australian Institute of Technology (formerly of the Department of Microbiology, University of W.A.), Mr Barry Carbon, Alcoa of Australia (formerly of the Division of Land Resources Management, CSIRO), Mr Brian Whelan, Dr Jim Barrow and Mr Greg Bartle, CSIRO.

Thanks are also due to the helpful officers of various State government organisations and to numerous householders who "volunteered" their gardens and without whose co-operation the study would have been incomplete.

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* See Carbon and Murray (1980), Whelan et al. (1981), and Whelan and Titmanis (1982).

OBJECTIVES

The objectives of this study were to determine whether the use of septic tanks is a potential source of microbial pollution of groundwater in the sand soils of the Swan Coastal Plain, Western Australia, and, to provide an understanding of the interaction of sewage micro-organisms with three selected sands.

The information presented in this report, it is hoped, will contribute to the management guidelines for septic tank installation and operation in the Perth Metropolitan Area, and in other communities situated on coarse sand soils on the Swan Coastal Plain.

SUMMARY

A fifteen-site field survey studying two coarse sands showed that microbes were removed from the sand with a small clay/silt fraction (Spearwood sand) within 0.65 metres of travel. In contrast, no maximum distance for microbe travel was determined for Bassendean sand which has no clay fraction. In this sand, microbes travelled further, but the reduction of counts of indicator bacteria to zero was not observed at the depths studied.

A large column study, designed to simulate in the laboratory the function of the septic tank and soil absorption system, gave essentially similar results. By comparison, relatively larger numbers of both E. coli and Salmonella spp. were present in the effluent. Despite this greater loading, there was very little breakthrough of the bacteria in Spearwood sand, compared with substantial breakthrough for Bassendean sand. The same type of experiment was done with a "model" virus. The fact that the removal was good for both soils for this organism should not be taken to mean that such a result would be universal for enteroviruses.

Considering the survival of the sewage bacteria in soils, it was shown that the differences between Bassendean and Spearwood sand, evident in terms of movement, did not affect survival. However, significant proportions of inoculated bacteria remained viable over 64 days.

Another important mechanism in soil purification, adsorption, was examined. Both a Salmonella sp. and a model virus were used in adsorption studies. It was found that,under controlled conditions and those likely to obtain in the soil beneath a septic tank, Bassendean sand did not adsorb the bacterium, but may have adsorbed the virus if exposure time was extended. Spearwood sand was a much better adsorbent.

The field and laboratory data indicate in general that Bassendean sand is unsuitable for septic tank effluent purification, whereas Spearwood sand can be regarded as effective for microbe removal.

Note:-

This report is an abbreviated version of a doctoral thesis (Parker 1983). The reader is urged to consult this thesis for a wider discussion of the subject.

RECOMMENDATIONS

It is evident from the data obtained in this study that the use of empirical methods for septic tank management should be revised. Whilst no specific health hazard has been predicted for septic tank operation in sandy areas of Perth, it is apparent that the potential is there under defined conditions. Coarse sand soils have limitations and should not be regarded as universally acceptable merely on hydraulic criteria.

The metropolitan area of Perth is built largely on Spearwood and Bassendean sands. The concentration of septic tank systems is in the peripheral areas of the city, mainly areas located on Bassendean sand. The demonstrated inadequacy of this sand for efficient effluent purification is compounded by the presence of high seasonal water tables in large areas. It seems prudent therefore, to concentrate the available resources of the sewerage programme in these areas.

There is always likely to be human contact with untreated groundwater (either surface manifestations or pumped bore water). All means should be used to minimise such contact. Specifically, this might be done by restricting access to drainage canals and compensating basins. The installation of bores in gardens presents some problems, particularly with respect to the position and depth of the bore. Current regulations require that a bore/well be

located at least 30 metres from a septic tank system. Although this may be possible to achieve within one garden, it is not necessarily possible with respect to neighbouring gardens. It is reasonable therefore, to suggest that some form of regulation of bore/well installations be made by qualified health surveyors. The objective here would be to ensure that proper construction is made and that spears penetrate the water table sufficiently far. (Limited evidence suggests that microbial pollutants do not penetrate the water table but travel along the surface.) At the same time, information regarding appropriate management could be provided to residents to ensure that any surface pollutants are not pumped. The use of groundwater should be restricted to garden irrigation, but where, for example, it is used to fill swimming pools, careful attention should be paid to chlorination. Untreated groundwater should not be drunk.

It is not possible on the basis of this study to construct maps showing where septic tanks could be used (both new and existing) in Bassendean sand areas, with respect to public health hazard. In Spearwood sand areas, a depth of not less than 2.0 m of permanently unsaturated soil is adequate, but where porous rock is found, the use of septic tanks should be avoided since saturated flow of effluent is possible. It would be feasible to monitor the effect of septic tanks on local groundwater by the use of a tracer dye. There may be some merit in selecting a number of households in Bassendean sand areas with high water table to establish the movement of effluent. A follow-up survey

might involve the use of a bacterium, particularly a sewage organism such as Escherichia coli. Mutant strains of this organism have been used with success and enable the environment to be monitored selectively.

Another approach might be to undertake a more general microbiological monitoring programme of an area with septic tanks and compare this with a sewered area.

In conventional water pollution bacteriology work, it has been possible to establish criteria regarding acceptable coliform counts for different waters*. Those counts are related to the likelihood of finding pathogenic organisms in such water samples. There is evidence that suggests that the relationship between indicator bacteria (coliforms) and pathogens found for waters is not valid for sediments and soils. For this reason no recommendations regarding "acceptable" coliform counts in surface manifestations of groundwater can be given.

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*Environmental Protection Authority (1981). Water quality criteria for marine and estuarine waters of Western Australia. Department of Conservation and Environment, Bulletin no. 103. Perth, Western Australia. pp 10, 19, 37, 41.

1.0 INTRODUCTION

1.1 Sewage disposal in the Swan River Colony and the inheritance of the septic tank problem in Perth.

In 1844, after fifteen years of settlement on the Swan River, there were only some 4,000 settlers (Seddon 1972). Fig. 1 shows the layout of the Perth townsite in 1838. Sewage was disposed of into cesspits adjacent to the house: the coarse sandy soils must have seemed ideal in this respect and their excellent porosity would be a contrast for those who came from a land where soils were often heavy clays which offered poor prospects for pit drainage. Indeed, there was no apparent need to do other than what was done in England.

A sewage disposal problem is largely one of scale. A small population might reasonably handle its sewage disposal by crude cesspit methods and not suffer serious water-borne infections. As the population of Perth grew, housing areas encroached on swampy land which became lakes in winter. The sinking of household wells to provide drinking water in proximity to cesspits inevitably led to widespread water-borne disease. The swampy areas of Perth townsite were already regarded as "unhealthy". Dr John Ferguson, the Colonial Surgeon, in a letter in 1847 to the Colonial Secretary gave "unhesitatingly" his opinion that the existence of swamps in the backstreets of the Perth townsite was prejudicial to health, but does not refer to specific diseases (CSO 1847). A year later (CSO 1848) a committee

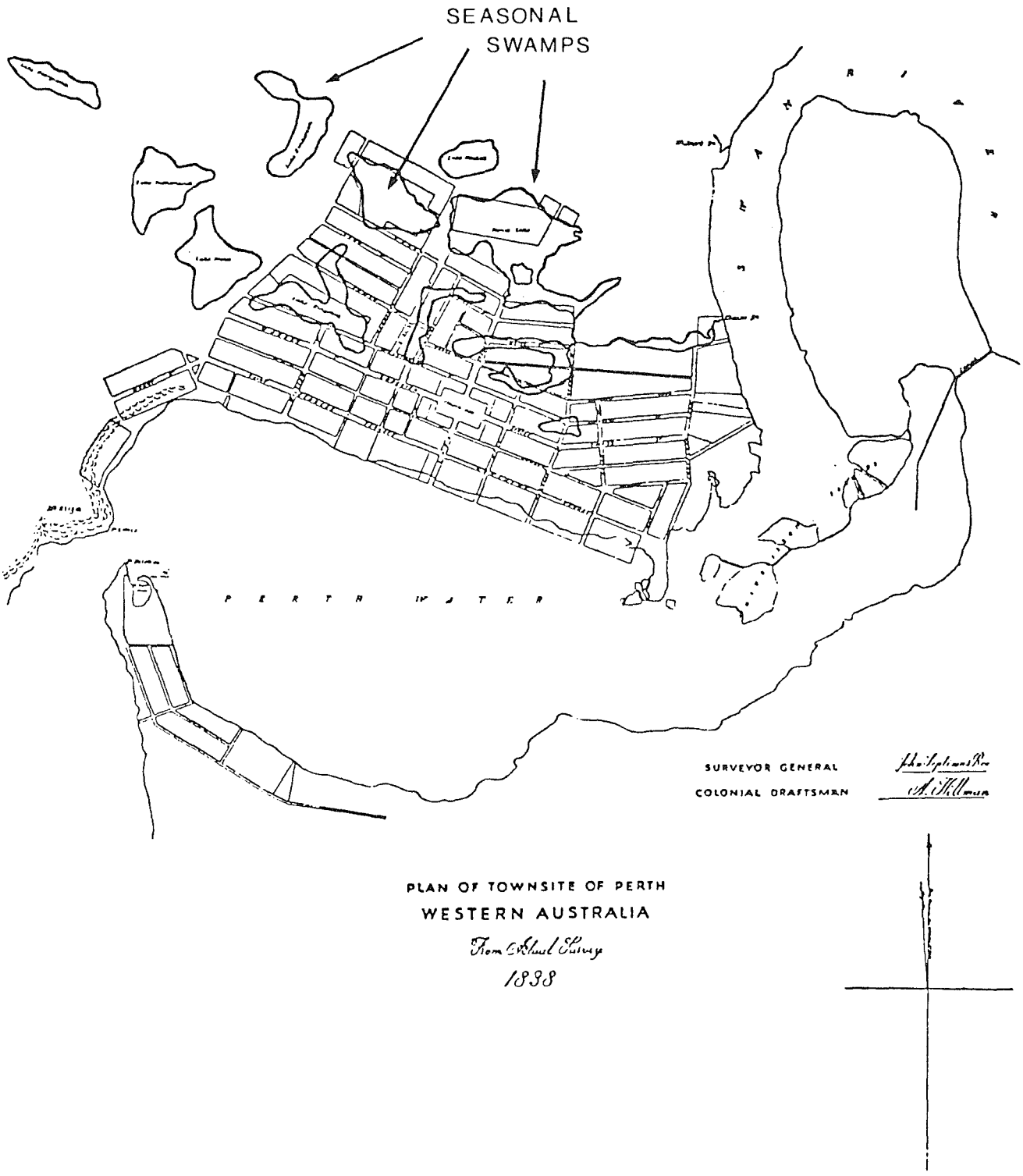

Figure 1 Map of Perth Townsite in 1838 showing seasonal swamp areas.

was set up to investigate and report on the draining of lakes and swamps adjacent to or in the Perth townsite. It was the unanimous opinion of the committee that health was being prejudiced and that growing population density would worsen the situation. Eventually all the lakes in the area were drained. We can only assume that swampy areas were prejudicial to health because of the widespread use of cesspits and their seasonal flooding. In retrospect, it can be concluded that cesspits are an inefficient way of disposing of sewage, particularly in swampy areas, but the alternatives were not available in 1847. The problem was seemingly intractable. It was a significant event in 1868 when the first "Inspector of Nuisances" was appointed by the Perth City Council (Stannage 1979). "Nuisances" was presumably the euphemism for faulty or leaking cesspits and other odour-causing problems. Stannage quotes the first appointee, Sergeant Dale, who observed that "so long as bad drainage exists, we are liable to periodical attacks ... of sickness". Dale at least had the sound belief that cesspits should be uniform in size and cleaned monthly (Perth City Council Minute Book, 8 Nov. 1869, quoted by Stannage (1979). By this time the seriousness of the problem went further than objectionable smells. The Acting Colonial Surgeon, Dr Shaw, in his report for 1874, stated that:

> "There is plenty of water to be obtained in Perth by sinking wells; it is more or less pure, but sometimes of an opalescent or muddy colour, nauseous taste and putrescent smell; this is no doubt to be inscribed in

> a great measure to the absence of any kind of sanitary precaution in preventing contamination of the water by soakage from cesspools into the wells; often, indeed, this occurrence is favoured by the construction of cesspits close to and on higher ground than the well. The houses are nearly all detached, and standing in about an acre of ground ... A few yards behind the house is a closet, with an open unbricked cesspit, and again a few yards from this the well, usually about twelve feet deep, from which water is drawn for drinking ... The cesspool is sometimes emptied, the soil being either carted away and used as manure or buried in the stable dung heap. From the above short notice it will be evident that, owing to the neglect of sanitary precautions in the towns at least, future years will bring more of sickness and death than the past one has done" (WAPP 1879).

In the report of the following year, Dr Alfred Waylen, the Colonial Surgeon,comments on the occurrence of "Colonial Fever". *

> "Most of the cases were sent to hospital from the low-lying portion of the city. This is of considerable extent and inhabited chiefly by artizans and labourers, who live in cottages built with but

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* Probable salmonella infections identified by Jameson (1889) as a "variant" of typhoid (Hunt and Bolton 1978).

> little regard to sanitation. There is often a total absence of ventilation as well as of drainage, and it is to these defects that when towards the middle of winter the ground becomes saturated with water, the presence of fever may be ascribed." (WAPP 1879).

By 1880, the real seriousness of the sewage problem was apparent: epidemics of cholera in London in the recent past must have alarmed the population of Perth, particularly those less well-off in the low-lying areas. Very little was done, and Dr Waylen's report as Colonial Surgeon for 1881 stated that "nothing definite has been arrived at as to do with sewage ... with an increasing population, delay will render more difficult initiation of a system of sewage disposal ..." (WAPP 1882). Hunt and Bolton (1978) have discussed the water supply and sanitation problems of the era and comment that Waylen had a great deal of insight into environmental sanitation, seemingly rare in those times.

A commission was appointed in 1884 with Waylen as Chairman to report into the sanitary condition of Perth and Fremantle. The bulk of the commission's report was concerned with sewage disposal problems, and significantly condemned the construction of sewers for Perth and Fremantle on the grounds that there was a lack of daily tidal flow in the Swan River to "carry off by scour what might be conveyed

thither by sewers" (WAPP 1885). There was some wisdom in this condemnation. The lack of adequate tidal flow on the Swan River meant that crude sewage would not disperse. Many years later, during the period 1912-1936, when partially treated sewage was discharged into the river at Burswood Island, there were eutrophication problems, although the sewage was never shown conclusively to be the cause (Swan River Reference Committee 1955). But for sewage disposal in 1885, the commission recommended a "night cart" system. It is also significant that they recommended piping water from the streams of the Darling Range, twenty five kilometres east of the city.

Again, very little action was taken on what were sound recommendations. By 1894, the problem was worse, especially in financial terms. Perth had a public health crusader in the personage of William Traylen, sometime Perth City Councillor and member of the Legislative Assembly. He was a persistent advocate of good sanitation, particularly pure water. Traylen's activities in Parliament led eventually to a night cart system being introduced (WAPD 1893). In 1894 Traylen suggested "that the healthfulness of Perth cannot be preserved without a sewerage scheme and water supply under the same control", but the cost was to be some £200,000 (WAPD 1894).

Inevitably, the problems of serious water-borne infectious diseases became prominent. In 1897 there were 1408 cases of typhoid with 134 deaths (Perth City Council Annual Reports,

1895 - 1900; cited by Hunt and Bolton (1978). In 1898, some of Perth's leading medical practitioners petitioned Parliament urging the need for deep sewerage (WAPD 1898).

The construction of Perth's sewerage was finally begun in 1906. The cost was estimated to be £500,000. The treatment plant was constructed at Claisebrook and discharged into the Swan River (Hunt and Bolton 1978). However, cesspits were still extensively used as was the night cart system. Descriptions of leaking and overflowing cesspits at the turn of the century are reminiscent of those of earlier decades in European cities. A resident of the city complained to the newspapers (PHD 1898a) and insisted that her health and that of her family "was in constant jeopardy, (her) husband was forced 2 or 3 times daily to burn off the foul gas arising from the ... well, which when lighted burns and roars furiously". The premises were inspected and, in the opinion of the inspector, the situation was not as extreme as the complainant had insisted, but nevertheless he thought that demolition would solve the problem when "the well or a portion of its disgusting surroundings may become enclosed within the body of a new building" (PHD 1898b). Below-ground cesspits were finally abolished by legislation in 1911 (Health Act W.A. 1911-1978).

Developments were underway in the use of septic tanks for individual houses. In the U.S.A., Philbrick (1883)

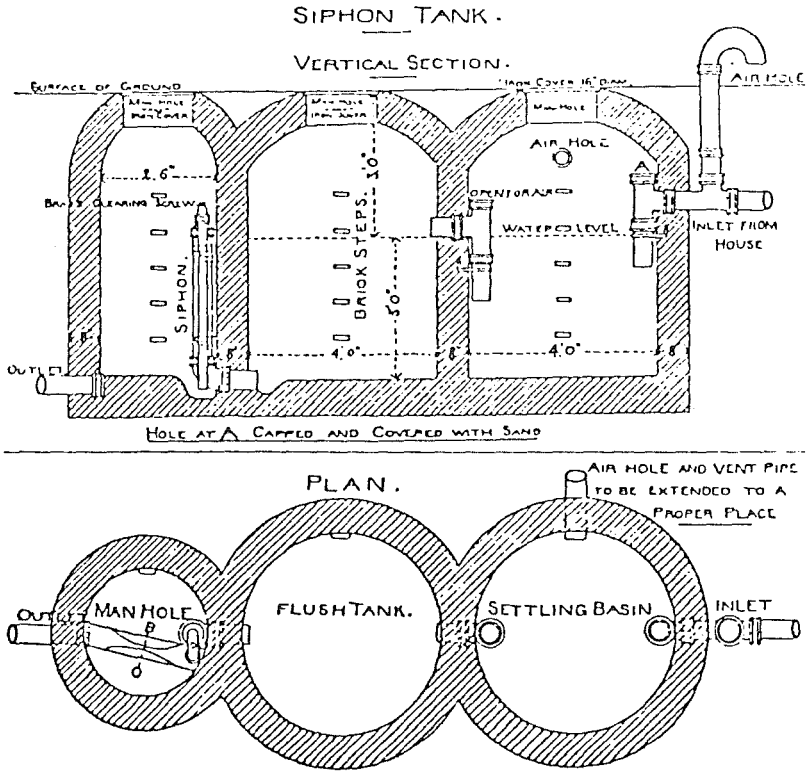


Figure 2 A nineteenth century septic tank design. (Philbrick 1883).

described a septic tank for use in suburban residences of New York (the design is shown in Fig. 2). He suggested that the system needed "a quarter of an acre of grassland for effluent disposal and that efficient effluent disposal would only be obtained by intermittent dosing of the effluent to the soil".

Greenhill (1927) described the early years of septic tank experimentation and stated that, although very few installations were made between 1902 and 1910, they were not of well-designed construction. As long as the water drained away no one cared about the quality of effluent or where it was disposed of. Often the effluent was permitted to drain in municipal storm water drains and thence into the Swan River. Perhaps the lack of legislation regarding septic tanks was understandable since the design and operation was done very much on a trial and error basis. The feelings at the time amongst officials in public health and works are expressed in a letter from the President of the Central Board of Health (CBH) to the Under Secretary of Public Works (PHD 1901), where the writer expressed the view that "experience in England has shown that the establishment (of a septic tank) is just as much a question for bacteriologists as for engineers", and suggested some form of interdepartmental co-operation. In later correspondence, again to the Under Secretary, it was noted that specific examples of such interdisciplinary exchange of ideas have been effective and a household septic tank (probably the first to be constructed properly in Western Australia) was installed in the Perth suburb of Cottesloe by a local civil

engineer. This functioned effectively for eighteen months. Some failures were recorded, and the CBH President concluded by stressing the need to "bring the experimentations to a successful issue and to raise them above the rank of empirical undertakings". He suggested that bacteriological and analytical tests of the operation of septic tanks would be necessary. For reasons of convenience, it was more common for hotels and theatres to have septic tanks installed. (Patrons would not then be required to use privies in adjacent back gardens at some distance from the main building). The operation of these tanks was again largely experimental and in 1904 (PHD 1904), for reasons that are now obscure, a bacteriological examination of the effluents of three hotels and the town hall in Fremantle was requested. In trying to gauge the efficiency of one septic tank, the analyst, Dr Blackburne (PHD 1904) complained that he had not been given a sample of raw sewage in order to arrive at a figure showing percentage reduction in bacteria. Curiously, he was unable to detect any "ordinary sewage bacteria" in the sample tested.

In 1923 a state licensing board compelled hoteliers to provide better sanitary conditions for the travelling public, and this led to legislation providing for properly designed and installed septic tanks to be administered by the Public Health Department. Undoubtedly, the disposal of sewage in Perth proceeded in a haphazard manner for some years. Stannage (1979) quotes the Perth City Council's Medical Officer, Dr Seed, in 1912 as saying that "if an efficient

scheme of disposing of the sewage is completed, we may expect typhoid to disappear from Perth at an early date". Later, in 1915, nine years after sewerage construction was begun, the decline in typhoid cases was "plainly due to the deep drainage system". But as Stannage continues, only one fifth of the city had sewers by 1914.

By association at least, we can see that some infectious disease was related to the use of crude cesspits, particularly in low-lying areas of the city. Apparently there was no active experimentation on either the survival or movement of typhoid organisms in water-saturated local sands to substantiate the views of people like Seed. In an era when it was thought the odour of sewage or putrefaction was enough to cause typhoid, it would perhaps have seemed an unnecessary task to undertake such experiments, especially considering the very small number of trained bacteriologists available at the time.

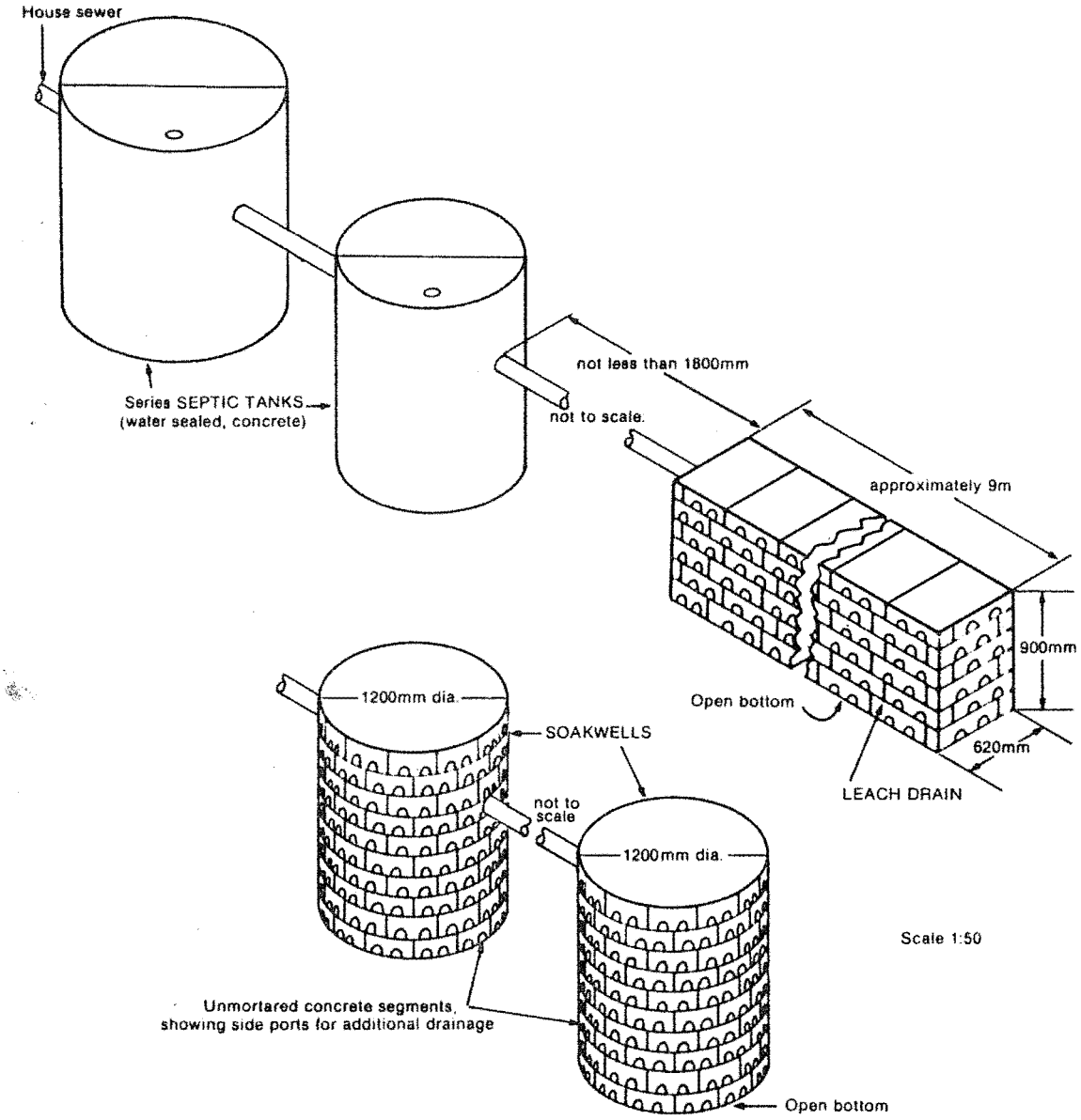
Unfortunately much of the documentation of the ensuing forty years has been lost and publicly available information about the management of sewage disposal in Perth is limited.

In 1955 (PHD 1953), some of the potential causes of sewage pollution of the Swan River were investigated. A thorough survey of the middle reaches of the river was made and two important, but not necessarily obvious,

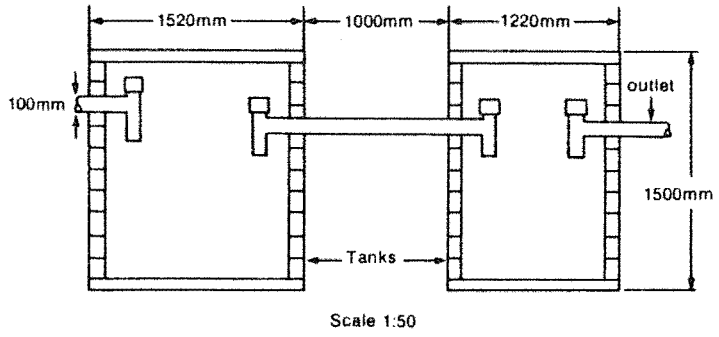
causes were suggested amongst other more visible causes such as animal yards. These were the discharge of effluent and sullage wastes to storm-water drains with ultimate disposal to the river; and seepage from septic tank systems (see Fig. 3) to storm-water drains when laid in close proximity. Several malfunctioning systems were described in detail. This extensive survey is the only one still extant in available archival sources that offers any guidelines for appropriate septic tank management.

Until relatively recently, it has been a corporate objective of the two major State government instrumentalities (the Public Health Department, and the Metropolitan Water Authority) that reticulated deep sewerage should be provided for the entire city. A report on waste management (PHD 1974) suggested that deep sewerage could be provided within ten years. Five years later, a further document discussing community wastes took the unavoidable view that septic tanks would have to be used permanently in some areas (PHD 1979).

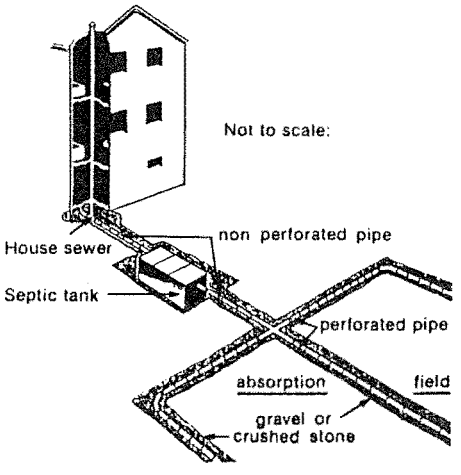
Some estimates have been made for the cost of providing deep sewerage for the whole city. Binnie International (1977) published figures showing that a total sewerage project might cost $A1000 million, and might take some 100 years to complete. The most recent estimates (MWB 1980; MWB 1981) are somewhat less, but nevertheless prohibitively expensive. It appears inevitable that "on-site" sewage disposal systems will be a permanent feature in Perth.



ISOMETRIC PROJECTION OF COMPLETE SEPTIC TANK SYSTEM
(leach drain installed as alternative to soakwells in localities with high water tables: complete system normally buried beneath garden no closer than 300 mm to ground surface.)



Cross Section of Septic Tank Installation



Tile Drain System—U.S.A. (US PHS, 1967)

Figure 3 Septic tank systems, compiled from Health Act (WA), 1911-1978 and US PHS (1967).

It is not surprising perhaps, that if the corporate aim since the first awareness of groundwater pollution has been to abandon the use of septic tanks, there must have been little impetus to undertake research into design, function or environmental effects of septic tanks.

Western Australia is not exceptional in this respect. For similar reasons, other countries have hitherto paid only minimal attention to the design, function and environmental effects of septic tanks in the widespread belief that they were a temporary measure. Financial stringencies world-wide and the increasingly higher capital expenditure needed for sewerage programmes have led to the re-evaluation of septic tank systems. The review of Thomas et al. (1960) and the study of Wilson et al. (1979) are typical of the changing attitude of those concerned with sewage management to what has been regarded as an inadequate means of sewage disposal. Thomas et al. (1960) suggested an eleven point research programme including design of tank(s); possible effluent disinfection methods; assessment of self-purification of polluted groundwater; a study of the evaporation and transpiration of water during soil absorption of effluent; investigation of soil clogging and hence more rational design of absorption systems. In the intervening years, several groups in the U.S.A. have devoted substantial effort to these problems.

Properly managed septic tanks should cause no pollution of the environment. In a pleasant modern suburban area we

might conclude that there was no problem: we detect no odour, there are no gross aesthetic problems and, as will be discussed later, no apparent disease association. However, the widespread use of uncontrolled on-site sewage infiltration systems may still be a health hazard under some circumstances. Whether or not there is a health hazard has become a very much more subtle question, involving in large part, an understanding of the ways in which the microbes of concern interact with the soil and groundwater environment.

1.2 The potential health hazard of septic tanks

The strict definition of a public health hazard associated with properly functioning septic tanks may be stated as existing when pathogenic microbes in the faeces or urine from an individual pass into the septic tank, through the sub-soil, into the groundwater and are ingested by another individual who then becomes infected: the faecal-oral route of transmission is completed. The recognition of such an event is very difficult, and the prediction of circumstances likely to lead to such an event may be impossible.

Assessing the public health hazard of septic tanks is essentially an assessment of the behaviour of a wide range of pathogenic bacteria and viruses both in the physical environment and in their relationship with man. Clearly the natural histories of a range of diseases are important,

but data derived from the incidence of an outbreak of water-borne salmonellosis for example, may offer no indication of the safety or otherwise of that environment with respect to enterovirus transmission, and vice versa. Moreover, as will be suggested later, it may well be unwise to promulgate management criteria based on the behaviour of coliforms. The interactions of salmonella or shigella, as well as enteric viruses, with soils may be quite different from coliform/soil interactions. As well as these fundamental difficulties, a further potential difficulty arises because of the complex host/parasite relationships of enteric pathogens and man. However, this should not lead to the rejection of epidemiological evidence, but rather to treat it with appropriate caution.

Two approaches can be adopted. Retrospectively, conclusions can be drawn from reported outbreaks of water-borne disease; and, a predictive approach can be taken by determining the behaviour of pathogens and indicator organisms in specified soil environments. Both approaches can at least offer some aid to the management of soil disposal systems. It is important to stress here the need to investigate the likely fate of enteric pathogens in a soil disposal system because, unlike a conventional waste-water treatment plant, there is essentially very little control beyond basic engineering design for a septic tank system. The disposal of effluent _per se_ has taken precedence over the fate of bacteria and viruses in design criteria and little consideration has been given to reduction of microbial populations in effluents.

1.2.1 Epidemiological studies

If septic tanks are indeed a health hazard, then it is reasonable to assume that water-borne disease statistics would reflect this. Water-borne disease statistics in the U.S.A. have been collated and published by the Centre for Disease Control (CDC) in conjunction with the Environmental Protection Agency since 1971 (Merson et al. 1974). Since that time and for some decades earlier (Craun and McCabe 1973) there has been evidence to suggest that septic tanks may be important in water-borne disease in the U.S.A. (Hughes et al. 1975; Haley et al. 1980). However, Hughes et al. (1975) concluded that data from the occurrence of acute water-related disease underestimate the problem. In short, the CDC surveillance programme cannot hope to detect all water-borne disease outbreaks. Extending this to establish adequate epidemiological data for septic tanks is more difficult in view of the need to have precise information concerning not only the patients, the natural history of the disease, but details of the soil and groundwater environment. Such information is very rarely available. For example, Table 1 is a compilation of eight outbreaks of water-borne illness in the U.S.A. selected because of the role of septic tanks and the postulated survival and movement of the infectious agent through soil and groundwater. The list is not exhaustive but is representative. Two major conclusions can be drawn from these data. One is that the use of the septic tanks adjacent to drinking water wells is a risk, and even when distancing criteria were met, microbes

Table 1: Outbreaks of water-related diseases associated with groundwater pollution from septic tanks and similar sources.

Reference	Persons affected	Organism	Water supply	Soil type	Well depth	Lateral distance between water supply well(s) & septic tank	Tracer dye used	Type of population & place	Comments
Neefe & Stokes (1947)	350	Hepatitis A virus	Private unchlorinated well	1.5m "ordinary top-soil" overlying limestone & shale.	63m	46m	No	Summer camp Philadelphia, USA.	-
Mosley & Smither (1957)	18 (9directly)	Hepatitis A virus	Private unchlorinated wells	Detailed log: horizons of clay, various shales, coal, limestones & sands.	25-65m	Closest 9m	Yes: fluorescein into one septic tank to all adjacent wells.	Rural farming community Kentucky, USA.	-
Wilcox et al.(1961)	90 (mainly children)	Hepatitis A virus	Private unchlorinated wells	Limestone	11-39m	"Often insufficient"	No	Rural farming community N. Michigan, USA.	Some wells poorly sealed & cased, 52% faecally contaminated (coliform counts), additional leaching effect of spring snow melt.
Lobel et al.(1969)	454	Not isolated	Private unchlorinated	Not stated	50m	60m (down-hill of cesspools).	Yes: fluorescein, but negative.	Picnic recreation area Pennsylvania, USA.	-
Garibaldi et al.(1972)	95	Hepatitis A virus	Unchlorinated	Stanley shale	15m	18m	Yes: fluorescein (transmission time 20 days).	Cafe in rural Arkansas, USA.	Well shown to be faecally contaminated (coliform counts), soil permitted lateral drainage.
Mack et al. (1972)	Several	Poliovirus II	Private unchlorinated well	Clay to 5m Shale 2.5m	30m	90m	No	Freeway restaurant, Michigan, USA.	-
Baine et al. (1975)	208	Shigella sonnei (drug resistant)	Private unchlorinated wells	Not stated	"Shallow"	400m	Yes: fluorescein, but negative.	Rural School Iowa, USA.	Route of transmission probably via school shower, but not conclusive.
Weissman et al.(1976)	1200	Probably Shigella sonnei (drug resistant)	Public wells	"Porous"	15m	38m	Yes: fluorescein (transmission time 9 hours).	Residential community	Chlorination failure on town water supply.

were able to move considerable distances through the soil (Mack et al. 1972). This is in contrast with the popular literature on septic tanks in the U.S.A. (See for example Warshall (1979), who claims that septic tanks have never been associated with water-borne disease). Secondly, the predominance of coarse or fissured soils was significant in microbial transmission. However, it would be unreasonable to use such limited data in other situations. Whilst the collated CDC statistics and the specific case studies quoted provide some insight into water-borne disease patterns in the U.S.A., they should not be extrapolated without some qualification to other countries. Frequently in the U.S.A. where septic tanks are used, there are also on-site drinking water wells.

In order to use epidemiological data to formulate siting criteria for septic tank systems, it would be necessary, ideally, to (1) record all individuals with water-borne disease involving septic tanks, (2) have a complete soil profile analysis, and (3) obtain information concerning water table or other environmental factors thought to influence the movement of microbes through the soil. Clearly these ideals cannot be attained, and predicting the extent to which malfunctioning or poorly sited functioning septic tanks might contribute to levels of salmonellosis, shigellosis, infectious hepatitis and other gastrointestinal diseases is very difficult, if not impossible, to determine. Moore (1971) discusses epidemiological criteria in the light of some of the well-known documented cases of water-

borne disease and concludes that strict criteria apply in principle to physical, chemical or microbial hazards in the environment. However, fulfilling the criteria for microbial disease is more complex, because of the nature of the host/parasite relationship, and because of the difficulty often encountered in locating and identifying the true origin of a water-borne disease outbreak. Infected individuals may be some distance (both spatially and in time) from the suspected water. No overt illness may be apparent in a primary contact with water, yet secondary person-to-person spread may be significant, and historically the more insidious phenomenon of healthy symptomless "carriers" of disease has been critical. Further questions concerning the survival of pathogens and the minimal infective dose are also relevant. The review of Bryan (1977) includes data from 49 publications on survival of pathogens and 27 publications on clinical responses of adult humans to various challenge doses of enteric pathogens, indicating a wide range of responses. For example, it is usually assumed that salmonella needs to be ingested in large numbers in order to produce disease, yet during the outbreak of water-borne salmonellosis in Riverside, California in 1965 (Collaborative Report 1971) as few as 10^3 organisms per litre produced infection. These authors suggest that lower numbers of salmonella may initiate infection if consumed with water, because of the rapid movement through the stomach, compared with food, which remains in the acid environment of the stomach longer.

Studies performed with viruses fed to volunteers have had two shortcomings: they have used vaccine strains and it is not certain at all that such procedures duplicate natural infection (Plotkin and Katz 1967). However, the consensus of opinion indicates that relatively low numbers of viruses are able to produce infection. A further difficulty encountered with an epidemiological approach to water-borne viral infections is that an infection might be inapparent, or so mild as not to cause alarm. From this it follows that the first person to be infected (where water hypothetically was the route of infection) does not become clinically ill and yet transmits the pathogenic microbe to others, and a water-borne route cannot be established. The distance between source of infection and secondary contact may be a separation in time as well as space. Even where evidence of water-borne transmission could be clearly established, the sickness may go unreported or not reported as water-borne. Clearly, this latter observation applies equally to bacterial infections.

1.2.2 Predictive studies

Research on the transmission of bacterial pathogens through soils in groundwater has been conducted since the earliest recognition of bacterial infections. Recent studies of bacteria and viruses have been reviewed by Pettry et al. (1973); Pettry and Reneau (1974); Burge and Marsh (1978) and Hagedorn et al. (1981). However, none of

the data reviewed by these authors can compare with specific local studies. It might be assumed that the movement of microbes through soils would follow the same patterns in soils having similar textural properties, but there is growing evidence to suggest that this is not so, and no general "models" can be derived for virus adsorption, for example (Goyal and Gerba 1979; Sobsey et al. 1980). Presumably the same holds for bacteria.

Since the 1960s greater attention has been focussed on the pedology, hydrology and microbiology of septic tank effluent disposal. The majority of studies have been conducted in the U.S.A. where it is probable that the potential health hazards of septic tanks are the greatest in the developed world. Federal laws in the U.S.A. have been enacted in recent years with the objective of improving all drinking water (Levin 1978). Such legislation has provided the impetus for U.S. government agencies to commission detailed research programmes in specific areas such as septic tank management (Kriessl 1978). Other U.S. studies have attempted to relate the presence of bacterial contamination in rural water supplies (Sandhu et al. 1979; Lamka et al. 1980) and in residential canal waters (Goyal et al. 1977) to the use of septic tanks and small sewage treatment systems that discharge to soils or waters. The evidence provided by these authors suggests that septic tanks pose a potential health hazard as judged by the presence of bacterial and viral contamination of drinking and recreational waters. The

juxtaposition of the septic tank and the drinking water well may have the attendant risks even when established siting criteria are adhered to. The risks are not lessened by the provision of housing developments in areas that, judged by other criteria, are acceptable, but in terms of pedology and hydrology, are unsuitable.

The situation in Australia is not as potentially serious as in the U.S.A. Australian cities enjoy generally excellent water supply systems and on-site drinking water wells are rare.

In the Perth metropolitan area, it has become popular to install groundwater bores to supplement mains water for garden irrigation. It is not likely that such bores or wells are used for drinking purposes, although it is difficult to prevent people from drinking such untreated groundwater if they wish. Public health education is the only persuasion. By comparison with the U.S.A. it can be expected that the public health risks of septic tank usage are lower. However, certain suburbs of the Perth metropolitan area have large numbers of septic tanks. Some of these suburbs are in areas of high seasonal or perched water tables (see Fig. 4) where subsoils are impermeable or the land is hilly. The Cottesloe soil association where limestone occurs may also present problems with possible saturated flow of effluent. These may be regarded as marginal or inadequate soil environments and it is in these

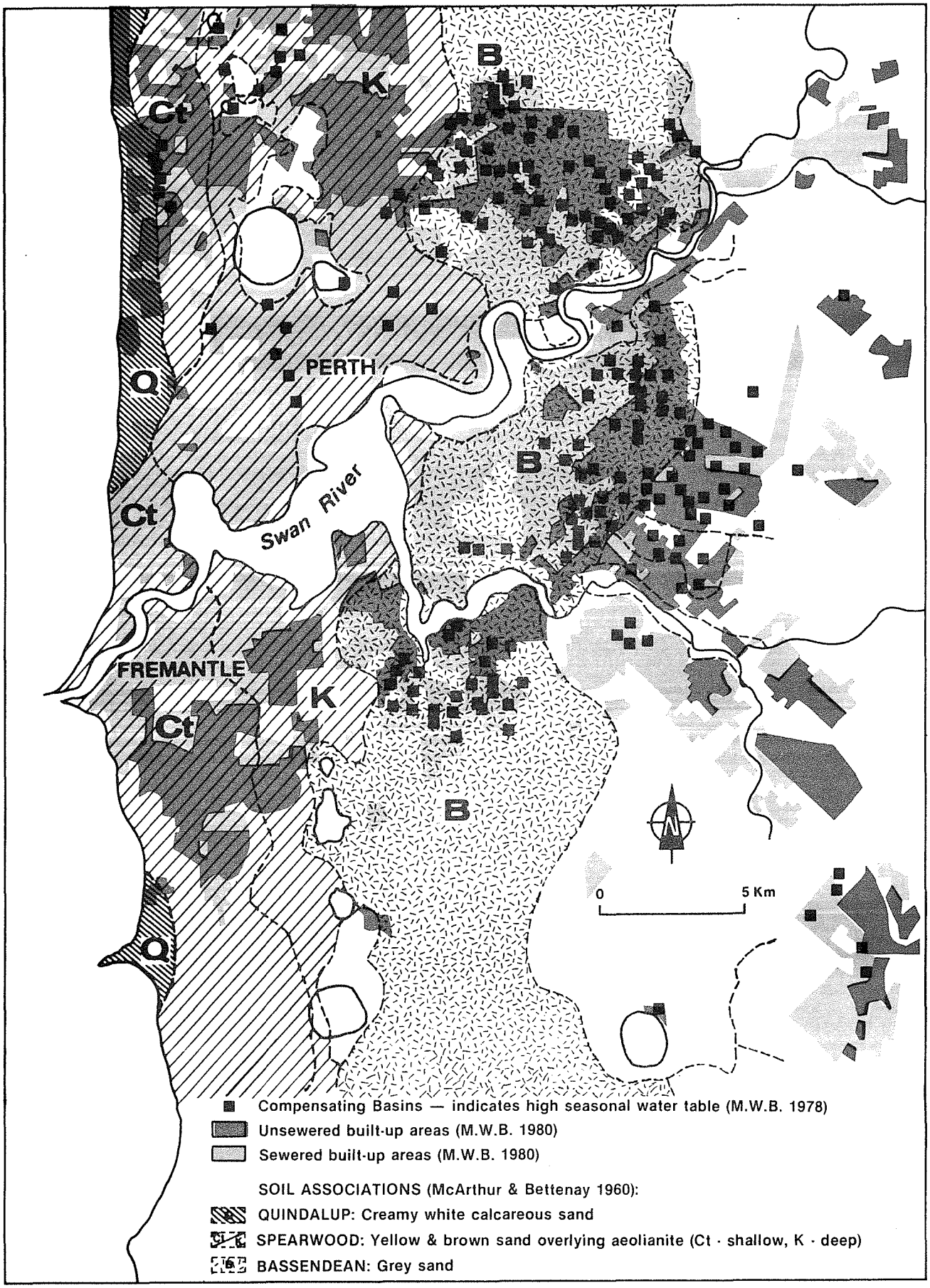


Figure 4 The Perth Metropolitan Area
A composite map showing the major soil types, with an indication of areas with high seasonal water tables, and sewered and unsewered areas.

areas that potential health problems may arise. There might be a risk in ingesting groundwater in these areas: children, for example, will gain access to drainage channels in high water table areas, and compensating basins draining such areas serviced by septic tanks are often landscaped and accessible as children's playgrounds. Another potential route of infection might arise from ingesting groundwater on vegetables that are consumed raw. Swimming pools filled with groundwater may also be a hazard.

Is there then a need for concern? Historically, septic tank installations world-wide have been used as a temporary measure until such time as money becomes available to provide sewerage. They have a poor image amongst public health officials and in some countries they are only tolerated in isolated rural areas (HMSO 1970). The name itself implies disease associations. The efficiency of a septic tank system, which is essentially uncontrolled and unmonitored soil disposal of sewage, probably reaches a peak (with respect to microbe removal) after about one year (Bouma 1975). But we have no ethical way of testing this observation for pathogenic bacteria or viruses, particularly in suburban areas, assuming that sufficient volunteers would permit such research activity in their gardens . Making generalizations on the basis of foreign (and admittedly inadequate) epidemiological data and on the basis of microbial movement in different soils (however closely the analyses compare with the soils of concern) is of dubious value. It is necessary therefore to adopt indirect methods of study

in order to be able to predict the survival and movement of organisms of interest.

1.3 The function of the septic tank and the soil disposal of effluent

The primary function of the septic tank is to reduce the amount of suspended solids in sewage by sedimentation and anaerobic digestion in order to prolong the life of the soil beneath the disposal system. In the early years of experimentation with septic tanks, the effluent was often claimed to be free of pathogenic microbes, and was discharged into any convenient waterway or onto the soil surface (Greenhill 1927; Malan 1964). An extensive field survey in France (Senault et al. 1965) demonstrated the presence of pathogenic bacteria and viruses in a substantial proportion of tested septic tanks. Data for coliform bacteria (Table 2) show that there were often very high numbers present in effluents, with little reduction on passage through the septic tank. Whilst this information does not demonstrate that pathogenic bacteria or viruses will behave in the same way, it confirms that septic tank effluents should be treated with caution. It is now mandatory in most health codes and regulations for effluents to be disposed of by sub-surface irrigation (USPHS 1967; Viraraghavan and Warnock 1975; Health Act, W.A. 1911-1978).

The designs for septic tanks and soil absorption systems specified for Western Australia have been referred to. Fig. 3 illustrates the typical systems and compares systems in the U.S.A. Many antiquated designs are still in use and

Table 2: Characteristics of septic tank effluents.

	Brandes[a] (1978)	Kriessl[b] (1978)		Sauer et al.[c] (1976)		Viraraghavan & Warnock[d] (1976)
Parameter	"Grey" water	"Black" water	Combined	Combined	Raw sewage	Combined effluent
5-day Biochemical Oxygen Demand (BOD_5) mg/L	35-245	38-160	7-480	81-159	397-582	166-478
Total Suspended Solids (TSS)mg/L	25-510	37-261	10-695	31-64	100-372	88-624
p^H	6.5 - 7.3	7.2 - 8.5	-	-	6.6 - 9.1	6.8 - 7.14
TC/mL	6×10^2 to 1.34×10^6	0.3×10^2 to 9×10^3	-	1.3×10^6 to 3.2×10^6	0.11×10^2 to 2.2×10^5	0.24×10^4 to 1.1×10^5
FC/mL	0.5×10^2 to 2.1×10^3	0.2×10^2 to 1.7×10^3	0.1 to 2×10^5	2.7×10^5 to 1.07×10^6	0.06×10^2 to 6×10^4	0.41×10^2 to 5.2×10^4

Legend

a - 18-19 samples (1 site)
b - 151 samples (7 sites)
c - 7-15 samples (1 site)
d - 19 samples (1 site)

it should be noted that they differ very little from the earliest designs of the last century (Philbrick 1883 - Fig. 2). It was common practice to separate toilet wastes ("black" water) and kitchen, bathroom and laundry wastes ("grey" water), but"combined" drainage facilities are now installed.

As sewage enters the septic tank, some of the suspended solids separate from the liquid phase and either settle to the bottom of the tank or rise to the surface and float as a scum. The surface scum forms from less dense materials such as fats and grease. Under these conditions a proportion of the sewage solids is decomposed by bacterial action and the resultant effluent (for an efficiently operating tank) is a relatively clear liquid. However, the household septic tank is something of a design compromise. Efficient sedimentation of suspended solids occurs when the sludge in the tank is permitted to remain in a quiescent state. Retention times in household systems are hard to assess because of the absence of regular monitoring of sludge and scum levels, but retention times may be as short as one day (Patterson et al. 1971). A Canadian study (Brandes 1977) has shown that 2.4 days is a typical retention time for a household system. These estimates suggest that such systems are not particularly efficient.

The biochemical action in a septic tank is the metabolism by bacteria of the organic solids to produce a

liquefaction of solids with the evolution of gases (predominantly carbon dioxide and methane). There is accompanying reduction in sludge volume. The data on basic sewage parameters shown in Table 2 do not indicate high efficiency: effluents have high BOD values and suspended solids are shown to increase in one case. In short, the effluent that is applied to the soil is of poor quality. The effluent from the septic tank drains into the soil through some form of sub-surface chamber. In Western Australia these are commonly soak wells or leach drains depending on the depth of unsaturated soil or soil type and topography. In the U.S.A. and Canada it is common practice to install "tile drains" (see inset Fig. 3) (USPHS 1967; Viraraghavan and Warnock 1975). Tile drains usually consist of unjointed earthenware pipes radiating from a distribution box. The function of these systems is to permit effluent to drain into the soil, and the choice of installing a soak well or leach drain merely reflects, as was suggested, the need for greater surface area when the latter construction is used.

It is desirable that all wastewater throughput should drain freely, yet it is known that purification processes in sands improve as they become clogged and a "crust" develops in or at the infiltrative surface (Bouma et al. 1972). The infiltrative surface becomes progressively clogged and effluent infiltration rates are reduced, ultimately tending to zero (McGauhey and Winneberger 1964). The time taken until such hydraulic failure occurs is not surprisingly

variable (Carbon and Murray 1980). A partial analogy may be drawn here between soil absorption of septic tank effluent and slow-sand filtration. In some cities, drinking water is purified by permitting it to pass through sand filters, and optimum removal of bacteria is normally achieved after a filter has matured and a "schmutzdecke" (Huisman and Wood 1974) is formed. This slimy surface layer on the filter has been shown to have an extensive range of biological activities as well as filtering properties. However, there are two major differences between the two systems. The slow-sand filter operates under aerobic conditions in the light. At the infiltrative surface in a septic tank soil absorption system, no light enters and due to the high BOD of the effluent, the "crust" or slime layer is anaerobic.

It is of interest to understand the nature of the crust or slime layer because it acts as a hydraulic barrier; appears to reduce microbes in percolating effluent (Kriessl 1978); and has obvious economic importance in the management of septic tanks. Whilst a restriction of flow, due to the formation of a schmutzdecke in a slow-sand filter is desirable, the phenomenon has been something of a problem for groundwater recharge schemes where large volumes of water infiltrate into the soil. Loss of infiltrative capacity was first observed in the 1940_S in California. Allison (1947) investigated the role of micro-organisms in soil permeability in ground-water recharge operations. In reviewing the literature, Allison discussed the gradual sealing of soil in terms of the disintegration of soil aggregates and the clogging of

soil pores by microbes and microbial products such as polysaccharides. Using ethylene oxide sterilized soils he demonstrated that permeability did not decline over 60 days compared with unsterilized controls and further, that the loss of permeability was not physical in origin. However, McGauhey and Winneberger (1964) claimed that clogging at the soil/effluent interface was caused by the mechanical effect of ferrous sulphide. This was not substantiated by the experiments of Avnimelech and Nevo (1964) who demonstrated the role of polysaccharides in coarse sand clogging, obtaining severe clogging in the absence of ferrous sulphide. Particularly important was strong correlation of polysaccharides with the loss of infiltrative capacity. The study of Mitchell and Nevo (1964) specifically examined the source of the polysaccharide material by amending flooded soil with organic matter. There was a decline in the abundance of fungi and actinomycetes with a rise in the bacterial population, particularly Flavobacterium spp. These organisms are known to produce yellow to red pigmented colonies on nutrient agar. Pure culture studies of these organisms demonstrated the production of large quantities of polysaccharide material from 0.01% glucose. Further studies to separate the effect of polysaccharide and bacterial cells showed that polysaccharide itself was the major constituent (90%) of the clogging material. It is interesting to note that the field study of Bouma et al. (1972) reported the presence of ">40% yellow, orange, pink or reddish" colonies in plate counts of crust samples.

Daniel and Bouma (1974) examined effluent quality (source) and the process of soil clogging in slowly permeable Almena silt loam. They compared septic tank effluent with effluent from an extended aeration treatment and distilled water, observing percolation rates with undisturbed cores. The data obtained suggested that the nature of effluent solids was important in soil clogging. They observed that extended aeration treatment solids were more finely divided than septic tank effluent solids and gave rise to more rapid clogging.

The importance of crust development and its ability to retard microbes can only be assumed in septic tank absorption systems since there is little comparative data available on very new systems. Analogies have been made with slow-sand filtration and the implication is straightforward: some form of microbial activity needs to occur in or at the infiltrative surface for maximum removal of microbes from infiltrating effluent. The balance between pathogen removal in a system with a developed schmutzdecke against hydraulic failure caused by the schmutzdecke is hard to define.

1.4 Soil environment, soil analysis and texture

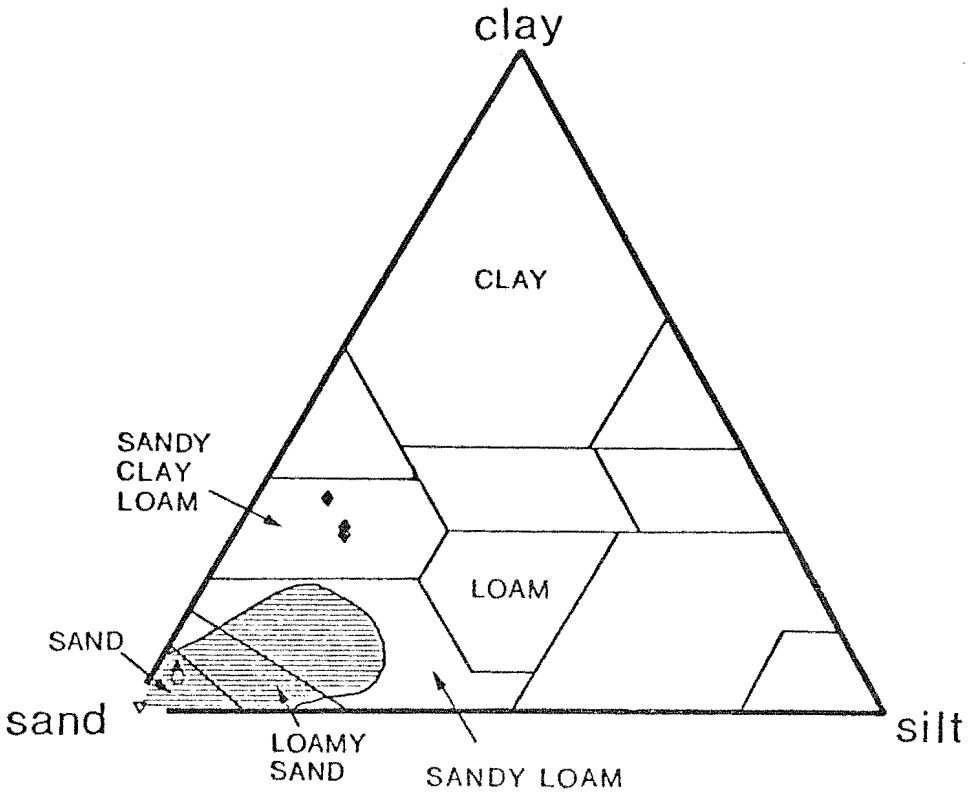
Soils can be analysed by determining the relative proportions of sand, silt and clay. Particle sizing conventions differ, but the International Soil Science Society system adopts the following sizing criteria: sands - coarse 2.0 - 0.2 mm, fine 0.2 - 0.02 mm ; silt 0.02 - 0.002 mm,and clay less than 0.002 mm. The textural

triangle shown in Figure 5 is based on this sizing system. The soils of interest of the Swan Coastal Plain (SCP) are shown on this triangle using the analysis of Whelan and Barrow (1980). In addition, other soils in Virginia and Wisconsin discussed in the text are shown. Clays can be described as the most "reactive" components of a soil. Some clays absorb water, causing soils to swell and shrink on wetting and drying, thus affecting water relationships. Clays are negatively charged and have an electrostatic double layer with exchangeable cations. It is these latter aspects which are important in considerations of sewage disposal: a soil with a significant clay fraction offers a more chemically active matrix than a soil with little or no clay. In this study there is then, a general comparison between a sand with a small clay fraction (Spearwood) and a sand where clay is absent (Bassendean).

The difference is important: Marshall (1976) and a number of other more recent studies suggesting that as the clay content of soils increases, bacterial and viral adsorption increases.

1.4.1 Soil/water relationships

The relative proportions of sand, silt and clay in a soil also determine the porosity, the moisture content under given conditions, the rate of infiltration of water, and the rate of movement of water within the soil. All these parameters are important in understanding the behaviour of septic tank soil disposal systems. Bouma



♦ Virginia Coastal Plain soils

▲ Plainfield Loamy sand

o Spearwood sand

▽ Bassendean sand

Data from Reneau and Pettry (1975)
Bouma et al. (1972)

Shaded area indicates glaciofluvial and aeolian materials.

PARTICLE SIZE DISTRIBUTION OF SWAN COASTAL PLAIN SANDS
(Whelan and Barrow 1980)

SAND	Coarse Sand 0.2-2mm	Fine Sand 0.02-0.2 mm	Silt 2-20μ	Clay <2μ
BASSENDEAN	96.9%	2.7%	1.2%	0
SPEARWOOD	76.9%	18.4%	2.5%	2.8%

Figure 5 Soil texture diagram showing data points for soils studied and other comparative studies. Table shows particle size analysis of Bassendean and Spearwood sands. (See also Bettenay et al. (1960), and McArthur and Bettenay (1960) for further details).

et al. (1972) summarize the importance of the relationship between soil and water in this connection by saying that:

> "... soil absorption and percolation of liquid waste is directly related to, and can be predicted by using the hydraulic characteristics of the soil. In addition, filtration and purification of wastes, while moving in soil materials, will be a function of travel-path and travel time through the very complex soil pore geometry"

It is important to note that coarse sands are somewhat unusual in their soil/water relationships, particularly when the rate of flow of water is considered. Figure 6 shows the time taken for liquid to travel 0.30 m through soil for a range of soils. Soil moisture tension here is related to the degree of capillary suction in a soil: i.e. at saturation (0 on the moisture tension scale) water moves relatively fast, but as the soil dries (+30 mbar) water moves much slower. The implication of this is that coarse sand soils are not efficient at conducting water under unsaturated conditions. Bouma (1975) suggested that the ideal soil for efficient drainage of effluent is a sandy loam. It should be noted that although initial drainage through coarse sands is rapid, once a crust layer has developed, the flow is reduced through the barrier, and moisture tensions below the crust are relatively high. In the operation of a soil absorption system, there is then a compromise between high liquid conductivity (early operation of infiltration system, with poor microbe removal) and a low liquid conductivity (later operation of infiltration system after crust development with better microbe removal, but slower liquid conductivity).

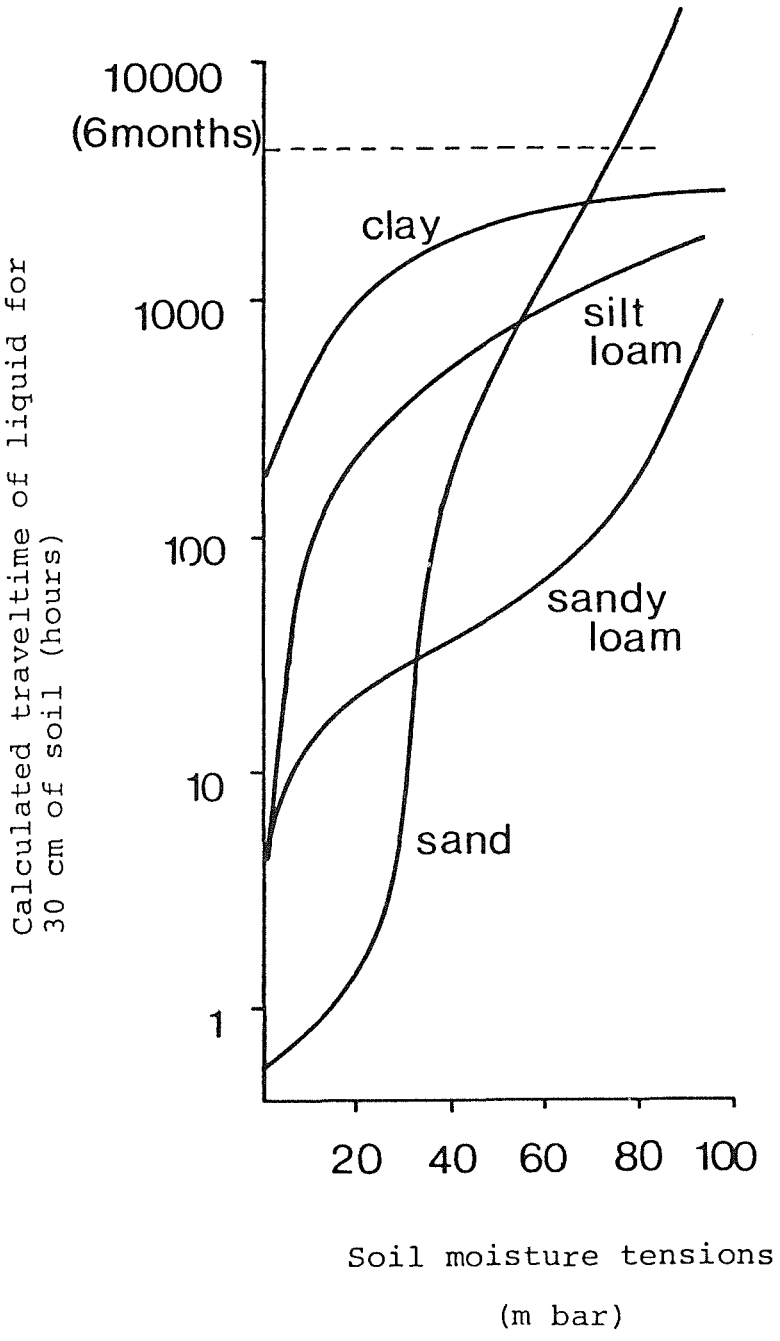


Figure 6 Movement of liquid in soils at different moisture tensions (after Bouma et al. 1972).

1.5 Interaction of bacteria and viruses with soil particles

The essential difference between a saturated and an unsaturated soil is the degree of intimate contact of microbes with soil particles.

Under unsaturated flow conditions, any organism / soil interaction is presumably favoured, and in general the provision of adequate depths of permanently unsaturated soil will ensure that groundwater does not become polluted by microbes. It is desirable therefore to have unsaturated flow conditions to achieve better microbe removal. It is generally observed that bacteria are removed from infiltrating effluents mainly by filtration, sedimentation and adsorption (Gerba et al. 1975), whereas viruses are mainly removed by adsorption (Bitton 1975; Gerba et al. 1975). The possibility that all three processes occur for both bacteria and viruses exists. It may be a matter of the degree to which each phenomenon contributes to removal: bacteria are adsorbed to soil particles and will be discussed below, but the possibility of large aggregations of viruses indicates that viruses may also be subject to filtration and sedimentation. Analogies are often made between sand purification of effluent and the slow-sand filter. In the latter, the filter material can be carefully graded, and particle size ranges selected. According to Huisman and Wood (1974), an ideal "effective diameter" (d_{10}) of sand particles (defined as the size of sieve opening which will permit 10% of the material to pass) should lie between 0.15mm and 0.35mm. On this basis, both Bassendean and Spearwood sands appear to be acceptable (Mathew (1981) has given sizes of 0.15mm and 0.19mm respectively). Sedimentation may be the predominant mechanism of physical removal of microbes where pores exist that are too small for the organism to pass. In theory, all upward facing surfaces are available as "retention sites" for microbes and other suspended particles.

Additionally, inertial and centrifugal forces will tend to deflect

particles away from flow lines and to deposit them on crevices. These forces and differences in flow rates through pores are important. Predictably, the gross differences between bacteria and viruses are likely to give rise to different behaviour in soils, not only with respect to each other, but with respect to different soils. The possible differences between the behaviour of bacteria and viruses have not been studied specifically. Some recent studies of virus breakthrough have been made under a variety of conditions. Differences in breakthrough behaviour due to virus type (Funderberg et al. 1981), carrier fluid (Duboise et al. 1976; Scheuerman et al. 1979) and soil type or condition (Funderberg et al. 1981) have been observed.

There is some evidence (Evans and Owens 1972) which suggests that differences between bacterial types may account for observed differences in movement between E. coli and S. faecalis in land drainage waters. Whether this is due to differences in their chemical surfaces, their shape, their degree of aggregation or all of these remains uncertain from their data. These findings are pertinent to the discussion on which organisms constitute appropriate 'indicators' in the soil environment rather than the aquatic environment. Because bacteria and viruses exhibit a charge it is also important to consider the chemistry of the soil environment in attempting to predict bacterial and viral movement.

Early studies of the interaction of bacteria and soil particles were reviewed by Peele (1936). At that time it was known that bacteria were attracted to soil particles and that a surface relationship existed between the two entities. It was also known that different soils adsorbed bacteria to different extents, and soils with finer fractions were better adsorbents.

Since the pioneering study of Peele, a considerable effort has been devoted to understanding the mechanisms of microbial adhesion to soil particles, and recently, a number of comprehensive reviews have been published (for example, Bitton 1980, Duboise et al. 1979 and Marshall 1980a and 1980b).

At the solid /solution interface there is an imbalance of forces, and the properties of the interface are related to the imbalance. Of interest are dispersive (attractive) and electrical (repulsive) forces.

Dispersive forces, van der Waals forces, or molecular forces are always attractive and independent of charge. The range over which van der Waals forces exert influence is small and is proportional to the sixth power of the distance between particles. Depending on the density and polar nature of the material in question, the relationship is valid theoretically up to distances of 20nm.

Electrical forces arise from the presence of charge on the surface of particles. The charge itself may be due to the ionization of surface radicals such as hydroxyl, carboxyl and phosphate. Such surfaces may be amphoteric, assuming a positive charge at low pH and a negative charge at a higher pH. The specific adsorption of ions at surfaces gives rise to charge, the ions becoming chemically bound, or adsorbed by van der Waals interactions: such as surfactants onto clays. Clay lattices may undergo atomic substitutions and acquire a charge: the replacement of silicon with aluminium is an example. This substitution effect is not dependent on solution composition. Clay minerals present in Spearwood sand are kaolinite and goethite (Marks and Newman 1981). Both of these materials are likely to be important in microbial sorption to this soil.

Very little study has been undertaken to extend the concepts of sorption in the area of land disposal of sewage. Whereas the literature in general describes viral adsorption it does not discuss bacteria in the same terms. The study of Roper and Marshall (1974) for example, appears to be the only work that has examined the survival of E. coli in terms of sorptive phenomena and these authors related their findings to the observed survival of faecal bacteria in sediments (Hendricks 1971; van Donsel and Geldreich 1971). It may be that the emphasis on enterovirus interaction with soil is presently justified in view of the dearth of useful

information. However, in this study an examination of bacterial adsorption is relevant for several reasons. The adsorption of bacteria of public health significance to coarse sands has not been investigated, and because of the general belief that bacteria are removed efficiently in soil disposal systems they have been neglected. The soils in this study are unusual in that they are coarse and have very low reactivity: it is likely that for one of them (Bassendean sand) the sorptive phenomena are minimal.

The degree to which microbes are adsorbed to soils can be ascertained by measuring the supernatant of a soil suspension before and after a suitable contact period and a percentage removal, allowing for death or loss of infectivity,can be calculated.

If different concentrations of test organisms are used in the same sort of test system then information on the capacity of a soil to adsorb the organisms can be obtained. At high organism concentrations the soil's ability to absorb organisms may decrease. Very little study of bacteria has been done in this way, although Hendricks et al.(1979) published an elaborate study of Staphylococcus aureus adsorption to various soils.

By comparison, a large number of virus adsorption studies have been done recently and the consensus of the discussion seems to be centred around models or indicators, and as such it has been noted (Goyal and Gerba 1979) "that no one enterovirus or coliphage can be used as the sole model for determining the adsorptive behaviour of viruses to soils and that no single soil can be used as the model for determining viral adsorptive capacity of all soil types". This is hardly surprising in view of the infinite variety of soils and the variable nature of a given soil surface, as well as the lack of homogeneity amongst enteroviruses.

1.6 Survival of enteric bacteria and viruses in the soil

1.6.1 Bacteria

Enteric bacteria, once removed from the gut environment and introduced into the soil, have limited potential for survival.

The first experimentation in the survival of enteric bacteria in soil was related presumably to the practice of land disposal of sewage.

An important conclusion to be drawn from all of the studies of bacterial survival is that, depending on particular soil environments, temperatures and moisture conditions, some organisms can have extended survival. Baird (1955) was able to isolate Salmonella typhi from sewage-contaminated soil 160 days after last known introduction. Studies of bacterial survival have been reviewed in Parker (1983), however it would seem that data from other experimental situations are of little value in predicting survival in the circumstances of the current study since there are too many variable influences.

1.6.2 Viruses

In contrast with the extensive studies of bacterial survival in soils, very few studies of virus survival in soil have been made. General reviews of virus association with soils have been presented by Duboise et al. (1979) and Gerba et al. (1975), but these contain only limited information on survival. It is not the intention here to present a comprehensive view of virus survival (particularly enteroviruses) because the virological aspects of this report are concerned with a bacterial virus.

The survival of enteric viruses in septic tanks, and soil and groundwater beneath septic tanks has received much less attention than, for example, survival in wastewater treatment processes. This is understandable considering the difficulties of operating in the gardens of private households. However, the results of a recent study (Hain and O'Brien 1979) suggest that Poliovirus I had excellent survival capability in the septic tank system. Measurements

were made using a membrane dialysis chamber. Added virus declined 50% in one day and thereafter more slowly, but less than 1 log total reduction was observed over 20 days. By comparison, coliform bacteria in identical systems declined by 2 logs in 5 days. Dialysis chamber experiments in groundwater observation wells beneath a septic tank were performed and in this case, more rapid virus decline was observed: > 1 log in 24 hours followed by a slower decline over 12 days, a 2 log reduction occurring by day 10. Both of these results can be criticized because exposure to particulate matter was not permitted, but it would seem that the chemical conditions obtaining in these systems were not unduly virucidal.

The application of virus-laden sewage to soils either in small quantities through septic tanks or through wastewater infiltration systems must be viewed with caution. Virus survival is extended by association with solids and soil particles, and although appropriate chemical conditions for adsorption may exist, the elution of viruses is facilitated by fluctuating water tables or rainfall. (Wellings et al. 1975 , Landry et al. 1979). The need to evaluate these problems is stressed by Berg (1971).

1.6.3 Antibiosis and predation in the soil

1.6.3.1 Antibiosis

Bouma et al. (1972) reported the presence of actinomycetes and genera such as Pseudomonas and Bacillus in the soil columns beneath septic tank systems. Noting that all three groups of organisms are potential antibiotic producers they postulated that such organisms might be responsible in part for the die-off of coliforms and streptococci.

Since the discovery of antibiotic production by soil organisms the question has always remained: Are these organisms capable of producing the same antibiotic substances in their natural habitats, and what is the ecological significance of this? The review of Brian (1957) is addressed to this question.

The available evidence then suggested that when nutritional requirements were met (addition of plant debris, nutrients in the rhizospheres of some plants), antibiotic substances were produced. But whether such antibiotic synthesis was antibacterial in normal mixed microflora could only be answered indirectly and was dependent on other variables such as soil type and flora. Consequently the answers were variable. But on the evidence presented by Brian, that high percentages of actinomycetes and microfungi isolated from soils were capable of producing antibiotics, the conclusion was that antibiotic production was quite common among soil saprophytes.

Turning to the highly artificial (or at least abnormal) situation in the effluent infiltration system, can it be argued that "die-off" of coliforms (and presumably pathogens) as suggested by Bouma et al. (1972) could be due at least in part to antibiotic induced death? Brian (1957) wrote that "antibiotic production in the soil is limited by the supply of suitable organic carbon compounds". The recent review of Demain et al. (1979) provides some interesting evidence for speculating on the role of organic compounds from effluent in stimulating antibiotic production in the soil. Antibiotic production did not necessarily require high concentrations of substrate. It was found with β-lactam-containing antibiotic synthesis that cell growth proceeded firstly, and when glucose was exhausted, antibiotic production began.

Another piece of evidence that might favour soil antibiotic production in sewage infiltration systems is that a wide variety of carbon sources (starch, dextrin, sucrose, soy bean oil and glycerol) can be used as preferred substrates in place of glucose in laboratory fermentations. Organic mats at soil/liquid interfaces (Avnimelech and Nevo 1964 ; Mitchell and Nevo 1964) consisting largely of polysaccharides and polyuronides are a potential source of carbon substrates which can leach into the soil. Combined with low

molecular weight compounds in the effluent itself this continuous supply of organic material may provide sufficient substrate for antibiotic synthesis. But the isolation of abnormally high numbers of actinomycetes (even if they are active antibiotic producers) from soil after effluent infiltration does not offer conclusive evidence of natural soil antibiotic activity. It does however suggest a selective pressure and given the input of low levels of appropriate organic material it may suggest a degree of in situ antibiotic production. Whether this activity is adequate to account for observed coliform die-off is another matter. The data of Kristiansen (1981), however, do not support the concept of antibiosis in soils beneath septic systems.

1.6.3.2 Predation

The role of protozoa in conventional sewage treatment plants has been studied at some length. Protozoa are widely distributed throughout all types of treatment and are found in anaerobic as well as aerobic situations. In conventional wastewater treatment plants protozoa are considered to be useful and the reduction of bacteria with and without protozoa has been tested. Reductions of pathogenic bacteria and E. coli have been also observed. Predation was seen as the major factor in aerobic treatment systems.

It seems reasonable to assume that die-off of bacteria in effluent percolating through the soil may also be due in part to predation by protozoa. None of the well-known studies of sewage infiltration systems (of any kind) has in fact referred to a possible protozoan role in die-off. Indeed, the assumptions in general are that lack of nutrients or competition for nutrients are of overriding importance. Thus the question of predation as a contributory factor in the die-off of faecal bacteria (and possibly viruses) in the soil is a matter of speculation. Since protozoa are ubiquitous organisms and are readily isolated from the soil (Darbyshire 1973), it follows that protozoa can be expected in the soil beneath septic tank

systems, but the question remains as it did with antibiosis: Does the demonstration of protozoa in these circumstances necessarily indicate a contribution to observed die-off of faecal bacteria? Testing this relationship for working systems as for other relationships that depend on critically examining soil flora is impossible. Some assessment, however, is possible in column simulation systems.

1.7 Small sewage disposal systems: a review of field and laboratory studies

One of the essential differences between deliberate effluent disposal into soils from treatment plants and "passive" disposal through small disposal systems is the ability to incorporate design and management controls into the former systems. The management of small on-site systems is still largely empirical. However, some research on soil and groundwater pollution from domestic systems has been undertaken and this has been reviewed in the wider context of soil disposal of sewage by Hutchinson (1972) and Pettry and Reneau (1974). A specific review has been given by Hagedorn et al.(1981). These reviews have emphasized the "out of sight, out of mind" attitudes with regard to groundwater pollution. Standards apply worldwide to both air and surface water, but as Pettry and Reneau (1974) suggest "soil pollution remains an undefined entity". This is not to suggest that the installation of septic tanks and similar sewage disposal systems is necessarily haphazard. Regulation of installations is enforced as has been discussed earlier. However, it is unreasonable to assume that uniform siting criteria can be applied universally. In essence, the major criterion for microbiologically safe septic tank siting is the absolute depth of adequate, permanently unsaturated soil between the point where effluent enters the soil and the water table. Clearly this depth varies according to soil texture, homogeneity, composition and local topography. It may be suggested also that such a depth can be defined by the ability to attenuate or remove microorganisms irrespective of "input" concentration. Whilst this latter criterion

may be difficult to achieve, the need to undertake local study is emphasized. Generalizations are not possible on the basis of investigations world-wide.

Field research with small disposal systems is fraught with difficulties. Septic systems are normally located in the gardens of private households, generally beneath carefully tended lawns. Obtaining suitable sites for field study is difficult since householders may not be willing to permit excavations or well installations and frequent intrusions into family life. The site / site variability and the lack of adequate scientific control over selected sites is a further consideration. However, this does not absolve the need for research, particularly in view of the dearth of studies of functioning systems.

The essential question concerning on-site disposal systems is how far do pollutants travel in the soil after discharge. In the following discussion two phases are considered:
a) when pollutants are discharged into saturated soil, and
b) when pollutants are discharged into unsaturated soil.
It has been anticipated that the response of bacteria and viruses to the soil environment would differ between the two phases. Whilst it is convenient to separate studies on this basis, it must be stressed that in natural circumstances there may be no such convenient differences. It has also been shown how important a consideration of time is in terms of bacterial and viral survival. Whether or not bacterial and viral survival is totally dependent on properties of the soil is unclear, at least in the field situation. However, it can be suggested that if survival potential is equal for both saturated and unsaturated conditions, then organisms will move substantially farther in the saturated phase.

1.7.1 Saturated flow studies

Early studies of on-site sewage disposal were made in the 1930s by Caldwell and Parr (1937), and Caldwell (1938a), (1938b). It was common practice to install bored-hole

latrines (described by Caldwell and Parr in 1937 as "essentially a septic tank in immediate contact with the infiltration bed") to penetrate the water table, to ensure septic decomposition. Caldwell undertook her studies over many years to assess the health hazard of latrines, particularly because household water wells were often located adjacently. A bored-hole latrine of the type studied by Caldwell is a hole in the ground sufficiently deep to penetrate the water table with free water at the bottom. At the study site, soil profiles were recorded. At the depth where bacteria would be expected to move laterally from the latrine, the soil was a medium - coarse sand with 9.4% clay. Below this depth, the soil was predominantly coarse sand with a lower clay content. In the 1937 study observation wells were arranged in arcs radiating out from the latrine at 1.5 m intervals to 10.7 m at various depths and screening arrangements to sample different groundwater strata. When the experimental latrine was first operated, Bacterium coli (E. coli) was detected in the 1.5 m wells in deeper, coarser soil in the absence of other pollution markers. The observation was surprising since a "filtration lag" was expected. The B. coli was subsequently detected in wells to the maximum distance of travel of 10.7 m.

Bacterial pollution reached maximum travel in 3 months, after which a regression was seen. The width of the plume did not exceed the original source. The greatest degree of bacterial pollution was observed in test wells installed to sample water moving through coarser horizons. Significantly less bacterial pollution was observed in shallower wells installed to sample water moving through the horizon with 9.4% clay.

In a subsequent study (Caldwell 1938a), the environment was selected so that an impermeable clay layer was present in the soil profile. In this case B. coli was detected in observation wells at 24.4 m from the source, and during 16 months the extent of travel of the organisms regressed to the 12.2 - 18.3 m zone.

Of further practical importance for public health considerations was a study conducted in the same year at the same site (Caldwell 1938b) where an "envelope" of fine clayey sand was placed around a pit latrine. Again, penetration of the water table was made, but no B. coli were detected beyond the "envelope" at any stage, although some chemical pollution was detected in wells at 3.05 m. The velocity of groundwater flow was stressed as an important criterion in all of these studies. Some similar experiments were performed in India by Dyer and Bhaskaran (1943, 1945a, 1945b). In this case the soil was an alluvial clayey silt to a depth of 4.8 m, below which was medium sand. An experimental bored-hole latrine was again surrounded by concentric rings of observation wells. A pumping regime was used to simulate village well usage, but 'gross' B. coli pollution was only observed at the 1.5 m wells in the direction of groundwater flow. Under these circumstances the authors expressed the opinion that soil type was important and that no bacterial contamination of a supply well would occur provided that, in a fine soil of this type, the distance between well and latrine was 6 m, and for a slightly coarser soil, the distance was 15 m. They recommended careful study for coarse soil installations.

In a study conducted in Alaska in a sandy gravel soil, Fournelle et al. (1957) introduced a Streptococcus sp. into an observation well and followed the movement of the organisms in the groundwater in the surrounding area. The results obtained were consistent with studies already quoted. However, the lack of any soil clogging is an important criticism of this study. The same criticism can be made of the studies of Hagedorn et al. (1978) and Rahe et al. (1978), where in both cases the survival and movement of enteric bacteria were tested under saturated conditions. No matter how they are constructed, all septic tank effluent absorption systems accumulate organic slime at the soil/ effluent interface. There will only be a very limited period during the life of a system when a clean soil interface is available. Notwithstanding this

objection the studies of Hagedorn and Rahe and their colleagues are important. Both studies employed an artificial drainfield system back-filled with gravel and covered with the excavated soil. In the study of Hagedorn et al. (1978) two experimental drainfields were dug. One, 0.30 m deep, only penetrated the A horizon of a silt loam. A second, 0.60 m deep, penetrated the B (silty clay loam) horizon; the bottom of the pit was clay loam. Both pits were backfilled with gravel and a plastic inoculation tube placed in the gravel. The organisms used in this case were streptomycin-resistant E. coli and S. faecalis. At both test sites the water table was such that the pits were saturated, and during a second sampling period the groundwater was within 0.05 m of the surface. The results obtained from samples taken from observation wells showed a distinct uni-directional movement of the groundwater, and bacterial numbers in observation wells were affected by rainfall patterns, which in turn caused fluctuations in the water table. It is interesting to note that whereas in the first sampling exercise, all observation wells (up to 15 m) showed a fluctuation of bacterial numbers with rainfall, in the second sampling exercise commencing some 40 days later, only the wells in close proximity to the pits showed any response to rainfall at the end of the experiment. The authors attribute this to dilution and "soil filtration". Comparisons between pits in two horizons showed that bacteria moved faster in silt loam than silty clay loam, although in general, the rapid movement was not explained by macropore presence in core samples (for example). In a complimentary study, Rahe et al. (1978) compared the movement of a streptomycin-resistant E. coli through two soils using 9 m dosing tubes buried to various depths with accessible injection ports at the ground surface. The soils were respectively, a silt loam (the profile containing a heavy clay starting at about 0.80 m), and a silty loam with a fractured saprolite starting at about 0.55 m. Both locations were on slopes.

The most significant observation in this study was the high speed with which bacteria moved in saturated soils.

Hydraulic conductivity data obtained both in the laboratory and *in situ* for the silt loam/heavy clay soil gave figures of 26 cm/hour and 15.2 cm/hr respectively, yet test organisms appeared in wells (at 45 cm depth) at 15 m 1 hour after inoculation. By comparison, the movement of organisms in the second soil was restricted by depth and moved slower (20 m in 12 hours). Gross soil analysis revealed the presence of many macropores in the former soil, which may have been an explanation for the high flow rates observed.

The studies undertaken by Bouma et al. (1972), Reneau and Pettry (1975), and Viraraghavan and Warnock (1976) have been performed with normal septic tank systems or systems only slightly modified (Viraraghavan and Warnock 1976) for experimental purposes. The extensive study of Bouma et al. (1972) was made throughout the State of Wisconsin (USA) and some 20 individual systems were examined. Bacteriological, chemical and physical measurements were made at each site. At one of these sites, located on a filled and developed wetland where the original soil was a coarse sand, no crusting or slime formation had occurred at the soil/effluent interface because of the high water table. Unlike other systems studied in the same soil type where unsaturated flow was possible, the groundwater at this site had a flushing effect. Extensive lateral movement of faecal indicator bacteria was observed. Bacterial counts were made on two occasions. On one occasion, FC in the disposal trench were 1.3×10^3/mL , and in an observation well, 28 m away in the direction of groundwater flow, the count was 1.1×10^2/mL. At the edge of an adjacent waterway a groundwater sample did not yield coliforms, but enterococci (9×10^2/mL) were detected. This latter well was 36 m from the putative source of pollution. It has been observed elsewhere that enterococci tend to survive longer in the soil than *E. coli* (Evans and Owens 1972) and these results may reflect a similar trend. But in order to show conclusively that the septic tank system is the source of pollution a more elaborate study is necessary. Many more

wells, continual monitoring and the possible use of marked strains can be suggested. The difficulties of on-site experimentation may prevent these steps. Compared with other sites in the same soil where unsaturated flow was possible the authors suggest that the presence of groundwater strongly decreased the effectiveness of the soil in purifying the effluent. Reneau and Pettry (1975) were able to observe three septic systems continually for three years in three coastal plain soils of Virginia, U.S.A. The soils examined were sandy loams with about 55% sand, 15-20% silt and 26-30% clay (averaged over profiles taken to 1.8 m depths). Different arrangements of test wells were installed both to obtain samples at different distances and at different depths. Water table data were not published, but described as "high, fluctuating, seasonal". Varying arrangements of observation wells were used but generally they were placed at depths of approximately 0.15, 0.5, 1.5, 3.0 m and at different extended distances, according to site, up to 28 m. The results for all three sites show the tendency for coliform organisms to move considerable distances in the upper strata of the profiles. In deeper strata there was a marked reduction in numbers. The septic tank system at one site had been in use for 15 years. It would be reasonable to expect an efficient removal of organisms at such a site yet there was only a ten-fold reduction in the coliform count between the end of the tile drain and an observation well 6.1 m away. The importance of an impermeable or slowly permeable layer in the soil profile was stressed. Organisms tended to descend to and move along the upper surface of such a layer, in this study, a hardpan. This phenomenon had in fact been observed some decades before (Stiles and Crohurst 1923; Caldwell 1938a).

Viraraghavan and Warnock (1976), during a 15 month study, confirmed that efficiency (in terms of pollutant removal) of an experimental tile drain specially installed adjacent to the normal drainfield was dependent on the depth of unsaturated soil.

1.7.2 Unsaturated flow studies

The movement of bacteria, viruses and other pollutants in saturated soils presents a greater potential hazard than in unsaturated soils. Accordingly, emphasis has been placed on saturated flow studies. However, the study of pollutant travel in the unsaturated phase can provide useful data for assessing soils as sewage disposal media.

The study of microbe movement in saturated soils is relatively simple technically. Small diameter observation wells can be installed appropriately and, not only can samples be pumped for analysis, but data on water table depth and fluctuation obtained. To assess the movement of microbes in unsaturated soils, other techniques must be employed. The time-honoured method of physically extracting samples, shaking them with suitable diluents and examining the microbial flora of the supernatant still remains the most reliable method. Alternatively, ceramic materials of known pore dimensions which may be used as tensiometers can be used to extract soil moisture. There may be difficulties in using ceramic devices for sampling due to losses by adsorption to the ceramic material itself. There is no agreement in the literature on the degree to which reduced counts are obtained with these devices. Dazzo and Rothwell (1974) condemned the use of ceramic cups for bacteriological sampling. However, when used in laboratory columns (Gerba and Lance 1978 and Magdoff et al. 1974a, b) they were apparently trouble-free and gave comparable counts inside and outside the devices. Brown et al. (1979) used porous ceramic cups to collect leachate samples in a study of coliform bacteria and coliphage movement beneath septic tanks and reported that preliminary bench tests showed "rapid flow of FC and coliphage". The study of Wang et al. (1980) revealed large differences when commercially available ceramic cups and tubes and a ceramic sampler constructed by the authors were tested.

The method generally used for assessing microbial flora

of soils is the direct physical extraction followed by dilution and either plating or some form of dilution count. Most of the studies reviewed here have employed such a method.

Caldwell (1938c) undertook a further extensive study of pollution flow from pit latrines under unsaturated soil conditions. She obtained data from three latrines installed on the edge of an excavated cliff. There was 3.7 m of unsaturated soil beneath these latrines (medium sand with about 5-9% silt plus clay). The latrines received the excreta of a family of 5-6 on a daily basis. In the first latrine, no additions were made apart from the sewage. In the second, a specially constructed roof was installed to permit rain to enter the latrine and surroundings. In the third, some 450 litres of water were added daily(comparable with the estimate of Williamson and Cole (1976) for water throughput for septic tanks in Perth). Samples were taken by using a 19 mm diameter tube inserted through the cliff face. The results overall showed a significant correlation between B. coli numbers and moisture conditions. In the latrine with excreta only, no B. coli were isolated 0.30 m below the base of the pit. In the 'rainfall' pit, B. coli were detected at 0.60 m, but only colon-aerogenes flora at 0.90m. In the pit simulating cesspool conditions there was gross contamination at 0.30 to 0.60 m and B. coli were recovered to a depth of 1.8 m. Consistent with studies previously quoted of latrines that penetrated the water table, there was significant regression of faecal flora with time, and after 2 months, B. coli could not be detected below 0.30 m. In the pit receiving heavy water loads, the effect of saturation surrounding the pit was revealed on the cliff face. Significant lateral movement was not observed over one month.

Another latrine was installed with maximally 0.6 m of unsaturated soil (coarse sand with 6% clay). In the latter case it is possible that the soil was at a

greater degree of saturation than expected because of the local rise in water table due to recharge (Hillel 1971), however, B. coli were not detected in adjacent (1.5 m) wells up to 5 months.

Baars (1957) considered the intermittent pollution of fine sandy soils at campsite latrines in the Netherlands. In this study the soil samples were removed physically and shaken with diluent. Portions of the supernatant were then used to assess coliform populations. In the site with well aerated, unsaturated sand (depth to water table 3.5 m) coliforms did not penetrate further than 1.5 m. However, 7 months after the end of the camping season, organisms were still present in the soil. Baars suggested that this result was exceptionally good, and a greater degree of bacterial pollution would be more typical. By comparison, at another site where the water table was only 1 m below the ground surface,there was bacterial contamination of adjacent drinking water wells, although these were 5-8 m deep (lateral distance unstated).

More recently there have been only two studies of bacterial and bacteriophage movement in unsaturated soils under field conditions: by Bouma et al. (1972) and Brown et al. (1979), respectively. The earlier study was conducted in a wide range of soils in Wisconsin and has been referred to above in the discussion on saturated flow.

The contrast between saturated and unsaturated flow in terms of pollutant travel is very marked. Data were obtained in the Wisconsin study for a large number of field sites of functioning septic systems for chemical, physical and bacteriological parameters. Most sites were systems where tile drains (Fig. 3 inset) were installed, although a few soak wells were studied. Samples were removed from specially dug trenches. The soils in the majority of cases were loamy sands or sandy loams (for example, the Plainfield soil, Fig. 5). Effluent infiltration volumes varied between

about 350 L/day up to an estimated 25,000 L/day. In nearly every case FC and FS organisms were removed from percolating effluents within 1-3 cm of downward travel. The TC tended to move somewhat further, confirming the earlier findings of Caldwell (1938c). Water tables were at depths between 3 and 20 m but in some cases there was sporadic occurrence of faecal indicator bacteria in test wells, due perhaps, to perched water tables. At one site such a situation may have been responsible for the recovery of FS in a well laterally situated 15 m from the tile drain. At this site the water table was at 7 m depth.

Ziebell et al. (1975) observed the movement of TC, FC, FS and "total bacteria" in Plainfield sandy loam. Within 0.30 m directly below a septic tile drain the reductions in these groups of organisms were respectively $4x10^4$ fold, $2x10^4$ fold, 300 fold and 600 fold. The total bacterial count at this level was said to be about that found in a control sample. At the same time these authors noted the presence of high numbers of actinomycetes, sporulating bacilli and pigmented bacteria. It has been noted earlier that pigmented organisms such as Flavobacterium spp. tended to predominate in the schmutzdecke.

Brown et al. (1979) made a more intensive study of the movement of coliforms and coliphage f2 beneath septic lines. Three soils (a sandy loam, a sandy clay and a clay) were studied. Septic tank effluent from a system serving nine families was used to dose either lysimeters containing a tile drain section or a field tile drain. Regular sampling (either physical extraction or ceramic samplers) was performed over two years for FC, but coliphage were only monitored in the final year. The results obtained for FC movement in the sandy clay showed a pattern of discontinuous flow typical of saturated flow through root channels and FC were detected at 0.90 m below the input zone. With time, numbers increased and organisms were

more widespread, but after one year a regression to very low numbers was seen, which is consistent with the earlier studies quoted. Predictably, the clay soil was most effective in retaining FC organisms. For the sandy loam, initial depth samples gave high counts at 0.15 m, but these had declined by 0.45m. No deeper samples could be extracted because of the wetness of the soil. In general, FC occurred only sporadically in leachate samples at 1.2 m, and 10-100 fold decreases were noted within 0.05 m. Phage isolation was made using E. coli K12 F^+, but the authors did not make it clear whether or not other phages were investigated in the effluent. It became necessary to 'spike' the effluent with additional f2 phage since natural base levels were low, about 65 plaque forming units (PFU)/mL, and showed a distinct winter decline. After spiking the effluent, f2 was detected at 0.15 m in the sandy clay. After six months f2 was sporadically detected down to 1.0 m after passage through a thin clay horizon. With the clay soil, f2 was detected within 2 weeks at 0.85 m and there was a regression with time to the input zone. Again, f2 occurred sporadically at 0.85 m. In general, the greatest concentration of coliphage (18 PFU/mL) was found 0.10 - 0.15 m below septic drain lines. After spiking with high concentrations of phage, leachate samples were positive on only two occasions. The reduction from effluent to leachate was 10^4 fold.

From these very limited studies of microbe movement in unsaturated soils of a wide range of textures, the general conclusion can be drawn that most soils are excellent media for microbe removal. However, the limitations have not been defined. Bouma et al. (1972) have urged the need for more elaborate study. There must ultimately be some relationship established between septic tanks and real public health hazards. Is the sporadic detection of faecal organisms significant? For example, is the isolation of FS organisms in a well 15 m laterally from the septic system with the water table at 7m an indication

of a potential health hazard? All of the studies quoted have contained data for TC, and in some cases so-called "total bacteria". Caldwell and Parr (1937) observed that colon-aerogenes (TC) flora moved further and survived longer. Assuming that they originated in the faeces, can we ask whether or not pathogenic enteric bacteria can survive in field situations as long as the TC flora? In other words, do the criteria for "indicator" bacteria developed for water environments still hold for soils? The question can be extended to include viruses. It is very unlikely that bacterial models, whether "indicators" or pathogens, can be used to predict virus behaviour in soils.

1.8 Use of laboratory columns to simulate field situations

1.8.1 Bacteria

It has been suggested that necessary experiments to examine the fate of pathogenic bacteria and viruses after soil discharge in operating household systems are unethical. Also, the use of drug-resistant bacteria and vaccine strains of human enteroviruses may not be advisable in suburban areas. Increasing numbers of workers have made use of a wide variety of column systems to obtain data on bacterial (usually faecal indicator organisms) and viral (usually vaccine strains of poliovirus) movement in soils. Most of the column studies that have been undertaken have been designed to simulate wastewater or raw water infiltration systems rather than septic tank systems, and general reviews have been published (Gerba et al. 1975; Bitton et al. 1979). There has been only a very limited number of studies designed to simulate septic tank soil absorption systems.

The major difference between large infiltration systems and on-site septic systems is that the latter are (after variable initial periods before the schmutzdecke appears) subject to continuous ponding and become anaerobic, at least in the surface layers. Such conditions are likely to lead to different behaviour in the soil when

compared with infiltration systems which are often cyclic, with dosing and resting periods, exposed to daylight and generally under complete control. Fig. 7 shows the soil column system constructed for use in this study. Bouma (1975) and Bitton et al. (1979) have both urged the need for appropriate soil science techniques. In terms of soil texture, there are some limitations on what can be achieved in the laboratory and extrapolated with confidence to the field environment. Clearly, it is impossible to replicate in a column the natural environment of a field soil. In a fine textured soil the presence of root channels and macropores will tend to result in local discontinuities of flow as was shown by Rahe et al. (1978). Column studies of microbial movement in such soils after drying, sieving and packing may be quite unrepresentative of the real situation (Bouma 1975). Not only are hydraulic properties likely to differ (Cassell 1974), but natural profile development would be impossible to simulate and soil/gas relations may differ. The technique of taking undisturbed cores has been used (Cassell 1974; Duboise et al. 1976) and may be a useful method, but some compression of the soil is inevitable. But as Bouma (1975) points out, there <u>has</u> to be some reliance on column studies because of the lack of available field systems. At the same time, the results of column studies, particularly with finer soils or artificial soil profiles (Magdoff et al. 1974a, b) should be viewed with these points in mind. The paper of Funderburg et al. (1979), in describing a technique for column packing for example, fails to acknowledge the problem of duplicating field hydraulic conductivity in the column. They presented data on a soil described as a sandy loam soil (although the stated silt and clay content did not exceed 0.6%), yet the authors have made fairly general claims regarding the usefulness of their technique for soils with a clay content that would make them unsuitable for coring.

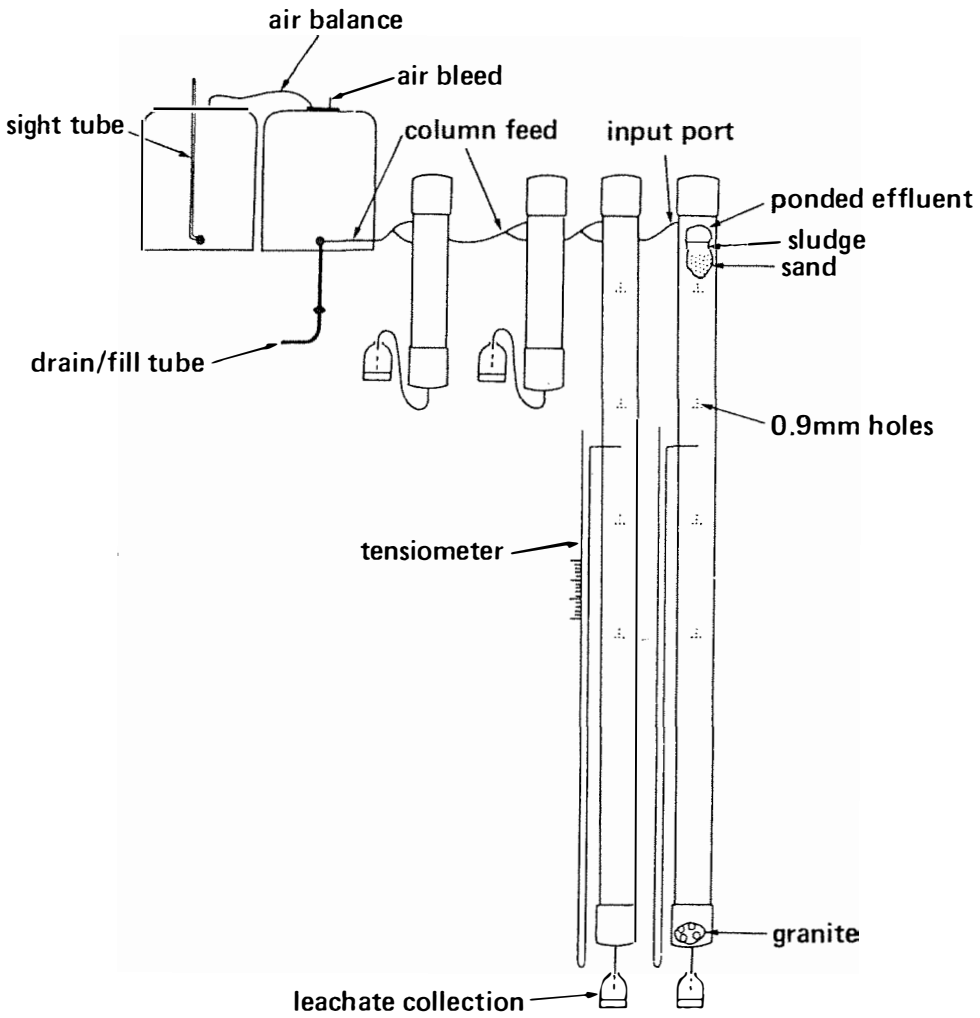


Figure 7 Large column system showing 1.8m and 0.3m columns. Other long columns omitted.

For the coarser, unstructured soils the problems of achieving appropriate hydraulic and moisture conditions are diminished. Sands have been popular materials in numerous studies for this reason. But again, a cautionary note was made by Bouma (1975). He has correctly pointed out that short term studies of septic tank effluent infiltration are not necessarily going to yield data in the same way as a field system that may have been in use for many years, because of slow chemical changes in the soil in response to effluent infiltration. Column systems designed to simulate functional effluent absorption systems must incorporate some degree of clogging at the surface since reductions in flow rates are quite marked (Jones and Taylor 1965). The time taken to clog depends on initial hydraulic conductivity (a function of soil texture), degree of aeration and loading. They showed that for a fine sand the average conductivity was reduced from 23 cm/hr to < 1 cm/hr in less than ten days with continuous ponding. At an infiltration rate of 23 cm/day, for a coarse sand the reduction from about 80 cm/hr to < 1 cm/hr took some ninety six days. Clearly, within the confines of short term experimental projects it might be necessary to accelerate the clogging process.

The Small Scale Waste Management Project at the University of Wisconsin (Kriessl 1978) has published a number of studies of bacterial and viral movement through sandy soils under the conditions described. Ziebell et al. (1975) compared various loading regimes and two soils in 0.10 m (diam) x 0.60 m (depth) columns. Four columns were packed with Plainfield loamy sand and four were filled with undisturbed cores of a silt loam. The sand columns had a tensiometer 0.05 m below the sand surface and holes in the walls for purposes of aeration. The silt loam columns had tensiometers at 0.05, 0.10, 0.20 and 0.30 m below soil surface. Sand columns were dosed at 5 or 10 cm/day, the lower figure being representative of field situations commonly encountered (Bouma et al. 1972). The silt loam

columns were dosed at 1 cm/day which was again representative of field measurements. The difference between the soils was the high clay content of the silt loam (21-27%) and the absence of clay and high coarse sand content (86%) of the loamy sand. Effluent supplies were obtained from a normal residence and supplemented with additional FC and FS cultures. The column inputs and leachates were monitored for 200 days. FC and FS input concentrations were approximately $5x10^6$/100 mL and $7x10^6$/100 mL respectively. Two sand columns were maintained at 25°C. The silt loam columns were maintained at 25°C. Results for FC removal in sand columns were essentially similar for the different flow regimes (5 and 10 cm/day). Breakthrough of FC occurred at about 10 days for both columns and for the 5 cm/day column the leachate FC rose to 10^3/100 mL. For the 10 cm/day column the concentration was $3x10^5$/100 mL. These values were reached after 90-100 days, thereafter declining to, but not falling below 10^2/100 mL at the termination of the experiment. FS organisms behaved differently. They never appeared in the leachate of the 5 cm/day column and appeared only after a long lag (40 days) in the faster flowing column. Whereas FC breakthrough curves were asymptotic, the FS curve was linear to a maximum of about 10^4/100 mL at day 95 and thereafter declined linearly. The removal of FS was greater than FC. FS were only sporadically detected and at low concentrations. It was concluded that although 0.60 m of sand could reduce bacterial numbers (in 100 days, the FC reductions were 94.1% for 10 cm/day column, 99.98% for the 5 cm/day column), it was an insufficient depth for complete removal. Flow regime was also significant as was the slow advent of "bacterial films" building-up on sand surfaces towards the end of the experiment.

The results for silt loam cores were very different. The bacteriological removal was much more efficient under saturated flow conditions. One column however, was inefficient in removing bacteria. It was felt that this was due to "short-circuiting" between aggregates or

macropores. Since flow through large pores was a function of loading rate, the authors reduced the loading to 3 mm/day in order to induce capillary flow in the aggregates with expected greater contact time, and therefore reduction in bacterial counts in the leachate. The reduced leachate counts confirmed this hypothesis.

It is interesting to compare the results of Ziebell et al. (1975) for sand columns with those of Gonchariuk et al. (1962) who dosed a sand filtration system at equivalent loading rates with septic tank effluent containing $2 \times 10^4 - 2 \times 10^6$/mL S. typhi and detected none of these organisms in the leachate after passage through > 0.50 m. After 'biological maturity' was attained the loading could be doubled with no breakthrough of organisms.

1.8.2 Viruses

Studies of animal virus retention in columns designed to simulate field conditions have been reported in Kriessl (1978). Extensive data were provided for a range of columns, generally 0.076 m (diam) x 0.60 m. Other 'mound' type columns were discussed and these data are discussed below. Further information was obtained using very small systems constructed from such materials as plastic membrane filter holder assemblies and these were useful for trial experiments.

Poliovirus I was very efficiently removed from effluent percolating (0.6 cm/day) through silt loam and as little as 0.10 m of soil was required to achieve a ten thousand-fold reduction. Continued use did not alter this efficiency. For sand columns the standard loading rate was 5 cm/day. Experiments were generally conducted at 6-8 ^{o}C. If sand (assumed to be Plainfield loamy sand) was pre-treated ("conditioned") by loading for three weeks with virus-free effluent the column was less retentive of virus than fresh sand, but this finding was not in agreement with the study of Nestor and Costin (1971) who obtained the opposite effect.

In a study using 0.60 m columns, Green and Cliver (1974) found that sand was less retentive for Poliovirus at low temperatures than at 20-22^{o}. In general, the consensus of opinion of the Wisconsin study group is that dose rates and depth of available sand are critical: there are limitations for any soil/loading regimes, and clearly these must be further investigated and a much wider range of viruses tested. The removal of > 99% of poliovirus in 0.60 m of sand under non-ideal conditions is efficient, but leachate concentrations are clearly a function of input effluent concentrations and this figure may be unacceptably low.

2.0 METHODS

Note: Wherever possible in this study the methods used were standard or as close to standard as possible. Greater detail of procedures is given in Parker (1983).

2.1 Microbiological

2.1.1 General

Bacteria and viruses were recovered from liquid samples by direct membrane filtration (MF), but in the case of sludge samples, the organisms were recovered after shaking the samples in dilute mineral salts solution (Vogel and Bonner 1956).

2.1.2 Total coliforms (TC) and faecal coliforms (FC)

Three media were used: 04 ET (HMSO 1969), mFC and m-endo (APHA 1976). Only the latter were employed consistently. The 04 ET medium was used in early trials and discontinued in favour of the more selective mFC medium.

2.1.3 Faecal streptococci (FS)

The medium of Slanetz and Bartley (1957) (MEA) was used generally, but some evaluation was made of the KF medium of Kenner et al. (1961).

2.1.4 Salmonella

The XLD medium of Taylor (1965) was employed exclusively. This medium was found to be excellent when mixtures of *Salmonella* spp. and *E. coli* were assayed, since both organisms could be assessed using one plate.

In general the membrane filtration technique (MF) was used wherever possible, but some Salmonella/*E.coli* counts were made by spread-plate technique.

2.1.5 MS2 coliphage

The method of propagation and enumeration of MS2 phage is given in Parker (1981).

2.1.6 Actinomycetes

The starch-casein agar of Küster and Williams (1964) was used.

2.1.7 'Total' bacteria

Plate-count agar was used according to standard methods (APHA 1976).

2.1.8 Protozoa

The "ring" method (dilution count) of Singh (1946), modified by The Macauley Institute (Darbyshire 1973), was employed.

2.2 Sampling

2.2.1 Effluent

During the course of preliminary effluent surveys and a later effluent and soil survey a standard method of grab sampling was used. Sterile 500 mL bottles suspended by nylon cord were submerged in the effluent and filled, leaving an air space at the top. They were immediately placed on crushed ice and returned to the laboratory within three hours. The subsequent necessity for bulk samples of effluent required a simple pumpable system. Fig. 8 shows the installation used. Such systems were installed in several septic tank systems also being monitored for chemical contents (Whelan and Titmanis 1982). 59 WES*, 21 LUD, 4 JEN and 97 EMP sites had sampling tubes installed. In the first three sites this tube was in the first soak well or leach drain. At the 97 EMP site the tube was placed in the septic tank inside the outlet pipe. Small quantities of effluent could be collected conveniently in a 5 litre flask under vacuum created by a water venturi, but the subsequent need for up to 100 litres per week was met by using a small electric pump.

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* Sample sites throughout the study are referred to by the house number and first three letters of the street name. Site 59 WES provided black water (sewage) only.

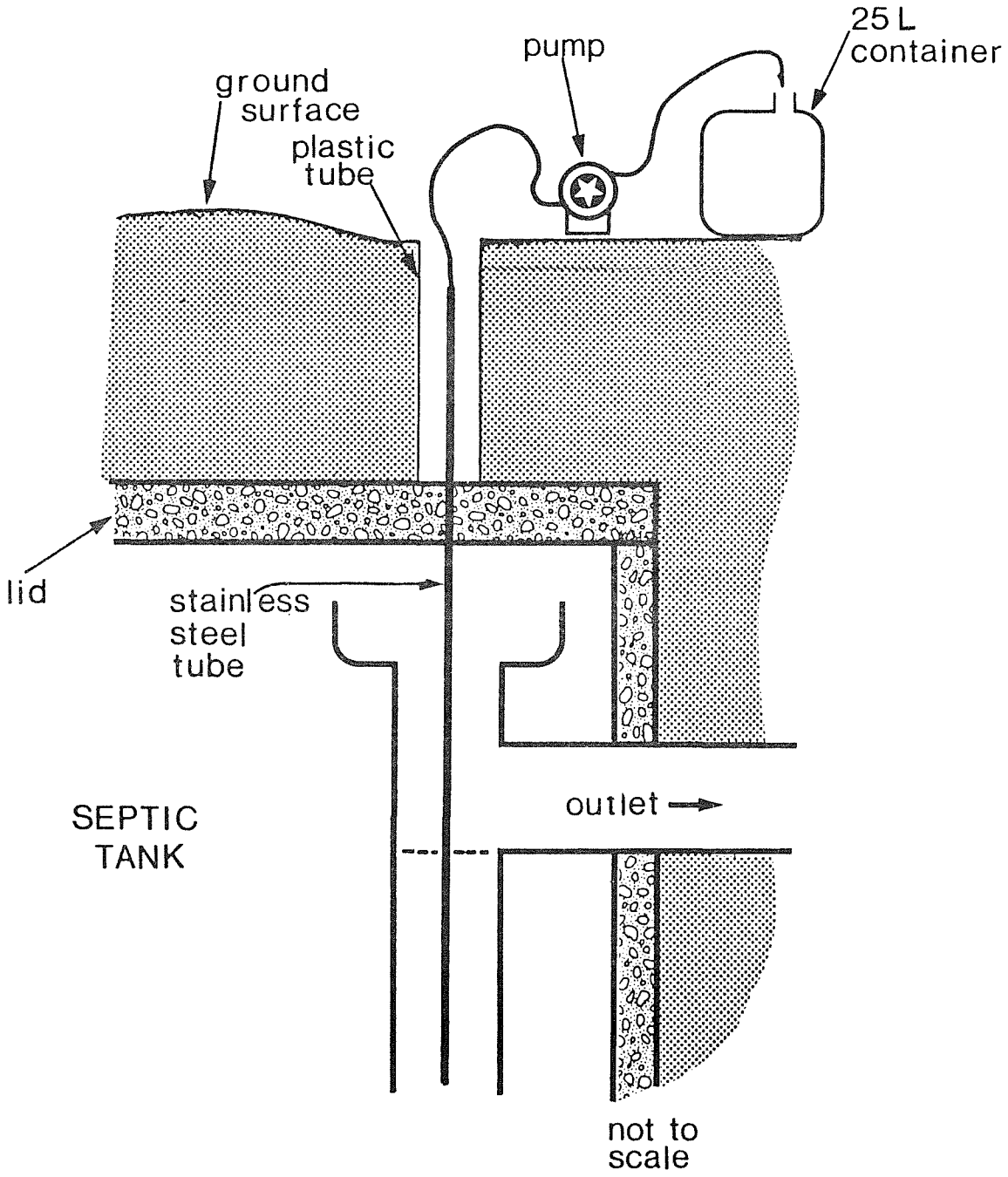


Figure 8 Side view of effluent pumping system installed at site 97 EMP.

2.2.2 Soil

Bulk samples of Spearwood and Bassendean sand were obtained from two suburban gardens in appropriate locations for these sands. Overlying organic material was removed and soils were sampled at about 1.5 to 2 m depth using a spade. Smaller samples were obtained by progressive coring to the same depth using a 50 mm aluminium tube. Samples were stored at 16-18^{o}C in sealed plastic bags. As far as possible, experiments were performed on aliquots of the same bulk sample.

The procedure for taking soil samples beneath a previously drained soak well or leach drain differed slightly from the conventional method which uses one tube with withdrawing and re-positioning. In the case of samples taken through a wet sludge layer, which itself has a high count of faecal bacteria, it was felt that progressive samples down the profile would become contaminated. A double-walled tube was employed and the following procedure was adopted:
The two tubes were cleaned thoroughly and the cutting end washed with 70% ethanol. To obtain as nearly as possible correct and comparable depths on each sampling occasion, the distance between the top of the sludge layer and a stout wooden plank placed across the opened well or drain was measured. Using the plank as datum, the tube was marked appropriately. Progressive coring was achieved by hammering in the whole assembly and withdrawing the inner tube only. A small stainless steel coring device was used to take a 10-15g sample from the cutting edge. The rest of the sample in the cutting tube was discarded, and the tube washed and rinsed with 70% ethanol. The procedure was repeated until a sufficient depth was reached. Four separate core samples were re-combined to give a composite sample. These samples were routinely returned to the laboratory on ice in sealed glass jars. Triplicate counts were made on the composite samples.

2.3 Laboratory preparation of soils

2.3.1 General

In survival, adsorption and column studies, soils were air dried on stainless steel trays at 37^0C for 1 hour and sieved through a 1.68 mm mesh standard soil sieve to remove debris. Moisture contents were determined by oven drying at 105^0C for 24 hours.

2.3.2 Adsorption

The adsorption of both S. adelaide and MS2 phage was measured by the following method:
5 g of test soil was weighed into a sterile plastic universal bottle and to this was added a suspension of bacteria or phage in an appropriate fluid. In all cases, both bacterial and viral suspensions were incubated in 30 mL plastic bottles or agitated by inversion at 18 r.p.m. on a purpose-built tumbler-rack. For time-course studies the mixture was quickly shaken and a sample of the supernatant removed. This was regarded as a t_o value. A control was always employed. This was prepared by taking a soil extract and adding the bacterial or viral suspension as before. In this way, survival could be accounted for. The extract was prepared by taking a bulk soil sample (100 - 500 g) and adding 100 - 500 mL of the fluid in question, shaking for 30 minutes, removing the supernatant, lightly centrifuging and filter sterilizing. Such extracts were stored at -20°C or used immediately. A range of fluids was tested, including large column leachates and distilled water.

2.4 Soil column methods

2.4.1 Large diameter columns: preparation, operation and sampling

Soil columns were packed with air-dried soils using a length of plastic tubing with a plastic funnel inserted in the upper end. To ensure even packing, the soil-filled funnel and tube were withdrawn slowly whilst tapping the column wall.

A range of soil column sizes was used. For long term studies under "ponded" conditions, 0.1m diameter PVC class 9 reticulation piping was used. Two lengths were used: 1.8 m and 0.3 m. In these cases a small quantity of washed and crushed granite ("blue-metal") was placed in the bottoms of columns to a depth of 0.03-0.04m. Large 1.8 m columns had tensiometers installed and the method used for measuring soil moisture tensions was that of Richards (1965).

The general arrangement of 0.1 m diameter ponded columns is shown in Fig. 7. The diagram shows only two 1.8 m columns, although in practice, four such columns were being dosed throughout a period of just under one year. The effluent feed was the same as shown, but the short (0.3 m) columns were placed in front of the four large columns. The columns are identified as follows: Bassendean 1 (Ba 1), Spearwood 1 (Sp 1), Bassendean 2 (Ba 2), Spearwood 2 (Sp 2) (all 1.8 m columns), and Bassendean 3 (Ba 3) and Spearwood 3 (Sp 3) (0.3 m columns). The large columns depicted in Fig. 7 are Ba 2 and Sp 2 and were the farthest from the effluent reservoirs. The columns were set up in such a way as to reproduce the conditions found in a normal effluent absorption system. To this end, they were overlaid with sludge to obtain conditions representative of a septic system at least one year old.

Columns were installed in a large temperature-controlled cabinet constructed by the author and designed to operate at 15^{o}-17^{0}C. (It was established by prior measurement with a

maximum-minimum thermometer placed at the bottom of a 2.5 m sealed tube in the ground, that this was the temperature range during the month of March). The columns were assembled in 0.5 m sections, each section being filled as described. The sections were bolted together progressively into the cabinet framework. The assembly was checked by spirit level and plumb-line. To ensure that the sand was wetted throughout, the columns were wet from the bottom upwards with distilled water. When the water reached the surface the columns were allowed to drain for three days. A layer of soak well sludge was placed on the surface of the wet sand. Originally a thickness of about 0.03 m was tried but this was later increased to 0.06 m because infiltration rates were too high. After sludge application, the tops of the columns were sealed with an air-tight cap and the effluent reservoir connected. Using an air bleed, the dead space above the sludge was filled with effluent to simulate the conditions found in a functioning septic tank system. Four 1.8 m columns were kept running in this way for about 350 days and two shorter 0.3 m columns for about 225 days. Effluent supplies for large columns were obtained on a regular basis from site 97 EMP. Some aspects of large column methodology are given in Parker and Carbon (1981), but since these are necessarily brief, further details are given here.

Column flow rates were obtained by aseptically weighing the leachates. pH and eH measurements were made using a digital meter. On a limited number of occasions the nitrate and ammonium composition of the leachates were measured.

Initially, large columns were ponded with normal effluent. No additional bacteria or viruses were added. The leachate FC levels were monitored during the initial phase. After five months of operation a "challenge" experiment was undertaken to assess the removal of drug-resistant Salmonella spp. and E. coli. This experiment is discussed in Parker and Carbon (1981). Also MS2 phage at high titre were added to the effluent reservoirs to obtain some information on the transmission of viruses.

2.4.2 Large column "dismantling" experiment

In addition to experiments designed to observe flow through large columns, several destructive experiments were undertaken to determine the populations of coliforms, "total" bacteria, actinomycetes and protozoans at specific depths below the soil / effluent interface. For each large column, the procedure was as follows: The effluent supply was cut off and ponded effluent at the top of the column withdrawn. An approximate 25 mL sample of the sludge was withdrawn. By taking the sludge surface as datum, measurements were taken down the column using a plumb-line and the column wall marked at 0.05, 0.15, 0.35, 0.50, 0.65, 0.80, 0.95, 1.10, 1.25, 1.40, 1.55 and 1.70 metres. The wall was cut at these points using a 50 mm hole-cutter. Duplicate soil samples (ca. 50 g) were taken at each point by inserting a sterile 20 mm x 80 mm thin-walled centrifuge tube. The samples were transferred to the laboratory immediately and were used for a number of different analyses.

3.0 RESULTS

3.1 Preliminary survey of indicator populations of "failed" septic tanks.

The coliform and other indicator populations of ten "failed" septic systems are shown in Table 3. In each case the location of the site was obtained at short notice with the co-operation of disposal contractors who had been called by the householder. Either the septic tank was full and incapable of accepting more sewage or the infiltration of effluent had ceased. This preliminary work was done using the 04 ET medium and, as a result of its poor performance, subsequently rejected. However, some useful information was obtained, but it should be stressed that coliforms and "faecal E. coli" counts may have been under-estimated due to background growth on membranes.

Of the ten sites surveyed, six had two soak wells in series; well no.1 draining into well no. 2. Judging by the degree of sedimented solids present at the bottom of the first well, it is quite probable that in all of these cases the first soak well had failed hydraulically or the infiltration rate was very much reduced. The first well in this situation would have been starting to operate like a septic tank. Counts of all indicator organisms showed, in general, a reduction between the first and second well. Some sites showed an apparent increase, but in view of the unreliability of the counting technique, this was probably not significant. In terms of the four indicator types tested, the counts for widely differing sites and conditions were remarkably similar. The notable exception was site 8 SNE, a black water system. Comparing this with site 452 LEN (Table 5c), it can be suggested that if sullage is separated from sewage, the septic tank is more efficient in reducing indicator populations.

This preliminary survey also demonstrated that FS were never found in particularly high numbers. Numerically at least, Cl.perfringens was shown to be a potential test organism.

Table 3: Bacterial counts* in 10 "failed" septic tank systems and some adjacent groundwaters.

Site	Coliforms	Faecal *E. coli*	Faecal Streptococci	*Cl. perfringens*	Comments
8 SNE	9.4×10^2	1.4×10^2	1.14×10^3	nd	Black water only. First drainage in 25 years.
34 DUN	5.7×10^3	2.5×10^3	nd	nd	Second soak well in series of two. 11 years old.
	26/100mL	5/100mL	4/100mL	nd	Well water, 25m away laterally with free standing water at 9.75m. Water pumped from spear below this was negative for test organisms in 2x100mL.
59HAL	9.14×10^4	1.96×10^4	1.4×10^2	nd	Second soak well in series of two.
17 MCL					Two soak wells in series.
s/well 1	1.34×10^4	4×10^3	0.38×10^2	nd	
s/well 2	1.4×10^4	-	0.10×10^2	nd	
24 WAL					Two soak wells in series.
s/well 1	1.52×10^4	1.08×10^4	0.10×10^2	2.2×10^2	
s/well 2	1.48×10^4	2×10^3	nd	2.9×10^2	
	84/100mL	52/100mL	nd	nd	Bore water, 2m from second soak well laterally with water intake at ~~estimated~~ depth of 15m. Counts are for static water. After pumping bore 15 minutes 2 x 100mL sample negative.
41 PAN					Two soak wells in series.
s/well 1	7.4×10^4	1.5×10^4	0.34×10^2	nd	
s/well 2	5.2×10^4	7.6×10^3	0.14×10^2	nd	
6 VER					Two soak wells in series.
s/well 1	1.17×10^5	4.8×10^3	5	9×10^2	
s/well 2	4.88×10^4	6.2×10^3	5	1.28×10^2	
68 MAR					Two soak wells in series.
s/well 1	1.78×10^4	3.86×10^3	0.124×10^2	nd	
s/well 2	2.52×10^4	1.8×10^3	0.315×10^2	nd	
3 BEE					Two soak wells in series.
s/well 1	1.08×10^5	2.1×10^4	7	3.74×10^2	
s/well 2	1.74×10^4	4.8×10^3	0.12×10^2	2.88×10^2	
92 BRO	3.7×10^4	7.6×10^3	< 1	1.15×10^2	First of two soak wells.

Legend
* - all counts per mL except where otherwise indicated.
nd - not done.

The opportunity was provided on two occasions to examine groundwater samples. At site 34 DUN, well water standing free at 9.75 metres was sampled by descending the well and taking grab samples. The well was situated laterally 25 metres from the septic system and the nearest adjacent system was > 25 metres in any direction. Nevertheless, faecal contamination of this water was detected. Bore water pumped from below the water table was coliform-free. On the second occasion (24 WAL), there was no well, but bore water samples taken 30 seconds after pumping were coliform-positive, but became negative after fifteen minutes. It should be noted that this bore was installed adjacent to the effluent disposal area.

3.2 Periodic variation in FC count at one septic tank site.

During the course of bulk effluent sampling at one site (97 EMP), a check was made on the FC population of the effluent. The result is shown in Fig.9. It can be seen that the FC count fluctuates quite widely. It is of interest to know that following sludge removal, the FC count rises to 10^6 organisms per mL suggesting that tank function takes some time to return to normal.

3.3 Survey of fifteen functioning septic tank systems: effluent and sub-soil indicator populations.

The results obtained for ten of fifteen sites are presented in Parker et al.(1981). Since the preparation of that paper, an additional five sites were studied and the discussion which follows is based on results from all fifteen sites, with additional data for FS and Cl. perfringens. Table 4 gives information on all of these fifteen sites in as much detail as could be obtained. Tables 5a, 5b and 5c show results from the fifteen sites located on Quindalup, Spearwood and Bassendean sands, respectively. Similarly, Figs. 10a, b and c show the data graphically, both as numbers and as fractions of effluent values. In these figures, the data points represent the mean values of counts for specific depths for each soil as appropriate. Not all depths have been sampled and the reader is referred to Tables 5a, 5b,

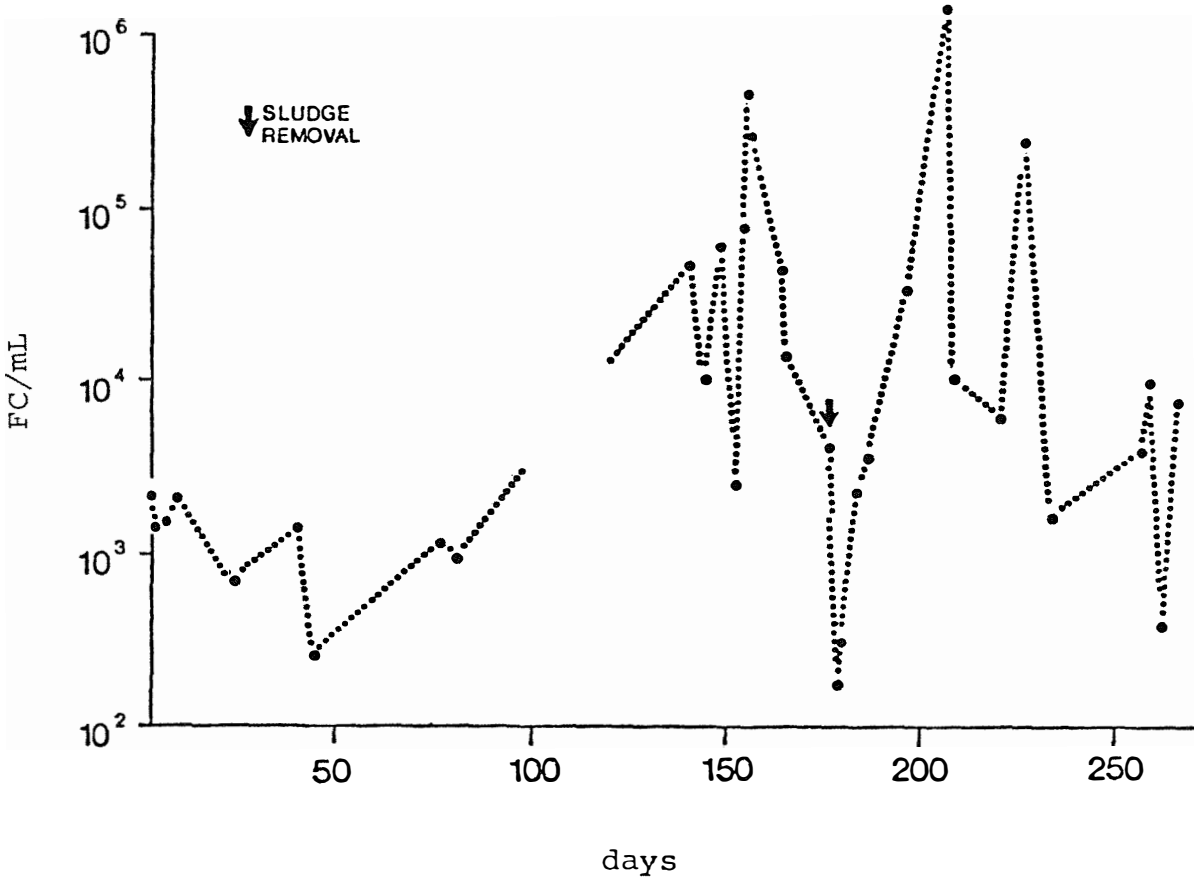


Figure 9 Periodic variation in FC count at site 97 EMP.

Table 4: Site descriptions of septic tank system field survey.

Site	Age	Family and site details
Spearwood sand		
22 HAR (3)*	11 years	2 adults, 3 children. Separate sullage disposal. First soak well sampled, no previous maintenance known, but excessive build-up of sludge, indicating poor septic tank function. Unfenced garden.
48 KEX (4)*	?	2 adults, combined. First soak well sampled, no history available, but evidence of previous heavy usage, excessive sludge build-up indicating poor septic tank function. Dog present.
14 EBE (5)*	14 years	2 adults, 2 children. First soak well sampled, maintenance 3 years prior to sampling. Dogs present.
35 GEO (6)*	4 years	2 adults, combined. Mid-point of leach drain sampled, no sludge at soil interface, but effluent of poor quality with high suspended solids.
16 EBE	15 years	2 adults, 4 children, combined. First soak well sampled, effluent in soak well ponding on garden surface. Excessive sludge at soil interface.
97 EMP	9 years	2 adults, 3 children, combined. Second soak well sampled. Limestone rock beneath system prevented adequate depth sampling or reference hole sampling.
Bassendean sand		
478 LEN (7)*	3 years	2 adults, 2 children, combined. One end of leach drain sampled. No sludge build-up at sampling point. No dogs or cats present.
452 LEN (8)*	25 years	1 adult. Separate sullage disposal. A new soak well had been installed 3 years before sampling. No ponded effluent. Dog present.
450 LEN (9)*	>20 years	2 adults. Separate sullage disposal. First soak well sampled. Excessive amounts of sludge on soil surface & walls, very hard encrustation.
7 ARA (10)*	>5 years?	2 adults, 1 child, combined. Leach drain sampled at midpoint. System on artificial mound because of high water table, elevation about 0.5m. No sludge accumulation. Dog present.
36 LOV	16 years	2 adults, 2 children, combined. Leach drain sampled at end nearest septic tank. Site also subject of dye study.
61 MCG	?	2 adults, combined. Leach drain sampled at end farthest from septic tank.
30 PAR	10 years	2 adults, 2 children, combined. Leach drain sampled at a central section. Effluent ponded on garden surface at distal end. Householder reported a water table at 1.5m. Attempts to obtain groundwater samples unsuccessful because of exceptionally hard ferruginous hardpan at 1.2m depth.
Quindalup sand		
25 SLA (1)*	9 years	2 adults, 3 children, combined. First soak well sampled, unknown maintenance. Dogs & cats present.
4 JEN (2)*	9 years	2 adults, 2 children, combined. First soak well sampled, history of hydraulic failure, sludge dispersed into soil to 0.15m.

* - site numbers as shown in Tables 2-4, Parker et al. (1981).

Table 5a: TC, FC and FS in effluents and the soil beneath septic tank sites in Quindalup sand.

	Site 25 SLA (1)[a]			Site 4 JEN (2)		
Sample	TC	FC	FS	TC	FC	FS
Effluent sample	$9.66 \pm 1.48 \times 10^4$	$3.45 \pm 0.42 \times 10^3$	$5.6 \pm 1.25 \times 10^2$	$1.808 \pm 0.32 \times 10^5$	$6.14 \pm 0.47 \times 10^4$	$2.55 \pm 1.32 \times 10^2$
b 0.15m	$1.59 \pm 0.38 \times 10^6$	$0.416 \pm 0.11 \times 10^2$	nd	s a m p l e u n o b t a i n a b l e		
b 0.35m	$3.95 \pm 2.0 \times 10^2$	$0.126 \pm 0.02 \times 10^2$	nd	tn	$6.05 \pm 0.75 \times 10^2$	$2.94 \pm 0.76 \times 10^2$
b 0.50m	$8.45 \pm 5.06 \times 10^2$	$0.115 \pm 0.01 \times 10^2$	nd	tn	nd	nd
b 0.65m	nd	nd	nd	nd	$1.15 \pm 0.113 \times 10^2$	nd
b 0.80m	nd	nd	nd	nd	$0.60 \pm 0.09 \times 10^2$	nd
*Reference sample	$0.224 \pm 0.02 \times 10^2$			0		

Legend

a - numbers in parenthesis indicate site described in Parker et al. (1981). All sites described in Table 4.

b - depth below effluent / soil interface at which sample taken (metres).

* - reference sample taken from approx. same depth as 0.35m sample and at distance of 3m from effluent drainage system.

nd - not done.

0 - no organisms isolated (detection limit - 2/gram).

tn - membranes too numerous to count.

Note: Means and standard deviations are reported, except in cases where single counts made and which are indicated by the symbol + (see Tables 5b and 5c).

Table 5b: TC, FC, FS and Cl. perfringens in effluents and the soil beneath septic tank sites in Spearwood sand.

	SITE 22 HAR (3)		SITE 16 EBE (15)+				SITE 48 KEX (4)	
Sample	TC	FC	TC	FC	FS	Cl. perfringens	TC	FC
Effluent sample	$3.2\pm0.93\times10^3$	$1.58\pm0.3\times10^2$	8.76×10^4	4.0×10^4	0.415×10^2	5.43×10^2	$4.32\pm0.41\times10^4$	$2.69\pm0.96\times10^4$
b 0.15m	0	0	1.50×10^2	1.2×10^2	nd	4.18×10^3	$1.528\pm0.44\times10^3$	$6.57\pm1.14\times10^2$
b 0.35m	nd	0	0		nd	nd	8.75±3.98	< 5
b 0.50m	8.38±3.78	0	0		nd	nd	0	0
b 0.65m	$0.23\pm0.049\times10^2$	0	nd		nd	nd	0	0
b 0.80m	nd	nd	nd		nd	nd	0	0
b 0.95m	0	0	nd		nd	nd	nd	nd
*Reference sample	15.46±3.18		0				0	

Legend as Table 5a.

Table 5c: TC, FC and FS in effluents and the soil beneath septic tank sites in Bassendean sand.

	SITE 478 LEN (7)			SITE 452 LEN (8)			SITE 450 LEN (9)		
Sample	TC	FC	FS	TC	FC	FS	TC	FC	FS
Effluent sample	$1.53\pm0.28\times10^5$	$1.98\pm0.58\times10^3$	<5	$1.86\pm0.69\times10^{4c}$	$1.55\pm0.44\times10^{3c}$	$1.2\times10^{2+c}$	$1.2\pm0.58\times10^2$	$0.40\pm0.24\times10^2$	$2.10\pm0.76\times10^2$
b 0.15m	tn	$1.18\pm0.11\times10^2$	0	$1.61\pm0.32\times10^2$	$0.72\pm0.67\times10^2$	<5	0	7.75±3.4	$1.97\pm0.18\times10^2$
b 0.35m	$1.41\pm0.27\times10^2$	5.36±3.24	0	7.1±30.2	<5	nd	0	0	nd
b 0.50m	$5.17\pm1.29\times10^2$	7.68±2.8	0	0	0	nd	0	0	nd
b 0.65m	nd	nd	nd	Samples unobtainable, insufficient headroom for sampling device.			nd	nd	nd
b 0.80m	nd	nd	nd				nd	nd	nd
*Reference sample	0			0			0		

Legend as Table 5a.

c - no ponded effluent, soil surface sample.

SITE 14 EBE (5)		SITE 35 GEO (6)			SITE 97 EMP+			
TC	FC	TC	FC	FS	TC	FC	FS	Cl. perfringens
$2.46\pm0.31\times10^5$	$1.32\pm0.27\times10^4$	$2.26\pm0.65\times10^5$	$1.04\pm0.1\times10^5$	$8.53\pm4.67\times10^2$	7.91×10^5	7.625×10^3	0	0.94×10^2
< 5	0	0	0	$2.36\pm0.68\times10^2$	1.32×10^4	0	0	0.50×10^2
$0.11\pm0.024\times10^2$	< 5	0	0	0	Underlying limestone			
$0.17\pm0.034\times10^2$	$0.15\pm0.04\times10^2$	$2.14\pm0.57\times10^2$	$0.80\pm0.25\times10^2$	0	rock prevented			
nd	nd	nd	nd	nd	further depth			
nd	nd	nd	nd	nd	sampling			
nd	nd	nd	nd	nd				
0		0			nd			

SITE 7 ARA (10)			SITE 36 LOV (see also Whelan & Parker (1981)	SITE 61 MCG+			SITE 30 PAR+			
TC	FC	FS	FC	TC	FC	FS	TC	FC	FS	Cl. perfringens
$2.08\pm1.38\times10^5$	$1.15\pm0.48\times10^3$	$0.58\pm0.24\times10^2$	$1.65\pm0.85\times10^4$	1.09×10^5	6.0×10^3	0.35×10^2	1.55×10^4	1.4×10^4	<10	2.12×10^2
$2.32\pm0.27\times10^3$	$0.80\pm0.26\times10^2$	$1.08\pm0.63\times10^2$	$3.24\pm0.16\times10^3$	0.64×10^2	3.2×10^2	1.573×10^3	1.06×10^4	5.7×10^3	0	3.55×10^3
tn	$0.59\pm0.78\times10^2$	$0.56\pm0.18\times10^2$	$0.82\pm0.86\times10^2$	0.46×10^2	0.46×10^2	5.20×10^2	0.75×10^2	5.4×10^2	0	2.25×10^2
$6.55\pm0.75\times10^2$	$0.52\pm0.06\times10^2$	nd	0	0	0	7.2×10^2	<10	0	0	0.713×10^2
nd	nd	nd	0	nd	nd	nd	0	0	0	nd
nd	nd	nd	<5	nd	nd	nd	nd	nd	nd	nd
<5			0	0			0			

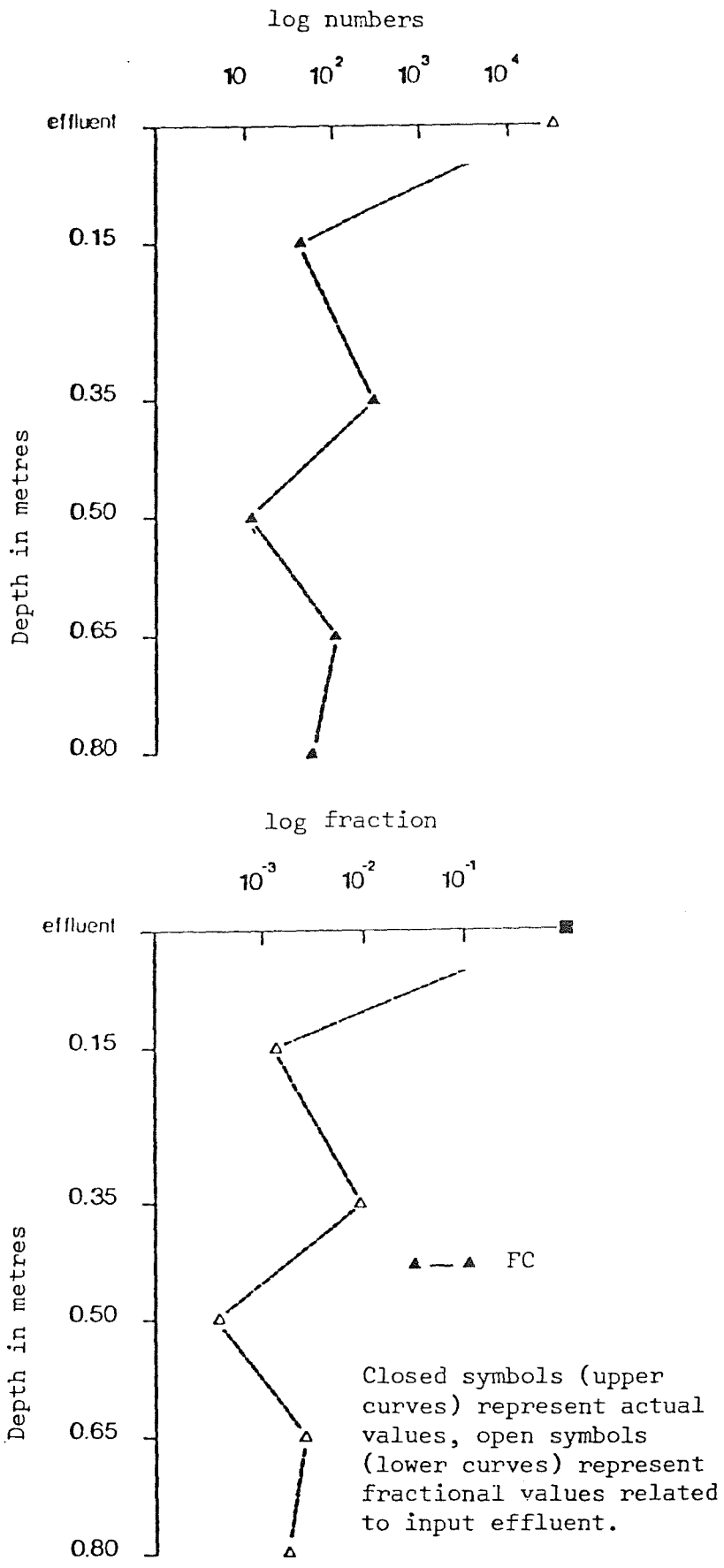


Figure 10a TC, FC and FS in the soil beneath septic tanks in Quindalup sand.

Each data point is the mean value of a number of values shown in Table 5a.

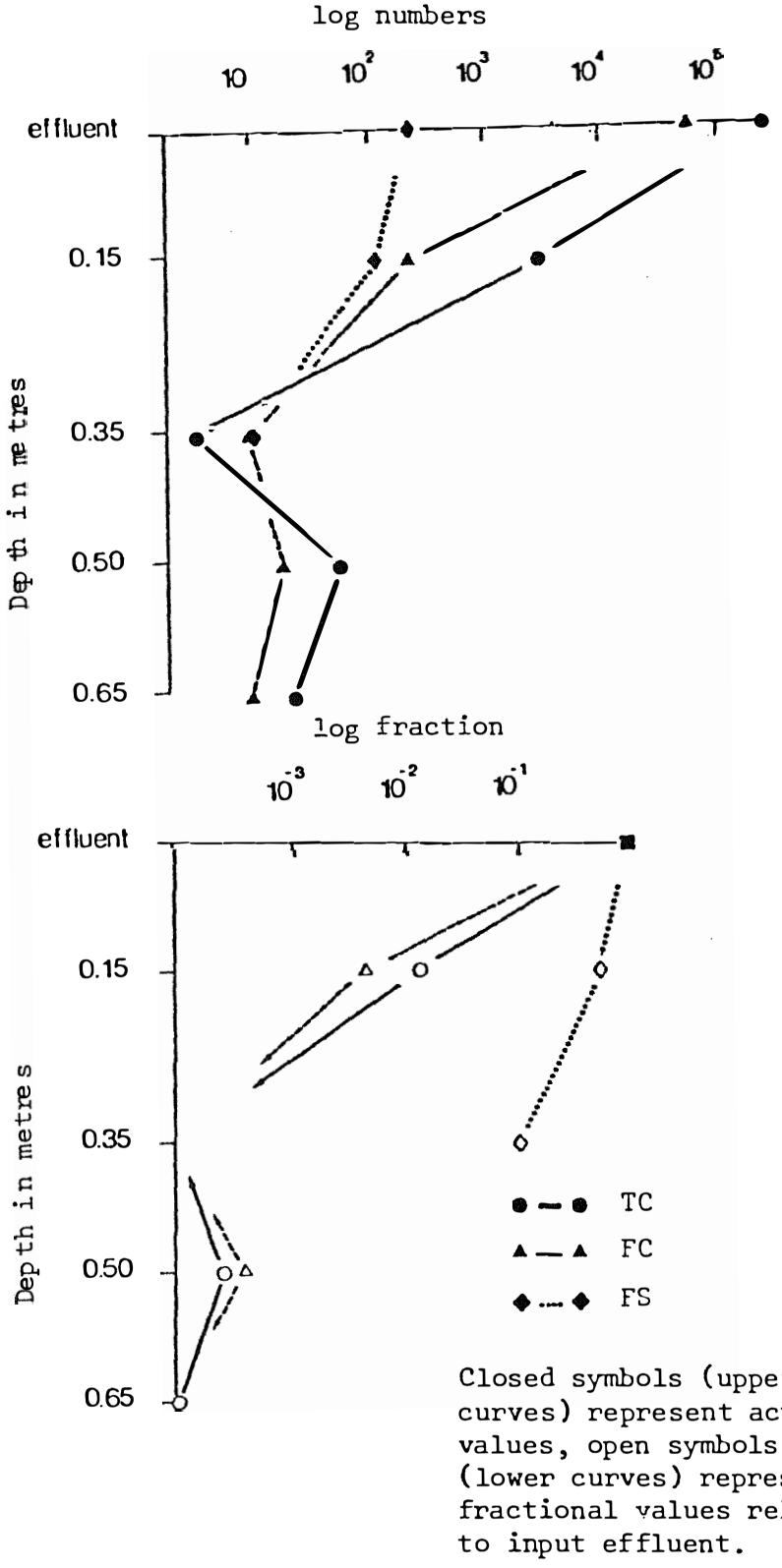


Figure 10b TC, FC and FS in the soil beneath septic tanks in Spearwood sand.

Each data point is the mean value of a number of values shown in Table 5b.

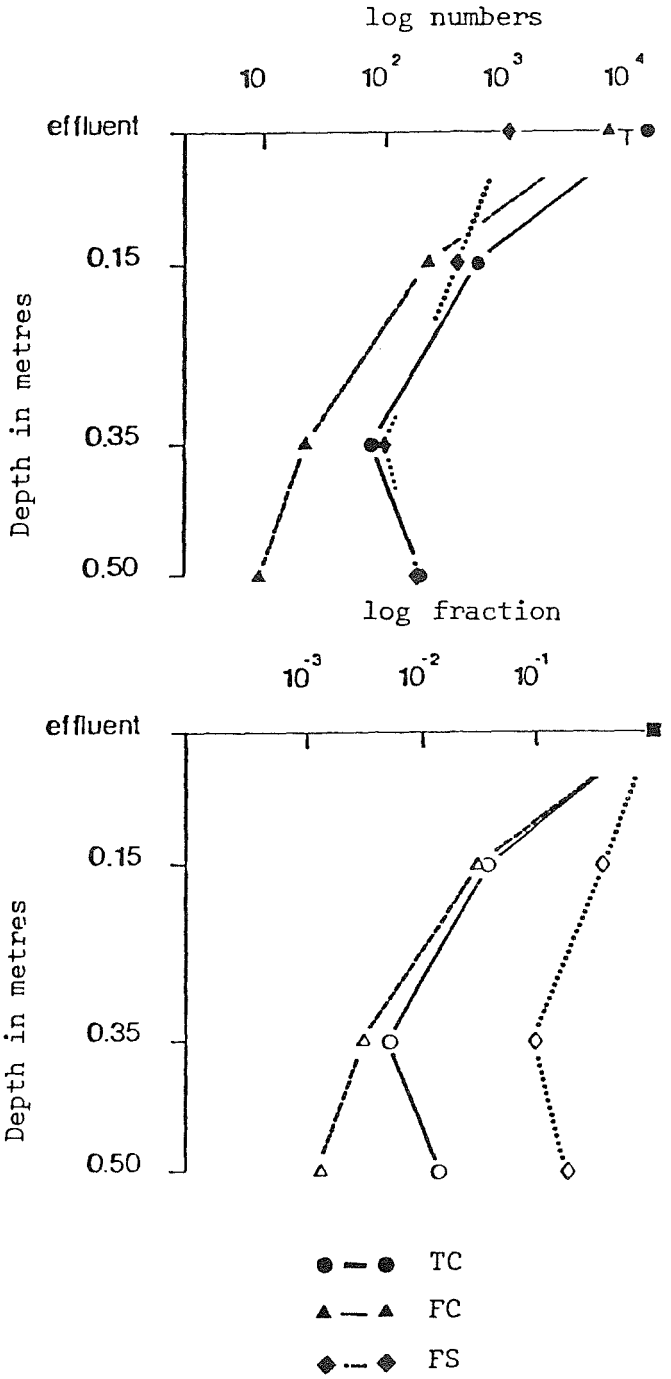


Figure 10c TC, FC and FS in the soil beneath septic tanks in Bassendean sand.

Each data point is the mean value of a number of values shown in Table 5c.

and 5c for more precise information regarding omissions and variability of counts.

The effluent counts for the fourteen sites where effluent was present showed a wide range for all of the indicator populations sampled: TC $1.2 \times 10^3 - 7.91 \times 10^5$; FC $0.4 \times 10^2 - 1.0 \times 10^5$; FS $0 - 2.19 \times 10^3$; and for the three sites where Cl. perfringens was assessed, $0.94 \times 10^2 - 5.43 \times 10^2$. The data for FS and Cl. perfringens can be compared with the earlier data (Table 3) and the results are essentially similar. No direct comparison can be made between coliform counts because of different methods, but it would appear that "failed" or "failing" systems did not give higher counts than functioning systems.

When the data from the additional five sample sites (2 Spearwood, 3 Bassendean) are included, it can be seen that the results are similar. There was a greater reduction in numbers of TC and FC between effluent and sub-soil for Spearwood sand than for either Bassendean or Quindalup. The reduction of FC counts to zero at a depth of 0.15 m in Spearwood was a frequent observation. For Bassendean and Quindalup these organisms were able to penetrate further. Further sampling for Quindalup may be necessary to confirm these findings, since at one Quindalup site (4JEN) excessive interface disturbance may have caused the deeper penetration found. The data for TC and FC taken overall for Spearwood and Bassendean sands demonstrate that the latter soil is less able to remove these organisms from percolating effluent. This phenomenon is more apparent if the fractional treatment of the data is considered. At a depth of 0.15 m, only an approximate 50-fold reduction was seen, compared with an approximate 100 to 500-fold reduction for Spearwood. At greater depths, the same pattern is maintained. For Spearwood sites the concentrations of TC and FC reduced to less than 10 per gram or to zero at 0.35 m, with an apparent increase at 0.50 m followed by further decline with depth. For Bassendean, the data show less rapid decline with depth and the maintenance of appreciable levels of TC at 0.50 m. Deeper samples would be

necessary to establish the full extent of penetration, but it should be noted that work on Bassendean sand sites (with the exception of 36 LOV (Whelan and Parker (1981)) was completed before the Spearwood work.

The limited FC data (Table 5a, Fig. 10a) for Quindalup show that, although initial reductions are as good as in Spearwood, there is effectively no further reduction over the depths sampled.

The results with FS as indicators of soil pollution reveal a quite different picture. It must be stressed here that FS, when present in effluent, were at lower concentrations than the coliforms, and some counts varied widely. For this reason, some caution is warranted in making comparisons between FS and coliform behaviour between soils. Considering mean effluent counts, it can be seen that values obtained from Bassendean sites were slightly higher than Spearwood. (This contrasts with the opposite pattern for coliforms.) However, irrespective of soil types, FS behaviour was quite different from TC or FC. There was little effective decline for either soil between the effluent and the 0.15 m depth. In real terms, there was only slightly more than a 10-fold decrease between effluent and 0.35 m for Spearwood sand and approximately the same for Bassendean. In the latter case, a slight increase was apparent at 0.50 m. If the soil data are compared as fractions of effluent counts, it can be seen that no decline occurred for the data available for Spearwood, and for Bassendean the decline overall was less than 10-fold.

Wherever possible, a reference sample was taken at a depth equivalent to 0.35 m below the soil/effluent interface of the infiltration system. The location of the hole chosen was to be sufficiently far away from the septic system to be beyond any lateral moisture influence or possible pollution caused by prior flooding of effluent. Accordingly, 3.0 m lateral distance was felt to be adequate. Such samples were only tested for TC. In general, the results were negative.

Positive samples were obtained from sites 25 SLA, 22 HAR and 7 ARA. Site 22 HAR was an unfenced, unkempt garden, and sites 25 SLA and 7 ARA fenced. The two latter sites had dogs and cats, and a dog respectively. The presence of animals or open access to stray animals cannot necessarily be correlated with positive results, but it is possible to offer this explanation. Other site samples with negative reference samples did have such animals present.

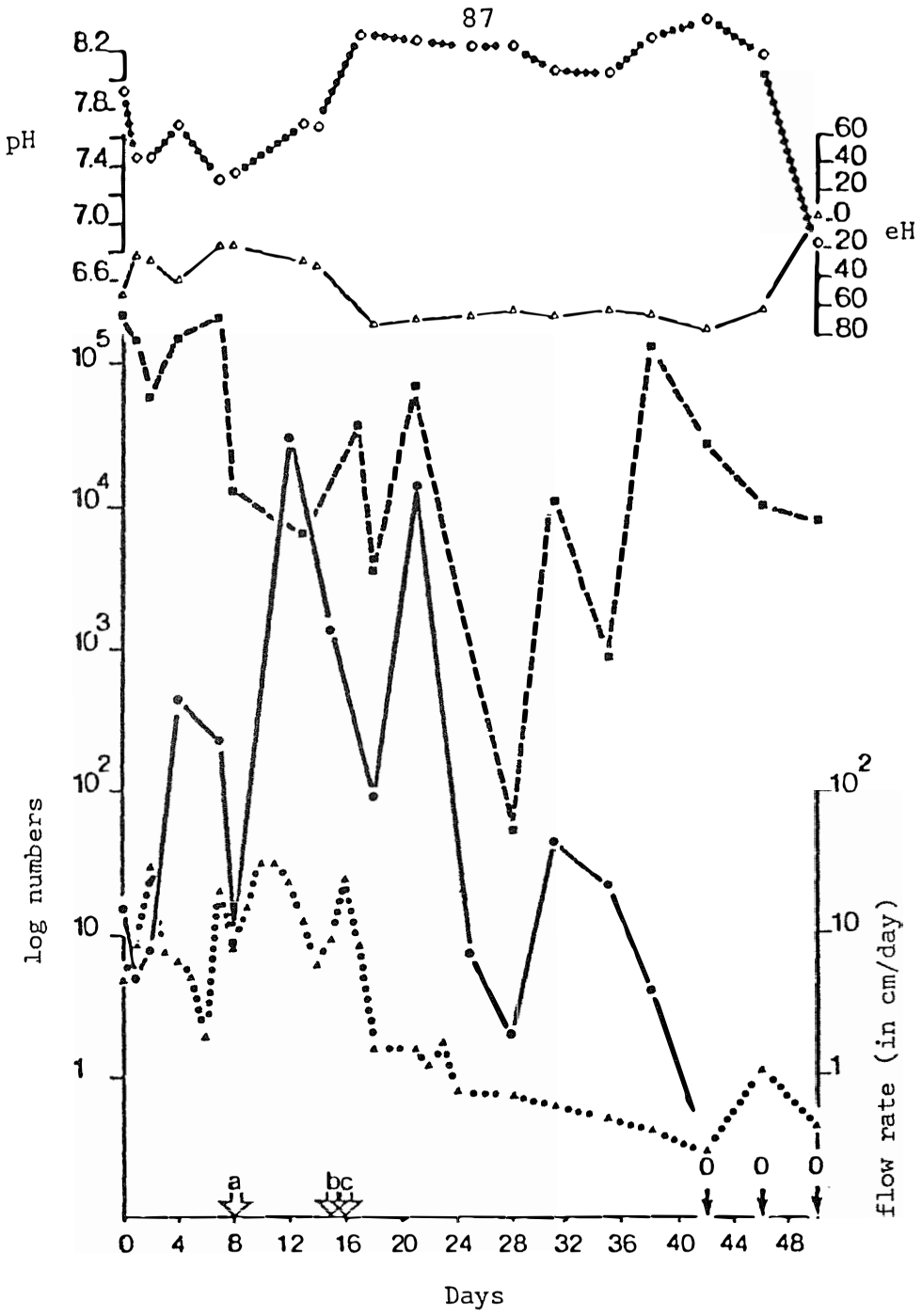
Soil and effluent samples were analysed for three sites for Cl. perfringens. Two of these, 16 EBE and 97 EMP,were located in Spearwood sand and the third, 30 PAR, in Bassendean. The data suggest that these organisms either undergo a limited reduction with depth (97 EMP) or no reduction at all until 0.50 m (30 PAR). There was an increase in concentration at two sites (30 HAR and 16 EBE) with differing soils, which was not consistent with data for coliforms for the same sites. In terms of effluent values obtained in the earlier study (Table 3), it can be seen that Cl. perfringens concentrations were between 94 and 900 organisms per mL, and that high coliform counts were not necessarily associated with high Cl. perfringens counts.

Parker, Carbon and Grubb (1981) refer to the accumulation of sludge at the soil/effluent interface. There was, not unexpectedly, a wide variation in this accumulation, ranging from (at the point of sampling) nil to 0.15-0.20 m of accumulation and in one case (4 JEN) extensive penetration into the soil. There was no correlation between the sludge accumulation and organism removal. The greatest removal in terms of FC was at sites 22 HAR, 35 GEO and 97 EMP, and least at site 30 PAR (comparing 0.15 m counts with effluent values). At site 35 GEO there was no sludge accumulation and at 30 PAR and 97 EMP there was sufficient accumulation to prevent adequate effluent infiltration. By comparison, sites 4 JEN, 48 KEX, 16 EBE & 450 LEN had excessive sludge accumulation and in addition to 35 GEO, site 478 LEN had no sludge accumulation. This aspect of the survey was least controllable. Adequate accurate information about each site was

difficult to obtain as few records of septic tank maintenance were kept by the householder. In some cases, no history was known because properties had either been recently purchased or were rented.

3.4 Preliminary monitoring of four large columns over 50 days.

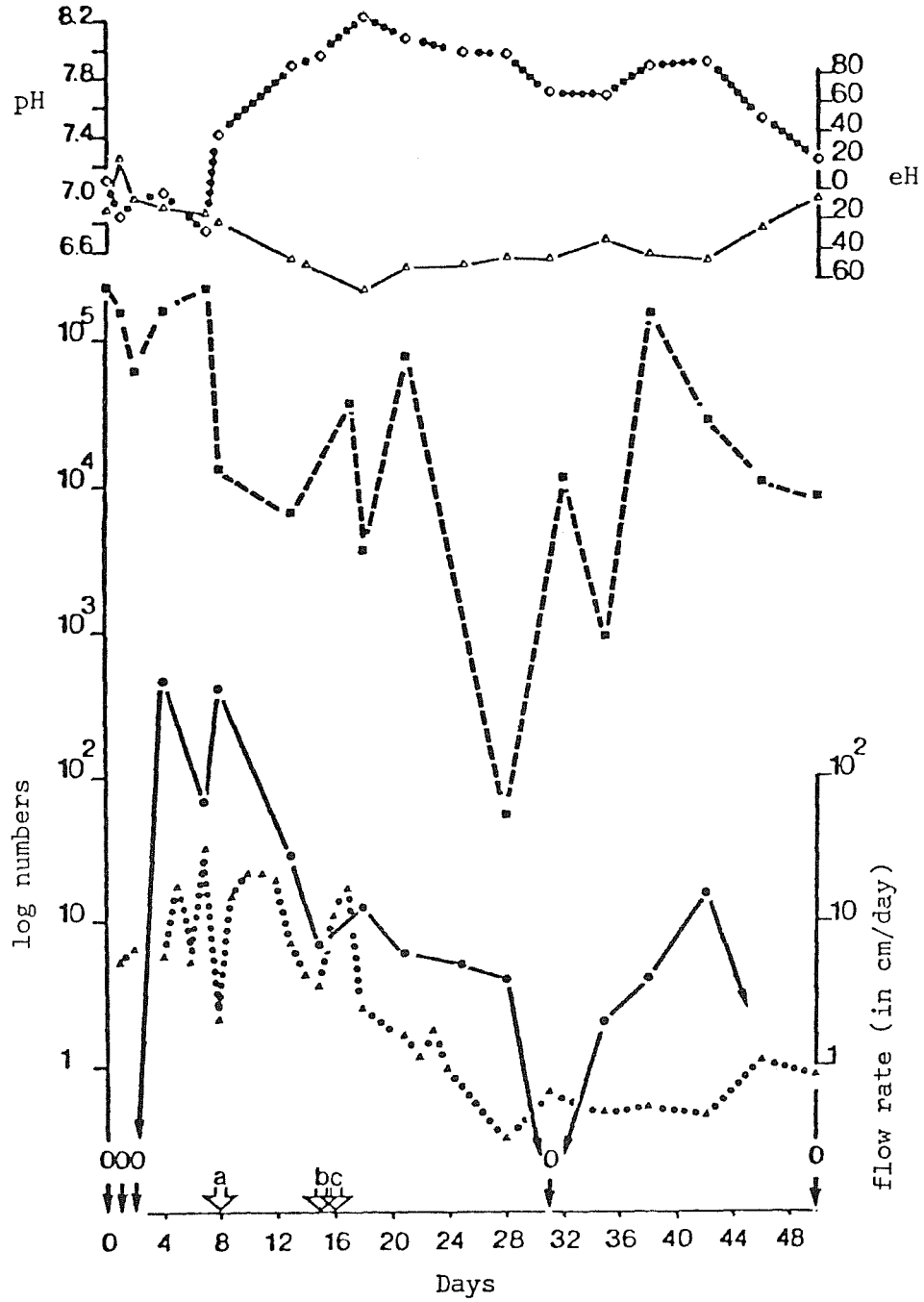
The purpose of this preliminary study was to ensure that a column system was an adequate laboratory model of the conditions found in the field. Adjustments were made to flow rate and aeration accordingly. Curves showing the variation of input FC, leachate FC, flow rate, pH and eH for each of the four columns for the initial fifty days are shown in Figs. 11a, b, c and d. The trends in leachate FC, pH and eH suggest that as flow rate was reduced and aeration increased, the columns began to function in a manner more closely corresponding to the field situation. At the commencement of the experimental programme, flow rates were too high (saturated flow) in all columns, permitting greater breakthrough of FC. Following the introduction of additional sludge onto the soil surfaces giving unsaturated flow (day 14), there was a tendency for leachate counts to decrease. Later studies have shown that sludge thickness alone would not account for this decline. At about the same time (day 15), a series of 0.9 mm holes were drilled in the column walls. From columns Ba 1 and Sp 1 the flow rate at that point was lower than for Ba 2 and Sp 2. The leachate, pH and eH for Ba 1 and Sp 1 changed slowly from day 15, showing an anaerobic trend until approximately day 42, when the trend reversed to a decrease in pH and an increase in eH indicating a nitrification process, as would be expected. By comparison, there was a sharper change in columns Ba 2 and Sp 2, but at the same time there was also a sharp reduction in flow rate from approximately 100 cm/day (saturated flow) to less than 1 cm/day (unsaturated flow). The tendency for these columns to operate apparently anaerobically, changing to an aerobic operation by day 50 was also apparent, but was more marked for Sp 2.



Legend

▪---▪	input effluent FC	◊·····◊	pH
•—•	Leachate FC	a	refrigeration failure
▲····▲	Flow rate (cm/day)	b	aeration holes
△—△	eH	c	sludge addition and tensiometer

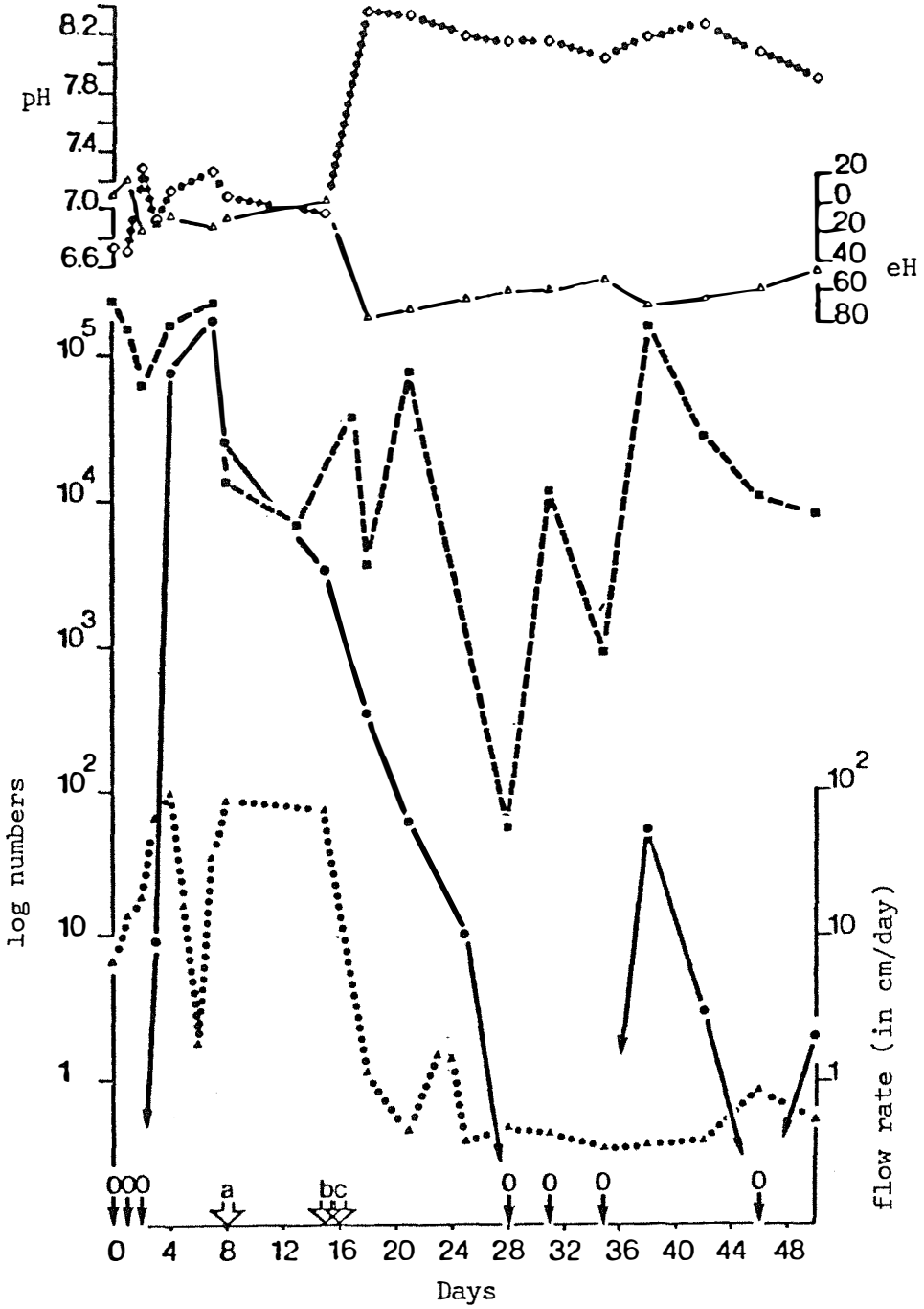
Figure 11a FC movement through column Ba 1, days 1-50.



Legend

▪---▪	input effluent FC	◊.....◊	pH
•——•	Leachate FC	a	refrigeration failure
▲.....▲	Flow rate (cm/day)	b	aeration holes
△——△	eH	c	sludge addition and tensiometer

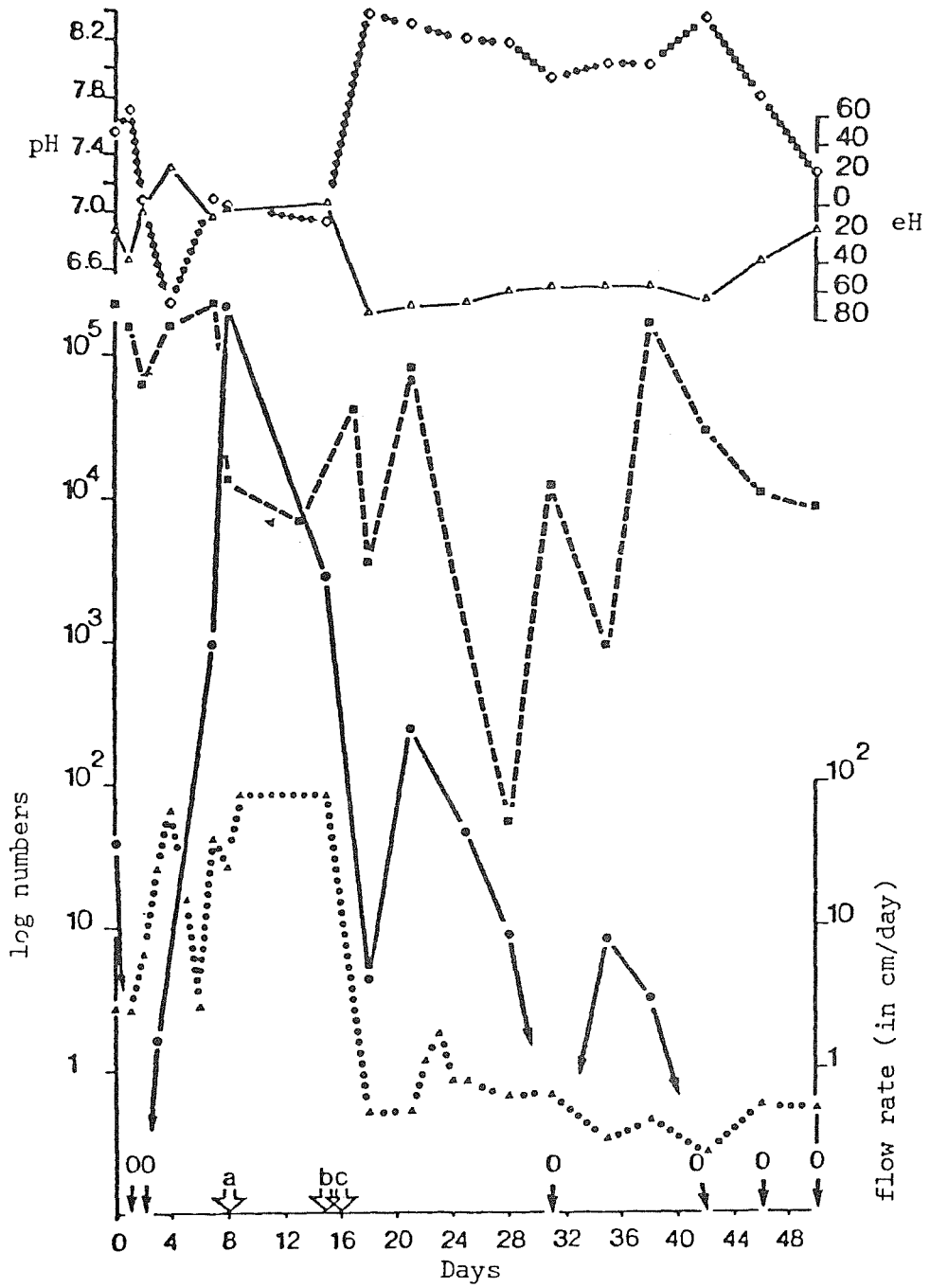
Figure 11b FC movement through column Sp 1, days 1-50.



Legend

▪---▪	input effluent FC	○·····○	pH
•——•	Leachate FC	a	refrigeration failure
▲····▲	Flow rate (cm/day)	b	aeration holes
△——△	eH	c	sludge addition and tensiometer

Figure 11c FC movement through column Ba 2, days 1-50.



Legend

▪---▪	input effluent FC	◇·····◇	pH
•—•	Leachate FC	a	refrigeration failure
▲····▲	Flow rate (cm/day)	b	aeration holes
△—△	eH	c	sludge addition and tensiometer

Figure 11d FC movement through column Sp 2, days 1-50.

A brief refrigeration failure of some twenty hours when the temperature increased from the desired 15-16°C to about 28°C, had the effect of causing die-off of FC organisms in the reservoir and leachate collected: it may not have affected bacterial populations moving in the soil.

In general, there were no marked differences found between soils during this preliminary phase. Organisms showed similar initial breakthrough characteristics for all columns except Ba 1 and Sp 2 where breakthrough was more rapid. Otherwise, FC did not appear in the leachate for 2-3 days. Towards the end of the period, all columns showed a similar trend with respect to FC removal. Between approximately days 30 and 50, although there was some FC breakthrough, the highest being 54/100 mL for column Ba 2, the trend was for counts to reduce to zero. This efficiency of removal continued and during the period between this experiment and the commencement of a dosing experiment with marked bacteria (day 188 onwards), no FC were isolated in the leachates of any column. The columns were maintained in the same condition for the duration of all experimental work.

The experimental work reported in Parker and Carbon (1981) was performed with columns Ba 2 and Sp 2 because columns Ba 1 and Sp 1 did not function satisfactorily in terms of flow rate. These latter columns had flow rates too low to be considered as being representative of field conditions. Repeated attempts to increase flow rates were unsuccessful. However, the flow rates of Ba 1 and Sp 1 did afford the opportunity to compare the effect of flow rate on bacterial and viral removal. The nitrification presumed to be occurring in large columns was confirmed just prior to the commencement of experiments with large columns. The input effluent and leachates from Ba 2, Sp 2, as well as Ba 3 and Sp 3 , were analysed for ammonium-nitrogen. This ion was not present in Ba 1, 2, Sp 1, 2 leachates, but was present in Ba 3 and Sp 3 leachates at concentrations of 70.2 and 64.4 mg/L,respectively. The effluent concentration was 103.3 mg/L. This indirect evidence confirmed that the columns were functioning as adequate models of the field situation, in terms of the

expected oxidation of ammonium ion.

3.5 Monitoring of six large columns: bacteriology and virology.

3.5.1 1.8m columns - bacteriology

Four columns, Ba 1, Ba 2, Sp 1 and Sp 2, were monitored regularly during three periods between day 188 and day 295. As noted above, it was considered that only two columns, Ba 2 and Sp 2, had proper hydraulic function representative of field systems, and some aspects of this study have been presented in Parker & Carbon (1981). Bacteriological, chemical and physical data for three monitoring periods between days 188 and 295 are shown in Figs. 12a and 12b. Following the initial 50-day monitoring period, it was found that no FC breakthrough occurred for the four large columns despite the fact that the input effluent FC concentration from site 97 EMP was between 1.66×10^4 and 3.43×10^7 cells/100 mL. Somewhat higher concentrations of marked Salmonella spp. and E. coli were subsequently introduced and breakthrough of these organisms occurred. It can be seen that greater breakthrough occurred with Bassendean sand (column Ba 2 , Fig. 12a) and although leachate concentrations of test organisms fell below the detection limits on some occasions, there were frequently high concentrations present. When the relative concentrations of Salmonella spp. and E. coli are compared between input and leachate, it can be seen that the Salmonella spp. exceed the E. coli. The effect was particularly apparent from day 211 onwards when leachate concentrations of Salmonella adelaide occasionally exceeded E. coli. For example, estimating 1½ to 2 days for column residence time, the leachate concentrations of S. adelaide between day 218 and 231 exceeded E. coli by 600 to 1000-fold. The same effect was seen after day 239. Apart from one isolated occasion of a high concentration of S. adelaide in the leachate of column Ba 2 on day 219, there was also some suggestion of a downward trend in leachate concentrations of both test organisms from day 188 to 258. This was accompanied by a slight increase in flow rate, but chemically, the leachate remained fairly constant. Evidence

Legend for Figures 12a, 12b, 13a and 13b

•—• *S. adelaide* or *S. typhimurium* (*S. typhimurium* used days 284-295)

o---o *E. coli*

▲ input of culture

▽ organisms not detected

Input effluent values shown in central region of graph, leachate values at the bottom. Other symbols as Figure 11.

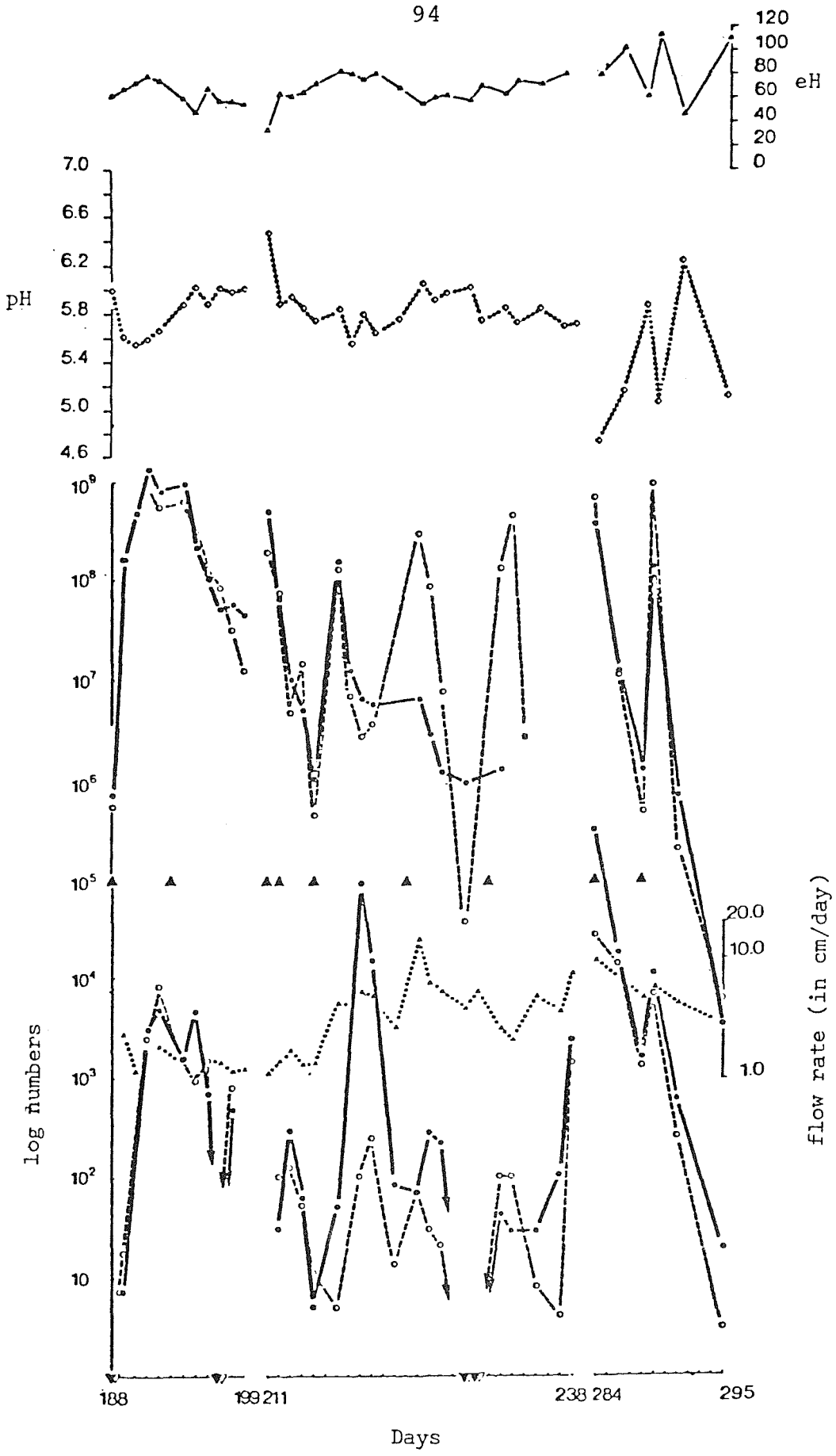


Figure 12a Movement of S. adelaide, S. typhimurium and E. coli through column Ba 2, days 188-295.

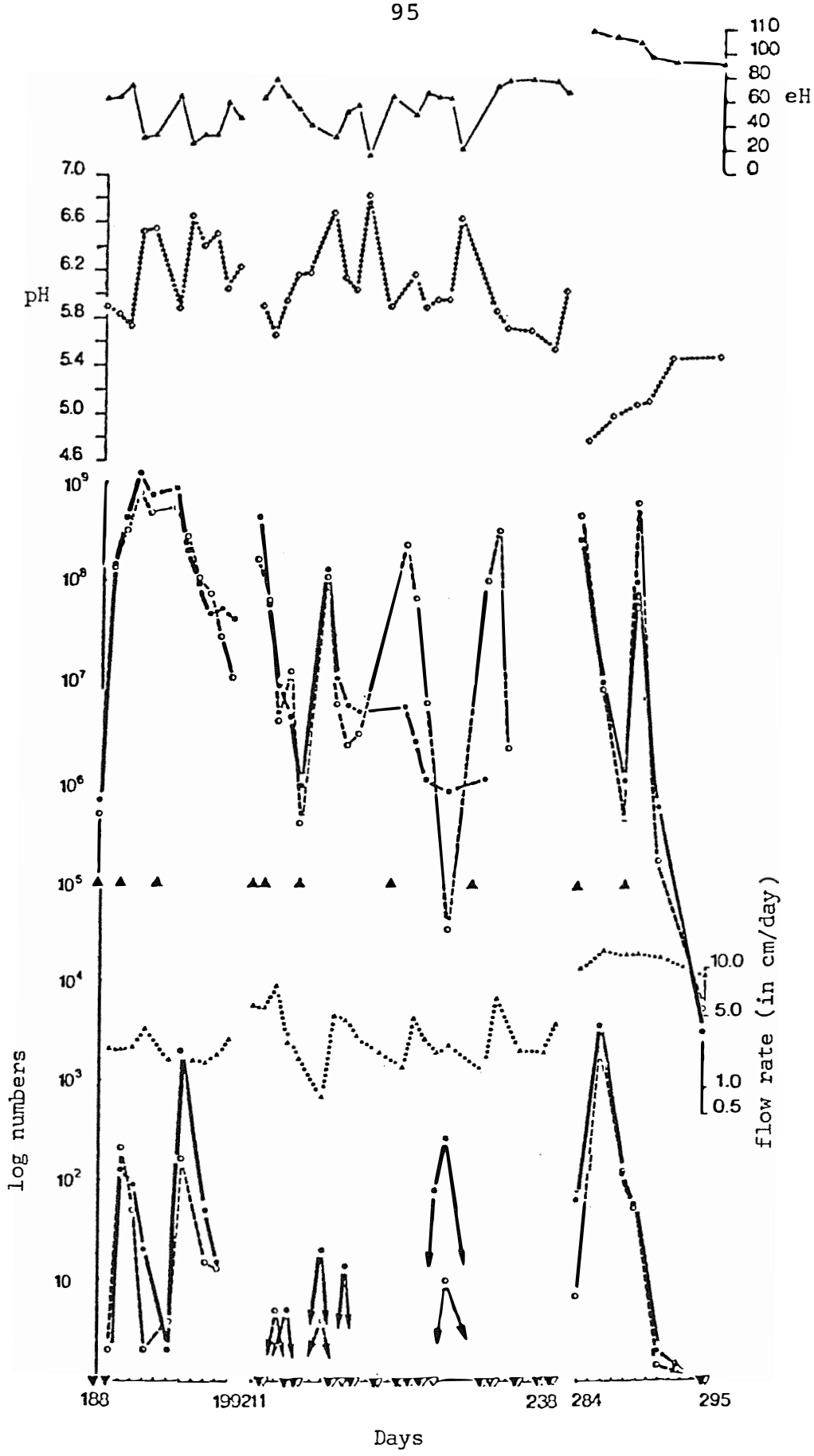


Figure 12b Movement of S. adelaide, S. typhimurium and E. coli through column Sp 2, days 188-295.

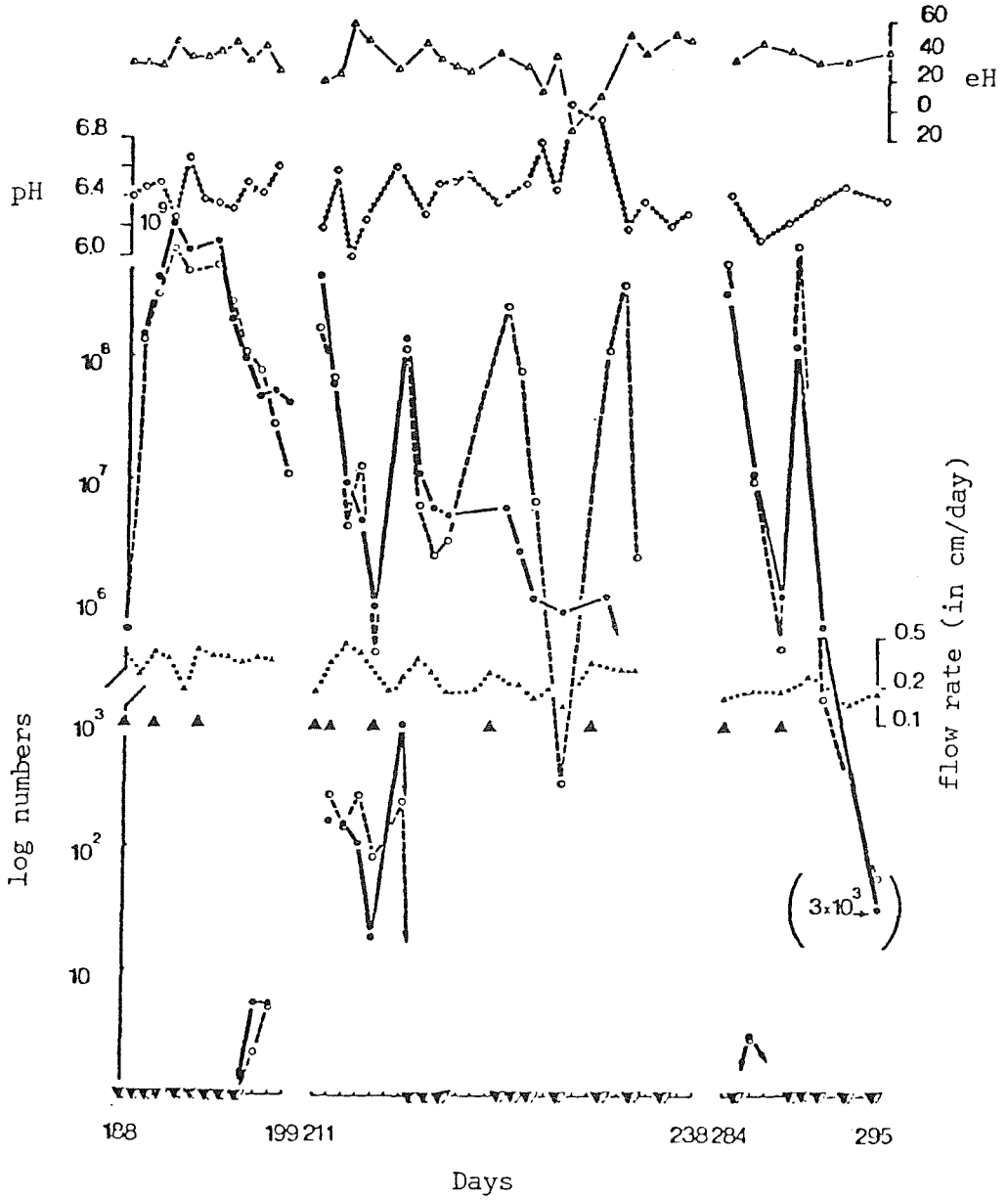


Figure 13a Movement of S. adelaide, S. typhimurium and E. coli through column Ba 1, days 188-295.

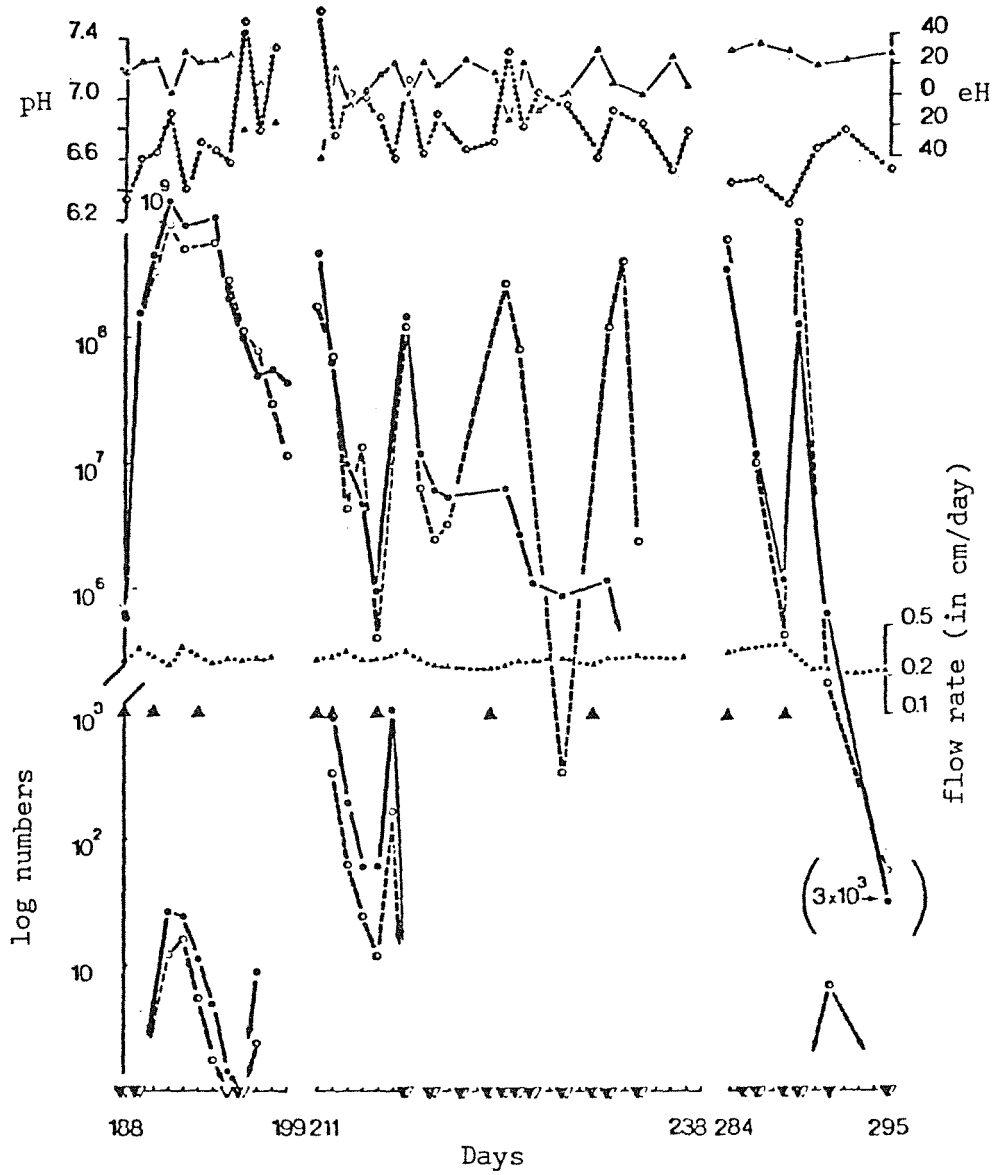


Figure 13b Movement of S. adelaide, S. typhimurium and E. coli through column Sp 1, days 188-295.

presented elsewhere suggests that this observed decline may have been associated with antagonism or predation. A gap of forty six days before the third monitoring period was perhaps sufficient for the possible stimulating effect to be removed. Following reintroduction of test organisms on day 284, the leachate concentrations declined much more rapidly than had previously occurred.

Fig. 12b shows the same analyses for Spearwood sand column Sp 2. The most obvious difference is that leachate counts frequently declined to zero, especially during the middle phase following day 211. Similar observations can also be made concerning comparative removal of test organisms. Following day 225 when E. coli exceeded S. adelaide in the effluent input, the leachate concentrations were reversed (day 229), and as with column Ba 2, the behaviour of S. typhimurium was similar to E. coli and the wide differences previously noted were not seen. Somewhat greater fluctuations in chemical data were apparent with column Sp 2, and occasions when pH was raised coincide with isolated breakthrough of S. adelaide.

If the data for both columns are compared overall (Parker and Carbon 1981), it can be seen that Salmonella spp. were more effectively removed than E. coli. However, mean counts did not reflect the daily variation actually observed.

Flow rate for columns Ba 2 and Sp 2 varied within the same general range but did not appear to affect other parameters. There was an overall tendency for flow rates to increase for both columns, although this was less marked for column Ba 2. The differences between columns (soils) were not great enough to give rise to the differences in bacteriological behaviour.

Although columns Ba 1 and Sp 1 were not considered to be representative of normal field conditions, they were monitored with the other columns. The flow rates for columns Ba 1 and Sp 1 were 12.8 times and 14.4 times less than Ba 2 and Sp 2 respectively, calculated on mean flow rates. This substantially

slower flow in both cases was associated with significantly different bacteriological behaviour. Figs. 13a and 13b show analyses for columns Ba 1 and Sp 1 respectively for the three monitoring periods between day 188 and 295. There was only a limited degree of breakthrough during the first and second monitoring periods. Although the actual numbers of organisms appearing in the leachate are small, and restrictions imposed by low volumes of leachate tended to limit the accuracy of counts, it appeared that differences due to soil type were minimized. There was some slight early breakthrough of both test organisms in column Sp 1 about two days following their introduction into the input effluent. This compares with a much lower breakthrough in the same period for Ba 1. In the second monitoring period, test organisms appeared in the leachates of both columns, S. adelaide tending to exceed E. coli in Sp 1. In both columns thereafter, neither test organism appeared in the leachate in the quantities available for analysis, except on one occasion at very low level. The pH and eH data (column Ba 1) were generally as predicted for nitrification. In the case of Sp 1, the values reverted occasionally to a more anaerobic mode but this was not associated with any particular change in leachate bacterial concentrations.

3.5.2 1.8m columns - virology

Shortly following the termination of bacteriological studies, the four large columns were dosed with large concentrations of MS2 phage. Results for columns Ba 1, Sp 1, Ba 2 and Sp 2 are presented in Figs. 14a, b, c and d respectively. Considering chemical and physical data, it can be seen that columns continued to function in the same manner as during the earlier experiments. Perhaps the most surprising result is that virus appeared in the leachates on days 306 and 308 for all four columns. For the slower flowing columns, Ba 1 and Sp 1, the concentrations were greater. Subsequent to day 308, no further virus breakthrough occurred despite the large concentration being introduced in the effluent (exceeding 10^{10}PFU/mL on day 317). Prior to the addition of virus, the effluent was tested (up to 0.5 mL) on lawns of the host strain and no plaques were observed.

Legend for Figures 14a to 14d

●——● Effluent virus titre

c---o Leachate virus titre

▲···· ▲ Flow rate (cm/day)

△—— △ eH

◊····· ◊ pH

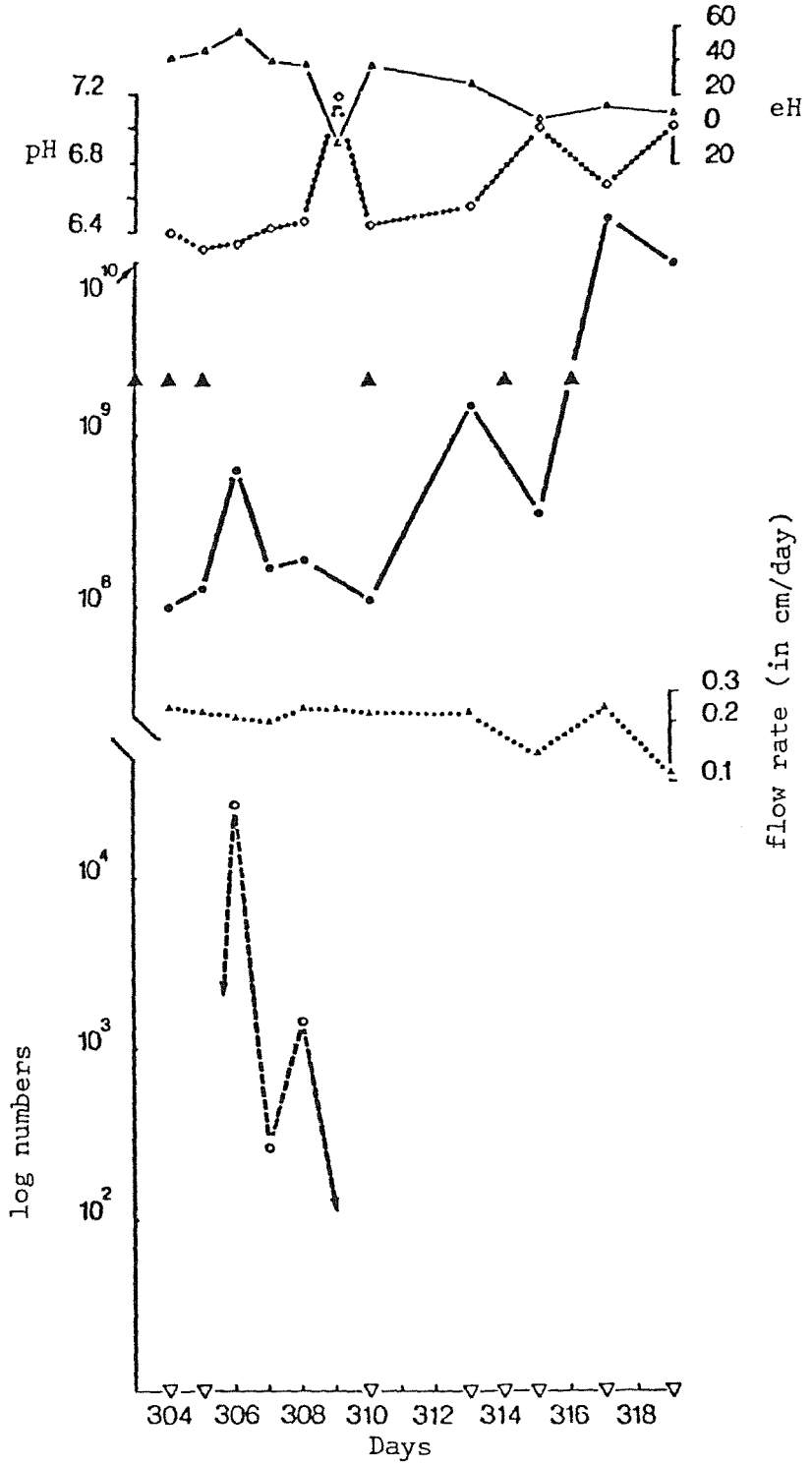


Figure 14a Movement of MS2 phage through column Ba 1, days 303-319.

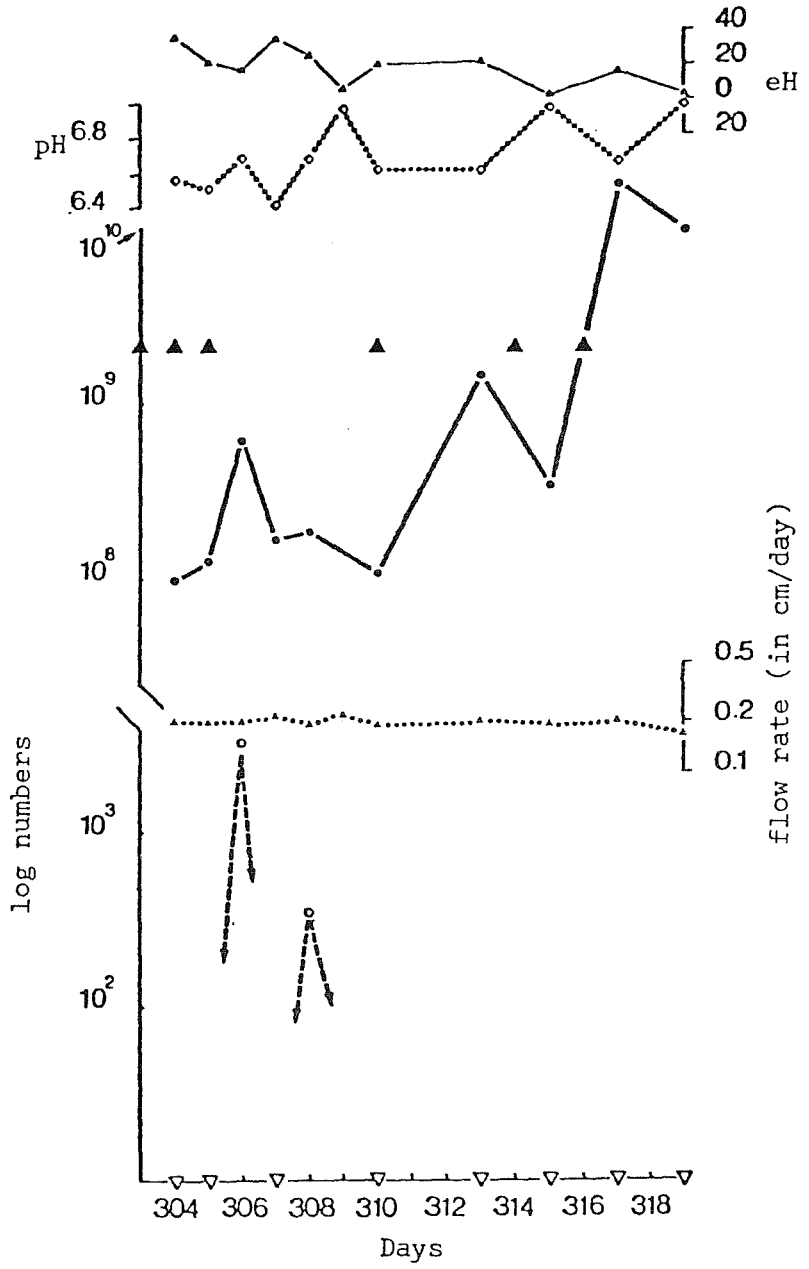


Figure 14b Movement of MS2 phage through column Sp 1, days 303-319.

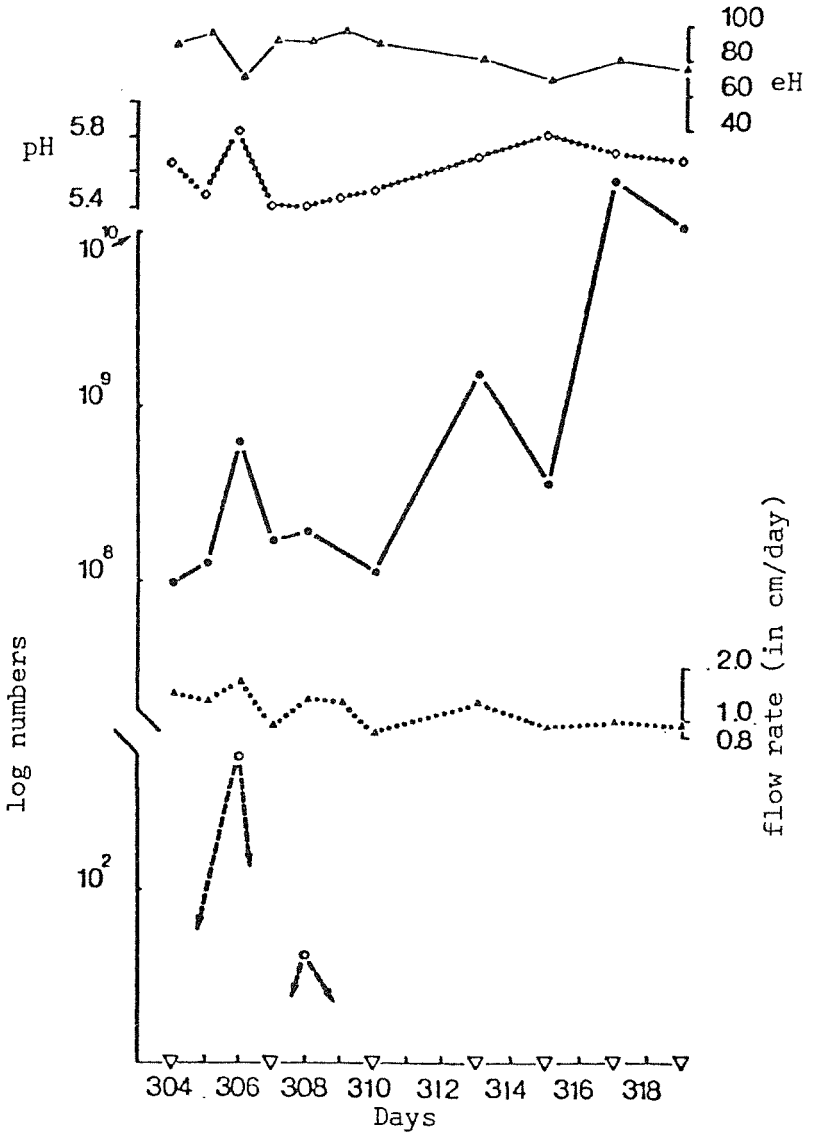


Figure 14c Movement of MS2 phage through column Ba 2, days 303-319.

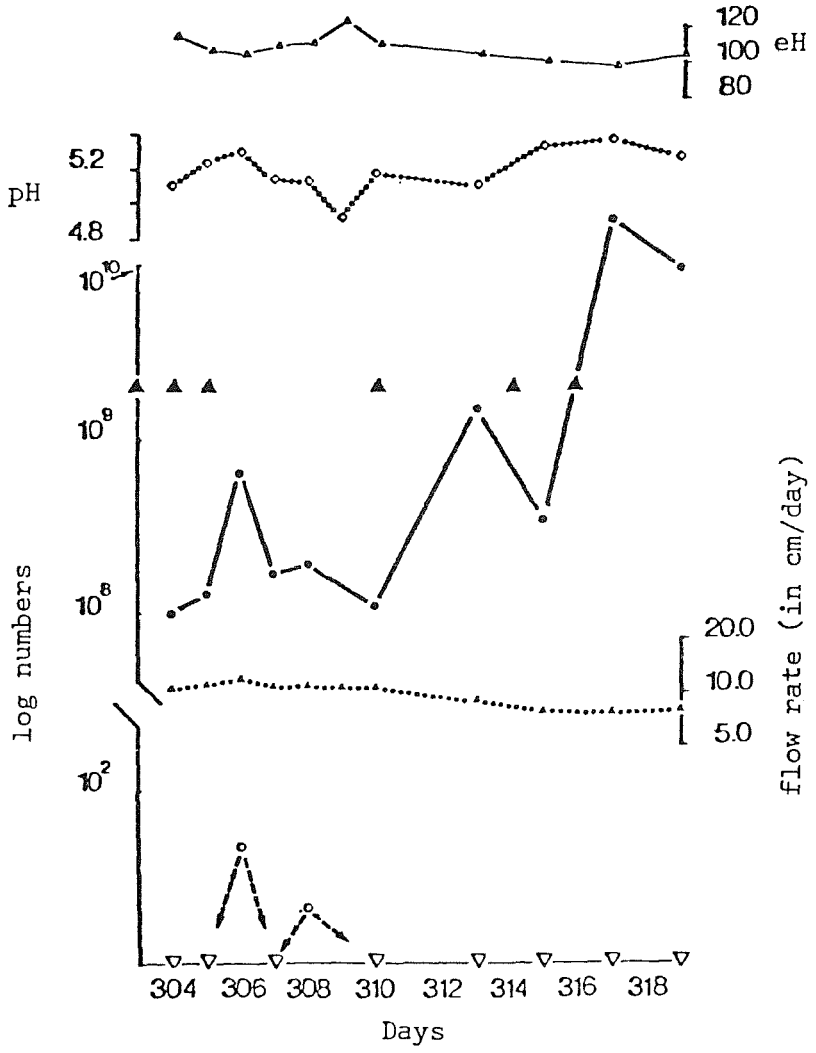


Figure 14d Movement of MS2 phage through column Sp 2, days 303-319.

3.5.3 0.3 m columns - bacteriology

Bacteriological, chemical and physical data for the same three phase monitoring period for columns Ba 3 and Sp 3 are presented in Figs. 15a and 15b respectively. For reasons of clarity, input effluent bacteriological data are shown in red. It can be seen from chemical data that the columns were anaerobic, pH values for both leachates being alkaline and eH values correspondingly negative. Values for ammonium nitrogen for Ba 3 and Sp 3 leachate were respectively 70.2 and 64.4 mg/L compared with 103.3 mg/L for fresh effluent.

For column Ba 3 there was little reduction in either *S. adelaide* or *E. coli* during the first monitoring period (day 188 to day 199) except on day 194 when *E. coli* was reduced to a somewhat greater extent than *S. adelaide*. However, during the second phase following day 211, the same 'selective' phenomenon as occurred in 1.8 m columns was observed. When input effluent concentrations of *E. coli* were greater, the reverse was true for the leachate and *S. adelaide* tended to exceed *E. coli* (days 220, 225 and 231). Unlike the 1.8 m columns, the same tendency was also apparent for *S. typhimurium* (day 284 onwards). Whether the same phenomenon of antagonism and/or predation was occurring is more difficult to interpret since leachate curves tended to follow more closely the patterns for input effluent. However, there was some evidence of a general downward decline during the middle phase. Differences due to or associated with soil type seen by comparing Bassendean sand with Spearwood sand in 1.8 m columns were not apparent for 0.3 m columns. The results for column Sp 3 (Fig. 15b) show very similar trends to Ba 3. Exceptional are the results for the initial phase where there was a marked reduction during approximately the first nine days. Relatively, the leachate concentrations at this point increase to a level similar to Ba 3. In the second monitoring period, the same 'selective' phenomenon was seen again and similarly in the final phase. As with Ba 3, there appeared to be the same overall decline in leachate concentrations in the

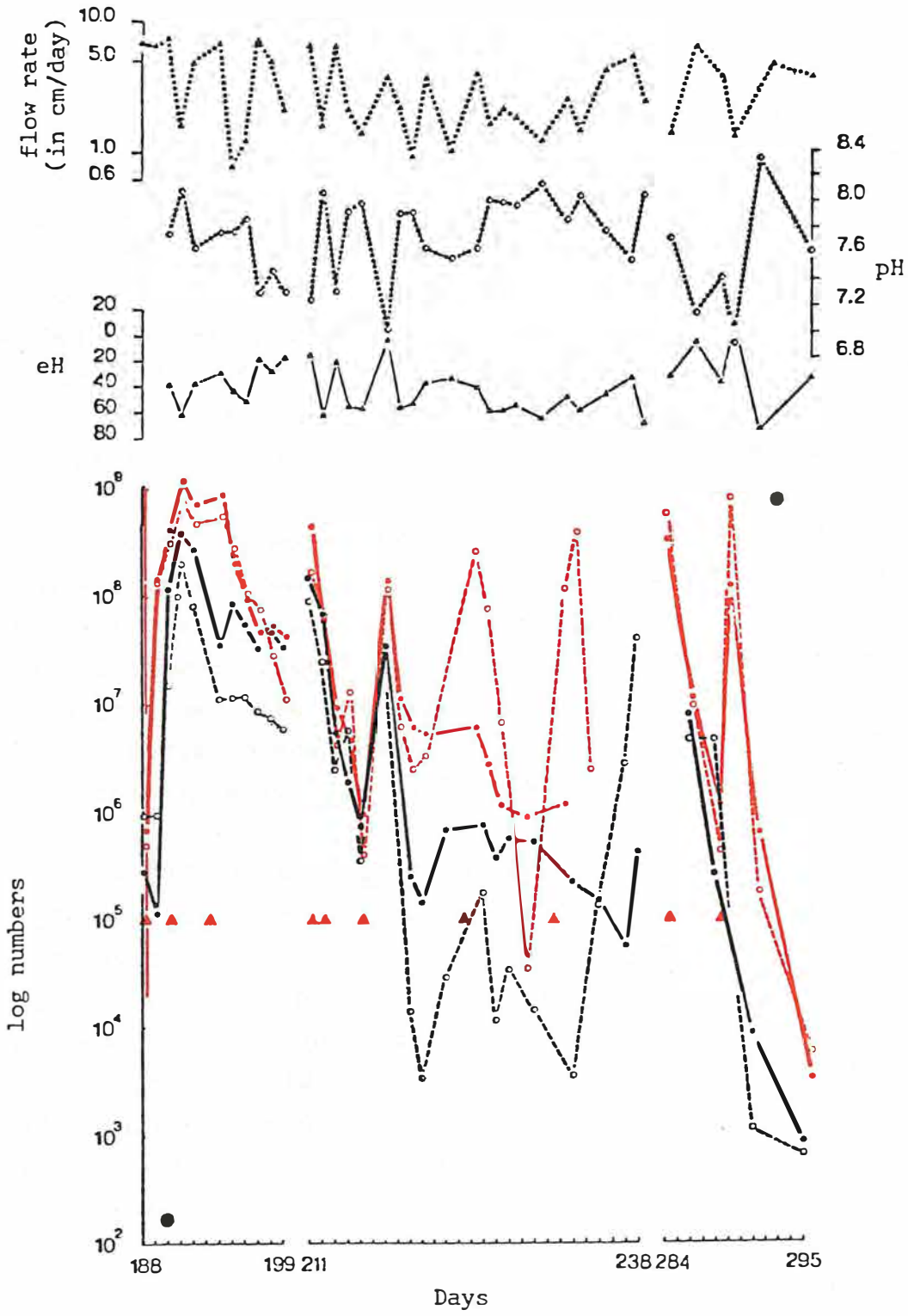


Figure 15a Movement of S. adelaide, S. typhimurium and E. coli through column Ba 3, days 188-295. S. typhimurium used between days 284-295.

All symbols as Figure 12.

Input effluent data shown in red.

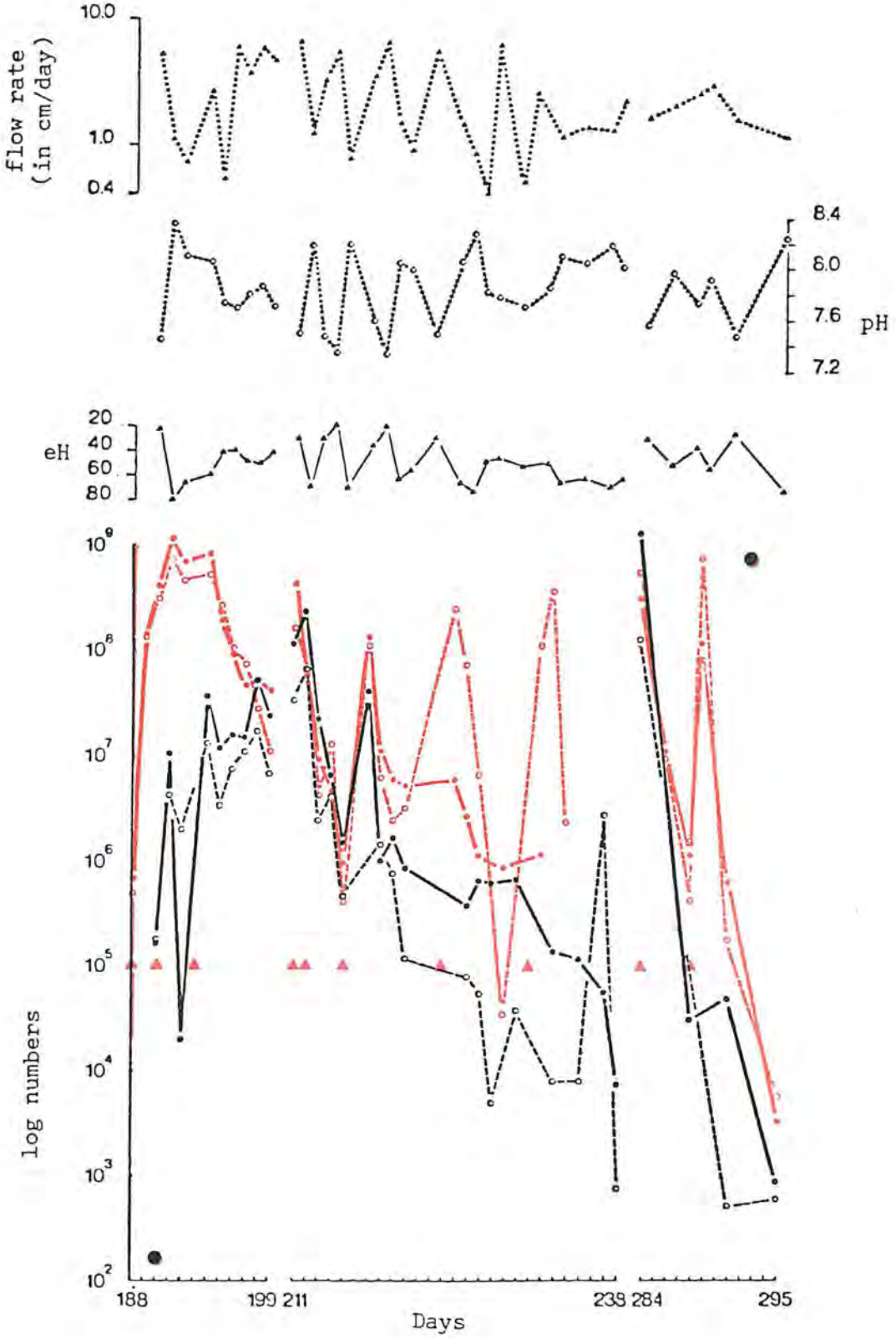


Figure 15b Movement of S. adelaide, S. typhimurium and E. coli through column Sp 3, days 188-295. S. typhimurium used between days 284-295.

All symbols as Figure 12.

Input effluent data shown in red.

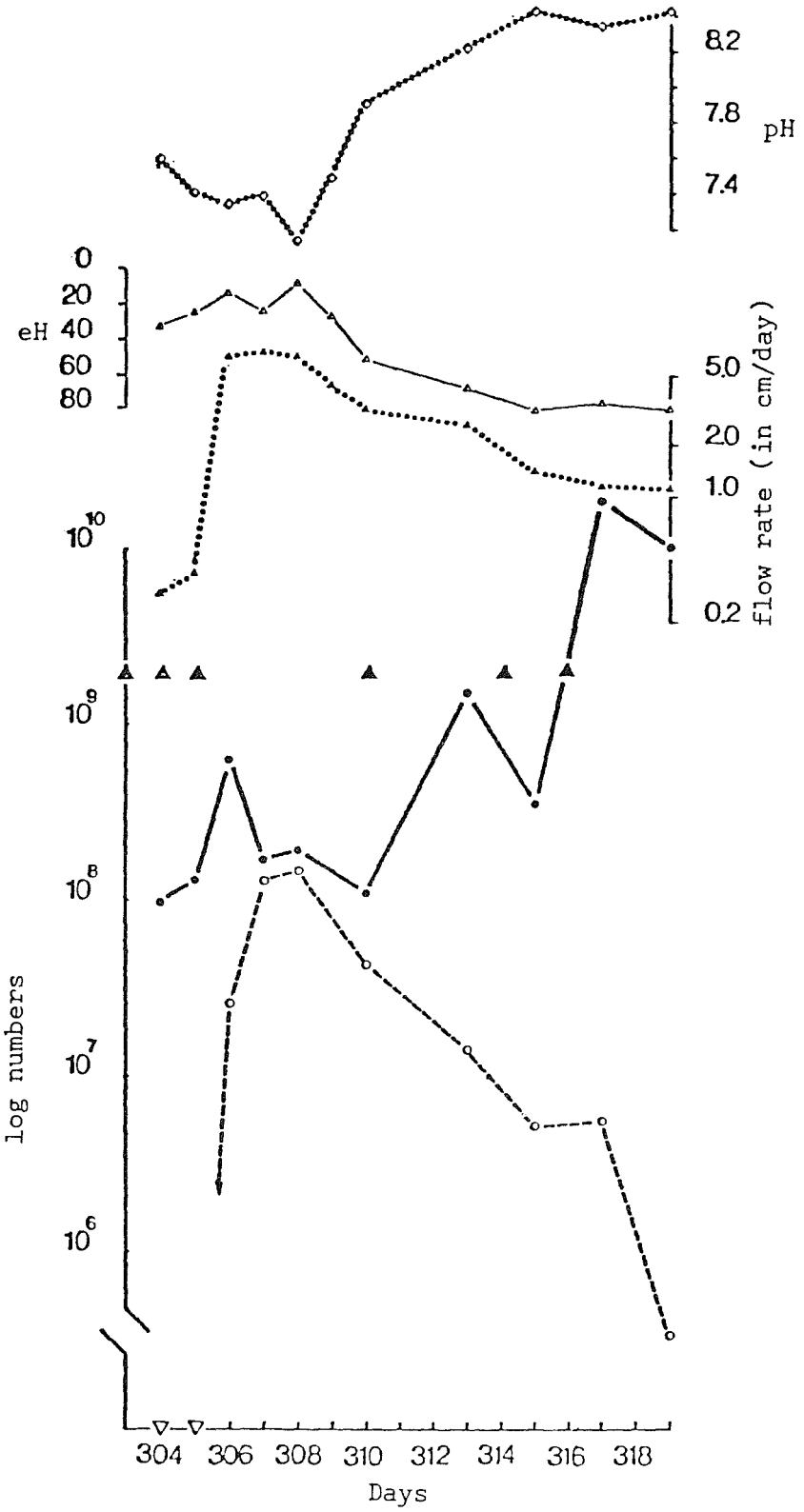


Figure 16a Movement of MS2 phage through column Ba 3, days 303-319.

Symbols as Figure 12.

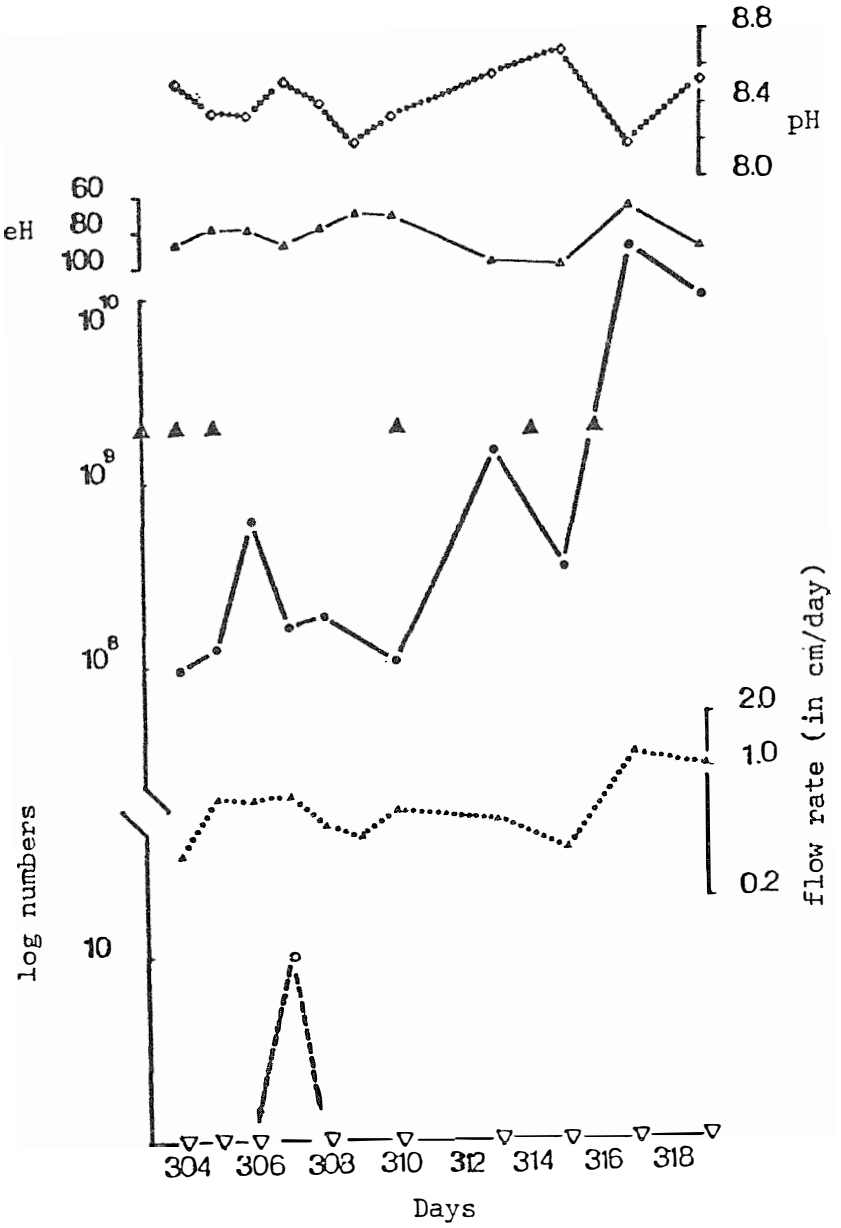


Figure 16b Movement of MS2 phage through column Sp 3, days 303-319.

Symbols as Figure 12.

second period. As observed in Parker & Carbon(1981), 0.3m columns were not efficient at removing these test organisms and it is possible that the data obtained may slightly underestimate the actual numbers because of potential predatory and antagonistic activity of organisms present in the outflow tubing.

3.5.4 0.3m columns - virology

The expected similarity between soils with respect to virus removal was not observed: the soils behaved very differently. Fig. 16a and 16b show virological, chemical and physical data for columns Ba 3 and Sp 3 respectively. Column Ba 3 leachate contained on day 307 and day 308 approximately the same virus concentration as the input effluent (1-2 x 10^8 PFU/mL). This degree of breakthrough also coincided with a sharp increase in flow rate: 0.4 cm/day to 6 cm/day. Such an increase may have accounted for the increase in virus breakthrough. However, even after flow rate reduced to about 1 cm/day, the concentration of virus in the leachate of this column was high at about 5 x 10^6 PFU/mL, effectively about a 3½ log reduction on day 317. By comparison, Fig.16b shows results for column Sp 3. The reduction during the same period was almost complete, breakthrough occurring only on one occasion at very low level. Flow rates for column Sp 3 were comparable to Ba 3 from day 313 onwards.

3.6 Direct "destructive" sampling of two large columns

Figures 17a and 17b show the concentrations of "total" bacteria (including pigmented forms), TC, FC, Actinomycetes, for columns Ba 2 and Sp 2, and for column Ba 2, protozoa and other soil organisms such as rotifers and nematodes are also shown. In general, the patterns obtained were predicted from field studies. Considering FC, it can be seen that there was, in both columns, an accumulation of these organisms in the sludge layer, resulting in a count (in both cases) slightly higher than for the ponded effluent. It is difficult to extend this comparison further because effluent counts were

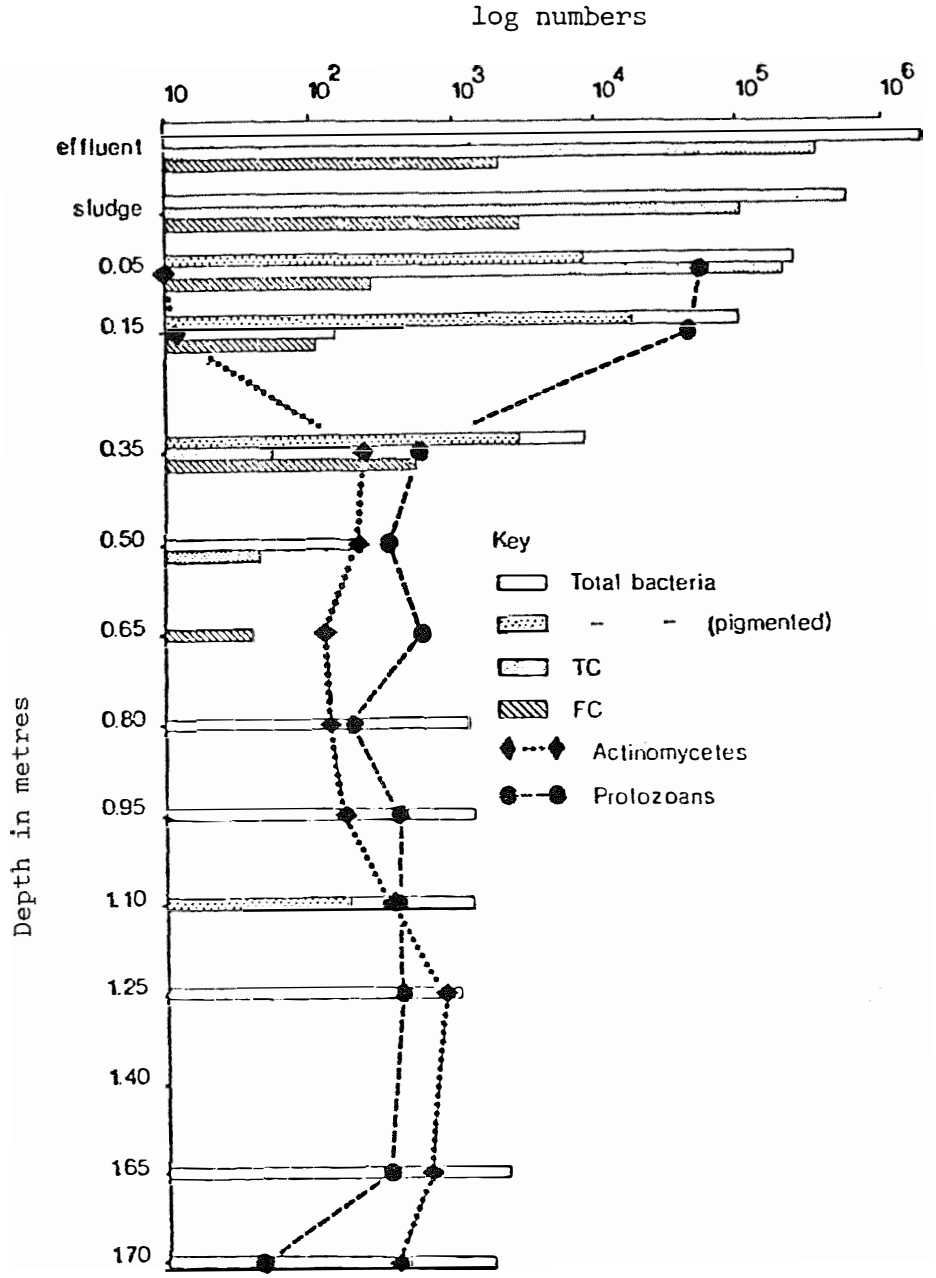


Figure 17a Numbers of "total" bacteria (including pigmented forms), TC, FC, Actinomycetes and Protozoans in column Ba 2.

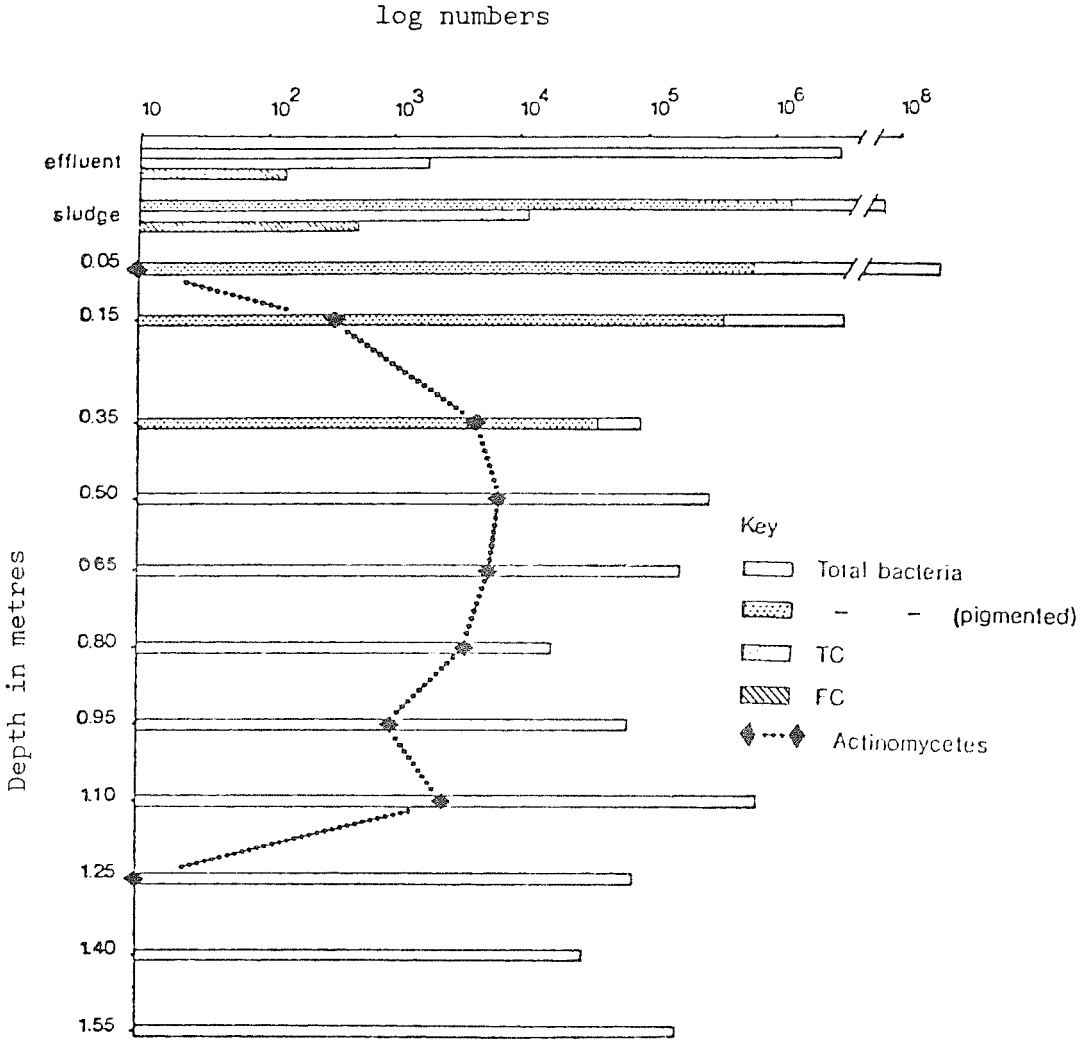


Figure 17b Numbers of "total" bacteria (including pigmented forms), TC, FC and Actinomycetes in column Sp 2.

made on liquids, and sludge counts on a semi-liquid slurry. The destruction of these columns was made one week apart, and the FC concentrations were different (Ba 2 > Sp 2). The observation that FC appeared to penetrate further (isolation at 0.65 m) in Ba 2 cannot therefore be explained solely on the basis of soil type. However, the decline in FC apparent in column Sp 2 was very rapid, with no organisms detected below the sludge. In terms of input effluent members, and subsequent decline with depth, the same trend was apparent for TC. There was a much greater difference in the input concentrations, however. By definition at least, TC should always exceed FC, but for column Ba 2 at a depth of 0.35 m this was not the case and at 0.65 m FC were isolated and not TC.

The "total" bacterial flora of both columns was assessed for two reasons: firstly, to obtain a general microbial picture, and secondly to assess numbers of pigmented flora, particularly any mucoid organisms that might be found at the sludge/soil region and thought to contribute to the filtering property of the schmutzdecke. "Total" bacterial flora (aerobic organisms on rich nutrient media) is misleading terminology because strictly anaerobic organisms are excluded, for example. Effluent "total" bacteria were similar for both sampling occasions, but the numbers obtained in the sludge and depth samples were different according to soil type. For column Ba 2, significantly lower counts were obtained in the sludge and at all depths tested compared with Sp 2. There was a concentration of pigmented types present in samples from 0.05 m to 0.35 m and again at 1.10 m. These organisms were exclusively fluorescent types. Following examination by Gram's stain and colonies under ultraviolet light, they were considered to be fluorescent pseudomonads. The concentration of "total" bacteria for column Sp 2 was significantly higher at all sample depths than Ba 2 and below 0.35 m there was no decline, although this latter phenomenon was the same for Ba 2. The pigmented types present, concentrated between the sludge and 0.35 m, were non-fluorescent and produced a range of pigments between pale yellow and red. On the basis of Gram's stain and morphology, they could be tentatively

described as Flavobacterium spp., or gliding bacteria, although this was not confirmed.

Counts of actinomycetes were dissimilar. For column Ba 2, the numbers rose to between 100 and 1000 organisms per gram at 0.35 m and remained at this level for the whole column depth. For column Sp 2 the trend was slightly different, in that following an initial rise between 0.35 m and 0.50 m, the numbers declined thereafter and were not isolated below 1.10 m.

Protozoa counts were only obtained reliably for column Ba 2. The attempt to obtain quantitative data for column Sp 2 was unsuccessful because of fungal contamination. The attempt was repeated on column Sp 1, but with the same result. However, despite dense mycelium in the culture wells, it was possible to observe active large protozoa in samples from 0.05 m to 0.35 m. The quantitative data for column Ba 2 showed a very active population immediately below the sludge layer, declining to a constant level at 0.35 m and being maintained down the column. Some attempt was made to identify the predominant species present, but the list is incomplete and only the larger organisms were tentatively identified. The most commonly occurring ciliates were Colpoda cuculus and the smaller Colpoda steini. A much larger and more active organism isolated predominantly from the upper soil samples (> 0.35 m) was probably Kahlia acrobates. A wider range of small flagellates was seen at all depths but not identified. No amoebae were seen at all. A number of rotifera were seen distributed throughout the column and were all Habrotricha spp. Some nematodes were also seen, but since no specific attempt was made to cultivate these, there may have been somewhat higher numbers present than actually observed.

3.7 Survival of Salmonella adelaide, FC, and FS in septic tank effluent & sludge

3.7.1 Effluent

The derivation of bulk samples of effluent for this study were from two diverse sources. It was known that one family was

heavy water users (4 JEN) and the clothes washing machine was used every second day. The detergent content of the effluent was therefore probably high although this was not tested. The second site, 59 WES, had by contrast, separate sewage/sullage systems. Only the effluent from the septic tank receiving the toilet waste was used for this experiment. The family at the second site consisted of 2 adults and 3 children and the septic system had been in use for at least 25 years. Whereas the effluent from site 4 JEN was excessively turbid, grey in colour and foamed when shaken, the effluent from 59 WES was green in colour with some suspended settling matter and did not foam.

Comparative data for S. adelaide, FC and FS in 4 JEN and 59 WES effluents are presented in Fig. 18 (15^{o} C) and Fig.19 (22^{o} C). At both temperatures the survival is in the order FS > FC > S. adelaide. S. adelaide underwent a fairly rapid decline which was retarded for eight days in 59 WES effluent. In both effluents at 15^{o}C the ultimate concentrations of either S. adelaide or FC at day 64 were similar, despite the dissimilarity of effluents. Of importance is the fact that S. adelaide populations, despite their initial numerical superiority, were less numerous at day 64 than were FC. In general, the differences in effluents did not give rise to large differences in survival for either FC or S. adelaide. There is, however, a contrast between effluents for FS at 15^{o}C over the eight days for which data are available, possibly indicating a better survival potential in 59 WES effluent.

The concentrations of S. adelaide at 22^{o}C (Fig. 19) at day 64 were different between effluents, survival being greater in 4 JEN effluent.The trend for FC suggests that these organisms would be in approximately similar concentrations at day 32.

3.7.2 Sludge

Three sites were selected for sludge survival studies: 4 JEN, 28 SOU and 11 DAN. The sludge in all three cases was the finely divided material sedimented on the bottom of the

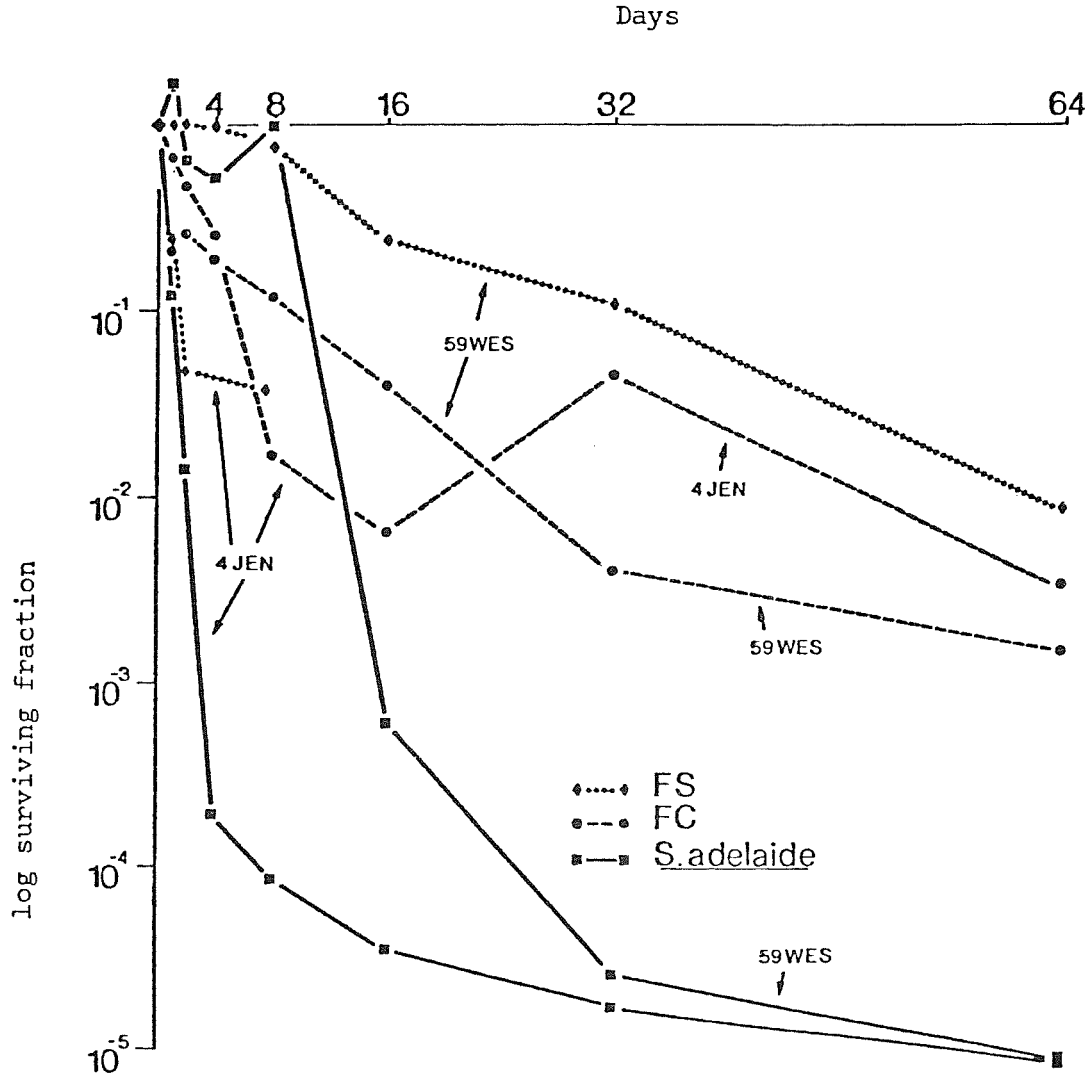


Figure 18 Comparative survival of S. adelaide, FC and FS in 59 WES and 4 JEN effluents at 15°C.

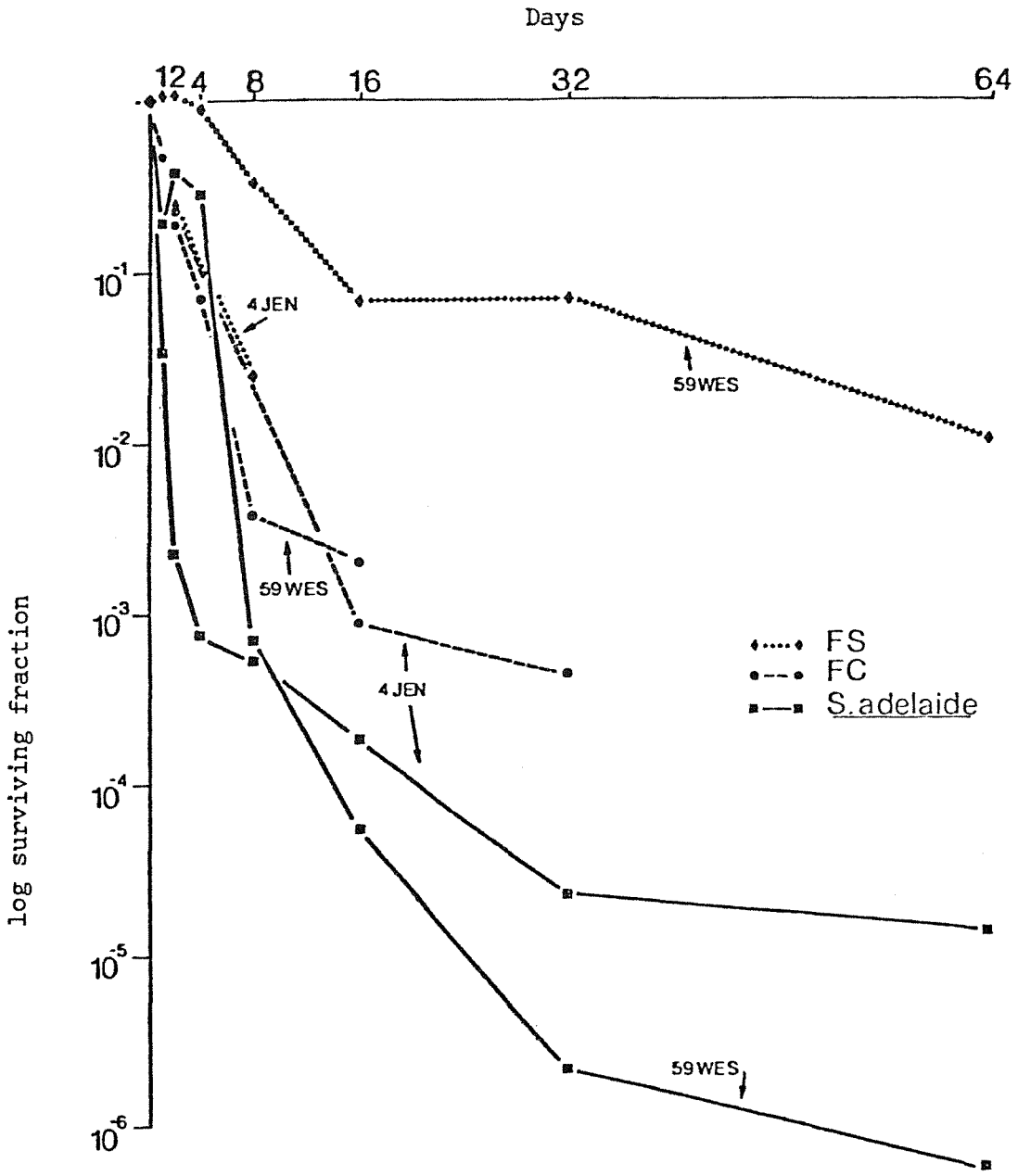


Figure 19 Comparative survival of S. adelaide, FC and FS in 59 WES and 4 JEN effluents at 22°C.

infiltration system. The material at all sites was collected subsequent to overlying effluent having been pumped out. The conditions at site 4 JEN have been discussed above. At site 28 SOU, the system had been in use some 11 years and the sludge was collected from a central segment of a 10 m leach drain. At site 11 DAN, the system was somewhat more recently installed although the date could not be established.

Data for FS were unfortunately unobtainable because of low initial counts at all of the sites. In general, the results are for 15° C only, although some work was done at 22° C with material from site 11 DAN. Survival curves for indigenous FC and added S. adelaide at 15° and 22° in sludge from 11 DAN are presented in Fig. 20. These curves are exceptional in that at 15° C there was effectively little die-off for S. adelaide and a considerable degree of actual growth of FC. At 22° C there was some S. adelaide die-off, but for FC the results were very similar to those for 15° C and these organisms showed growth between day 0 and day 4.

For sites 4 JEN and 28 SOU, the results were similar to those obtained for survival in effluents. Survival curves for these sites (at 15° C only) are shown combined with 11 DAN data (15° C) in Fig. 21.

The survival of S. adelaide was different in all three sludges. For 4 JEN there was rapid initial decline, re-growth, and decline at a lesser rate. For 28 SOU, following slight initial re-growth, the rate of decline was more uniform. In terms of surviving organisms at day 16, the range including data for 11 DAN, was very wide, from effectively no decline, to slightly more than 10-fold decline (4 JEN) to about 300-fold decline (28 SOU). The survival of FC in 4 JEN and 28 SOU sludges was very similar.

The survival of organisms in the accumulated sludge of septic tank systems could be more extended than in effluent because of possible 'protective' effects of the material itself and

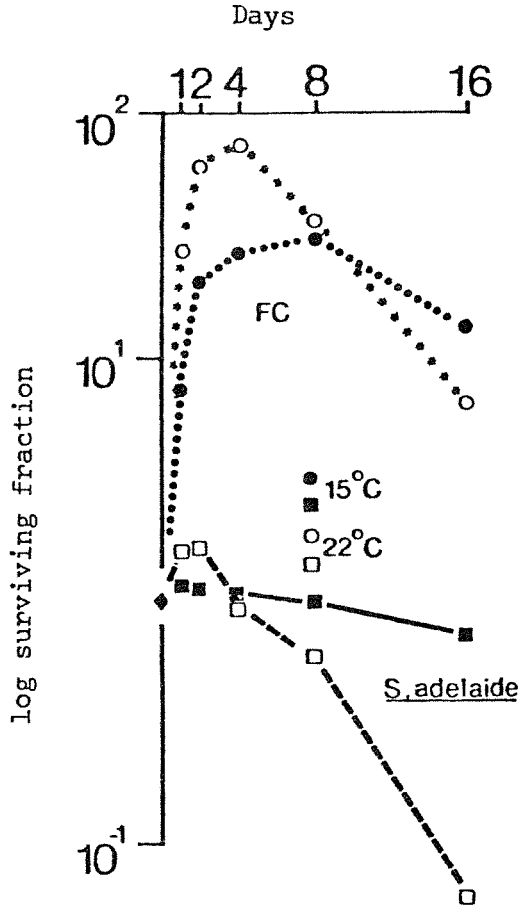


Figure 20 Comparative survival of S. adelaide and FC in 11 DAN sludge at 15°C and 22°C.

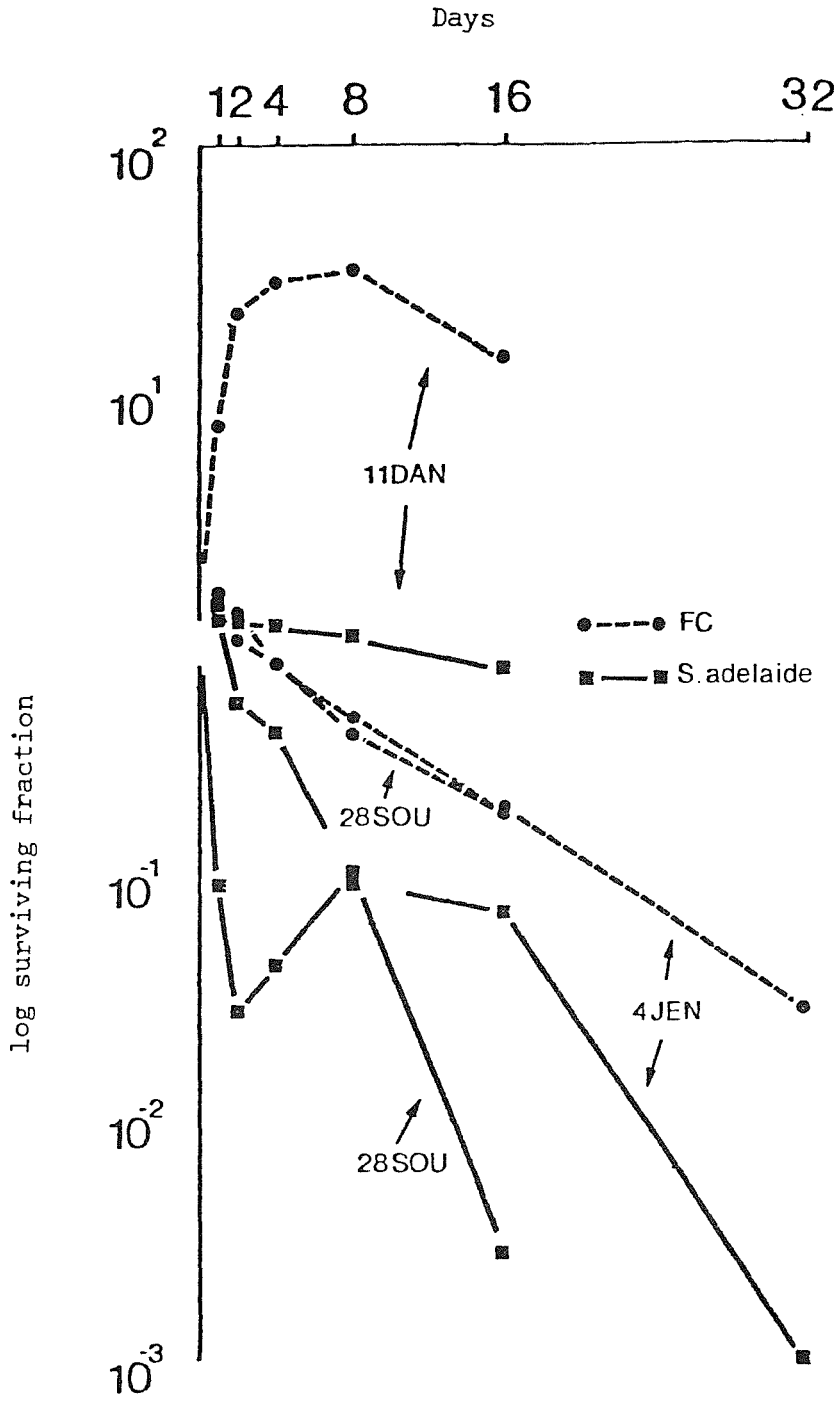


Figure 21 Comparative survival of S. adelaide and FC in sludge from sites 11 DAN, 28 SOU, and 4 JEN at 15°C.

possibly because of the higher nutrient levels that may occur there. Fig. 22 shows the survival of FC and S. adelaide in effluent and sludge at 15° C for site 4 JEN. However, the results do indicate that survival is better in sludge than in overlying effluent. The fact that S.adelaide cells, which were derived in exactly the same manner, behaved similarly in effluents and not sludge, is supportive evidence for this observation. At the same time, the limited amount of data for FC in the first effluent experiment, compared with the second, suggests the same trend. There was a very large difference between the concentrations of S. adelaide in the two materials at day 16. By contrast, the differences between the behaviour of FC were less marked and presumed to be due to the fact that the indigenous FC population sampled after perhaps 4-5 days' detention in the septic tank system might be better adapted to the environment.

Data obtained on survival of S. adelaide and FC in soils influenced by septic tank effluents are presented in Parker and Mee (1982).

3.8 Adsorption of S. adelaide and MS2 phage to sands

Adsorption, a further mechanism for bacterial and viral removal from percolating effluent, was tested using both fresh and leached soils with a variety of fluids as supernatants.

These fluids were derived from the leachates from large columns (both alkaline and acidic) as well as distilled water.

3.8.1 S.adelaide

Time-course experiments were run for four hours with sampling at 0, 1, 2 and 4 hours. The results of tests with both Bassendean and Spearwood sands (fresh and leached) are shown in Figs. 23a and b. Bassendean sand did not adsorb S. adelaide cells under the conditions of these experiments. Spearwood sand showed no effective adsorption when fresh soil and alkaline leachates were combined, but there were differences between tests and controls when either distilled water and

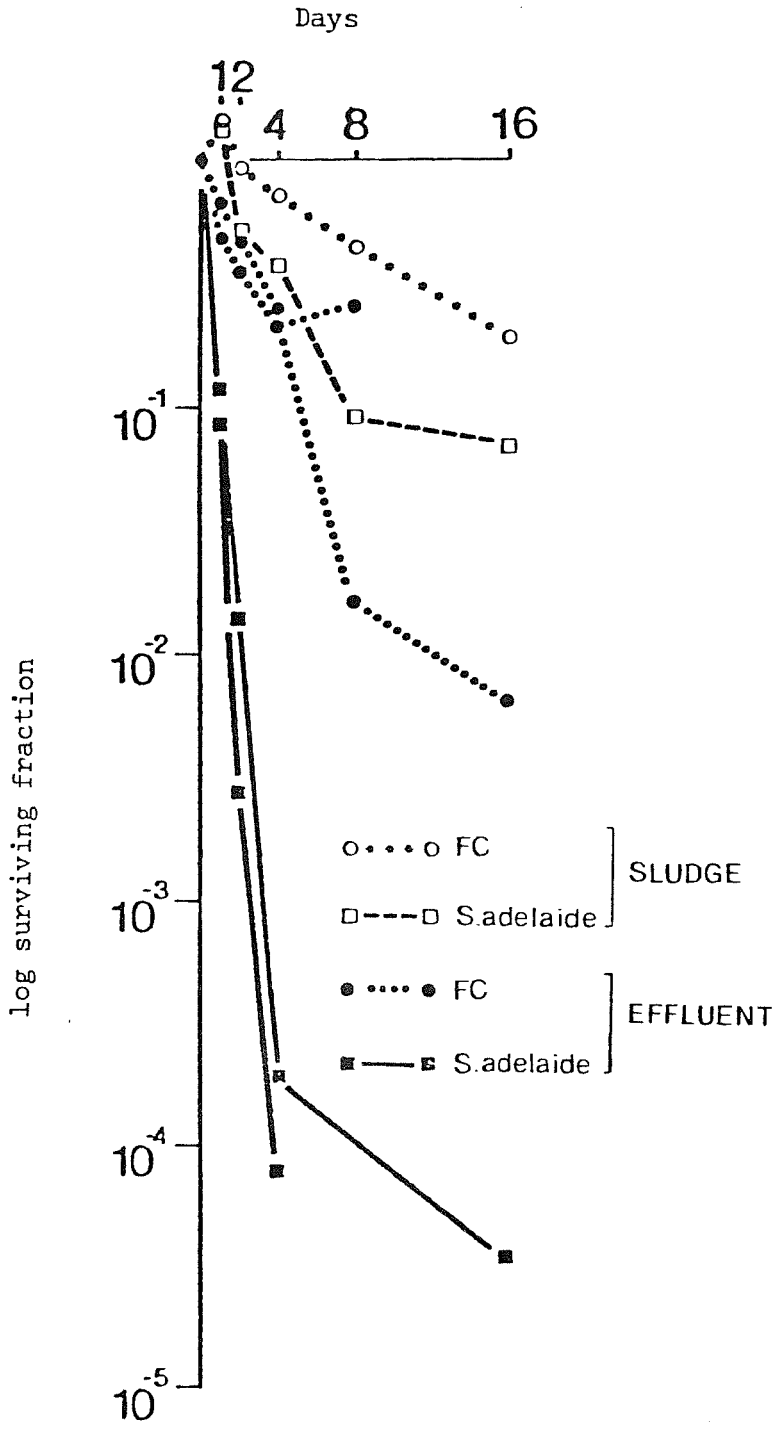


Figure 22 Comparative survival of S. adelaide and FC in sludge and effluent from site 4 JEN at 15°C.

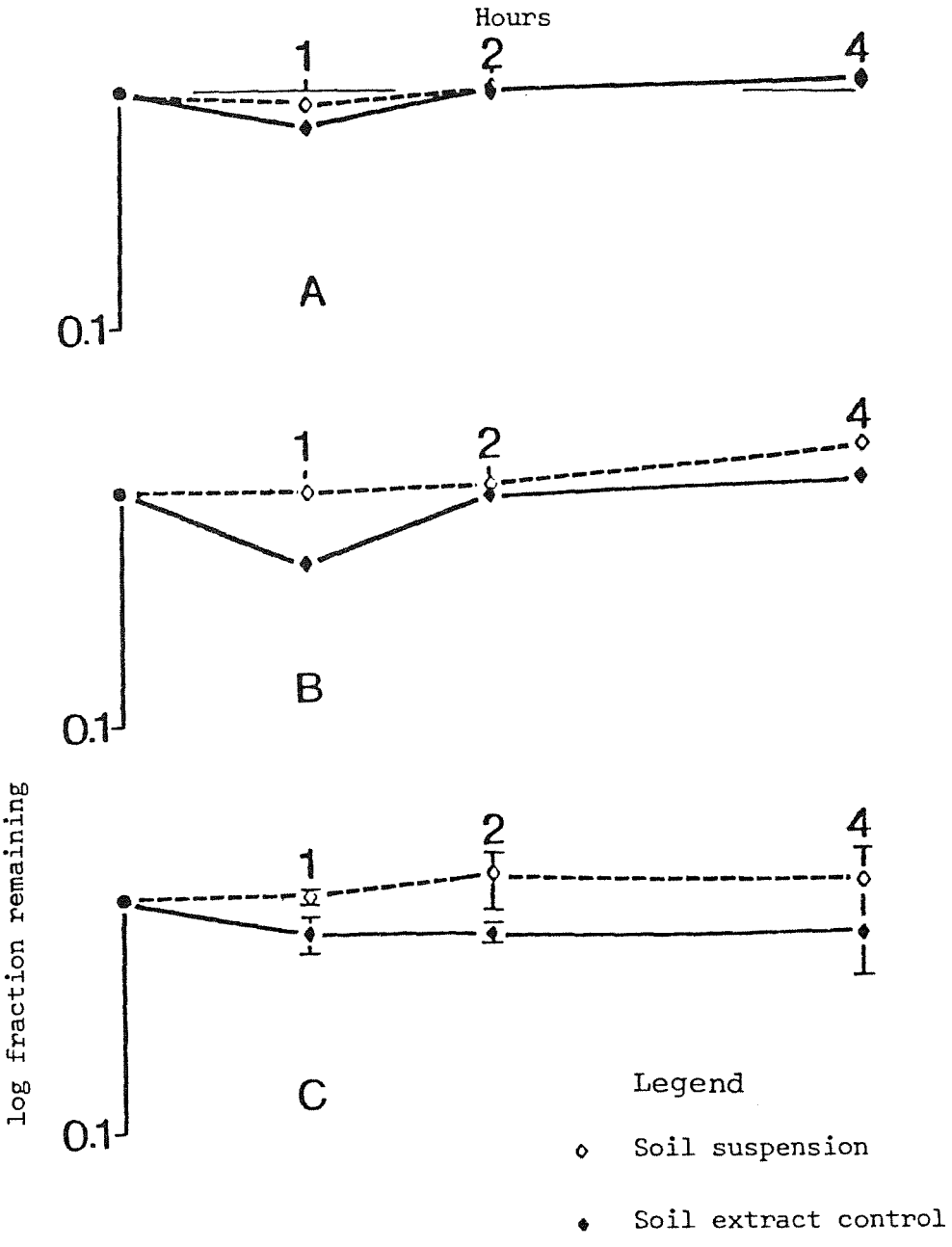


Figure 23a Adsorption of S. adelaide to Bassendean sand over four hours under the conditions :

a) fresh soil + distilled water

b) fresh soil + alkaline leachate

c) leached soil + acidic leachate

Each data point is the mean of four experiments. Bars indicate standard deviation (s.d.) and are absent when s.d. small.

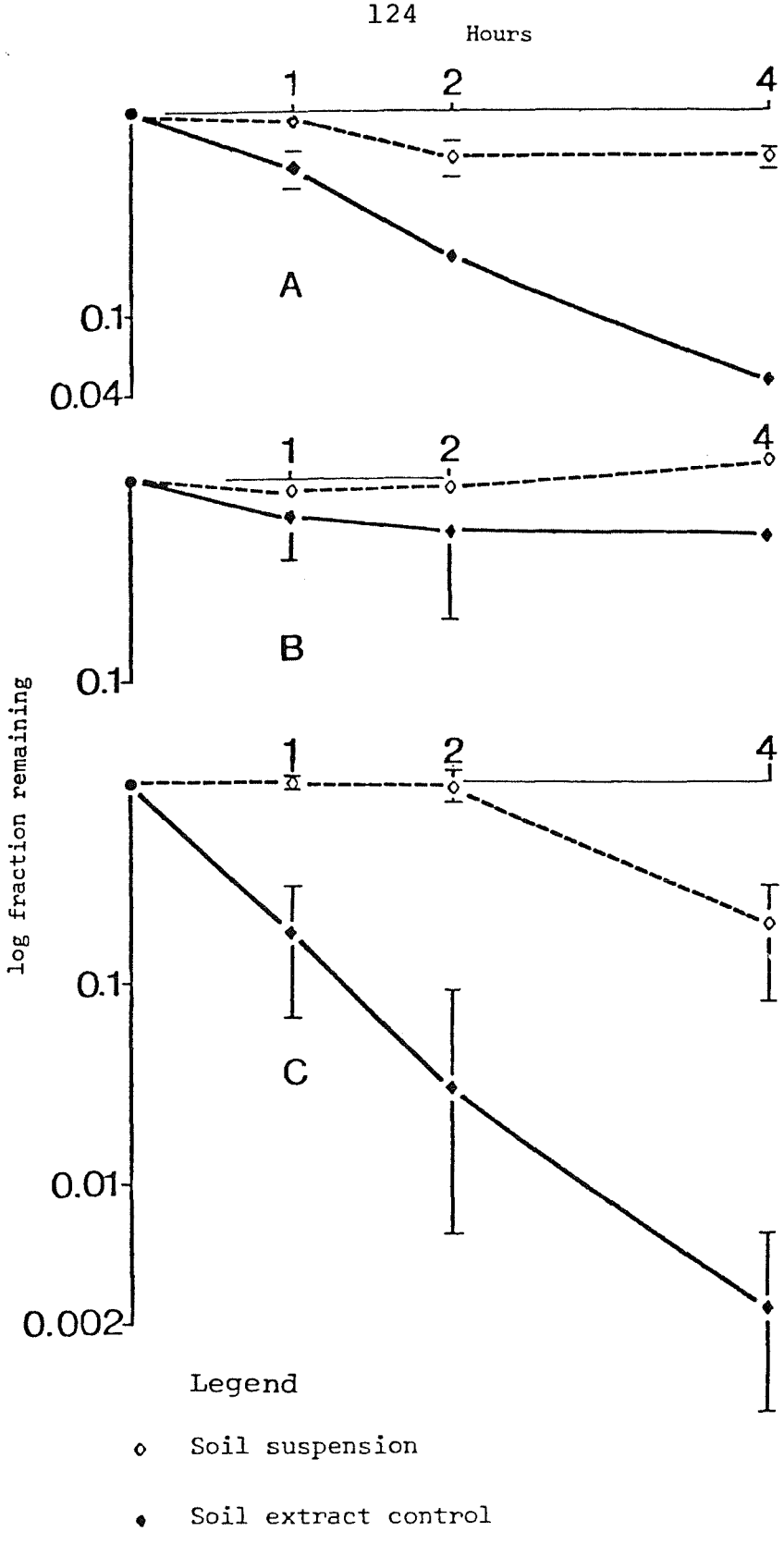


Figure 23b Adsorption of S. adelaide to Spearwood sand over four hours under the conditions;

a) fresh soil + distilled water

b) fresh soil + alkaline leachate

c) leached soil + acidic leachate

Each data point is the mean of four experiments. Bar indicates standard deviation (s.d.) and is absent when s.d. small.

fresh soil, or acidic leachates and leached soils were employed. In the latter case only, it was apparent that equilibrium was reached at four hours. For subsequent studies on the effect of cell concentration, a contact time of two hours was employed. The S. adelaide cells survived well over four hours, with little loss of viability.

A series of experiments was then conducted to examine the effect of cell concentration on adsorption, using the same protocols, but keeping incubation time constant and varying cell concentration. Experiments were thus performed using distilled water, alkaline and acidic leachates. The results of these experiments are shown in Tables 6a, b and c, and the data plotted as cells adsorbed per g vs. unadsorbed cells per mL , in Figs 24a, b, c and d. These latter plots are in the form of adsorption isotherms.

The observation in preliminary studies that when adsorption did occur it was generally of a low order, was confirmed by these studies. With distilled water as the supernatant fluid, the fraction of cells adsorbed by Spearwood sand was related to concentration (see Fig. 24a). These data suggested that as cell concentration increased, the fraction adsorbed decreased. At the highest cell concentration employed in one experiment (Table 6a) there was no statistically significant difference between counts. This suggests that at about 5×10^8 cells/g the maximum adsorption for this soil under these conditions is reached. However, the possibility of steric hindrance by the large numbers of cells present at this concentration may apparently decrease sorptive capacity. When the same soils in the fresh state were combined with alkaline leachates, the same soil comparisons were apparent (Table 6b). Bassendean sand, except for one result at high cell concentration, did not adsorb S. adelaide cells. However, for Spearwood sand the results were not consistent within each experiment and, for example, at one intermediate level no adsorption was detected. A relationship between cell fraction adsorbed and cell numbers adsorbed was not apparent in this case (see Fig. 24b) and the fraction adsorbed at all cell concentration levels except the highest was much lower than for

Table 6a: Adsorption of S. adelaide to Bassendean and Spearwood sands at four cell concentrations using distilled water and fresh soil.

Soil	Soil test supernatant (unadsorbed cells/mL)	Soil extract (cells/mL)	Cells adsorbed/g	% Cells adsorbed	pH of soil test supernatant at 2h
Bassendean sand	2.53×10^4	2.46×10^4	*	-	6.86
	2.75×10^3	1.22×10^3	*	-	6.81
	2.70×10^6	2.73×10^6	*	-	6.82
	3.16×10^6	2.32×10^6	*	-	6.79
	2.52×10^8	2.47×10^8	*	-	6.81
	2.13×10^8	2.08×10^8	*	-	6.90
	a 2.41×10^{10}	2.75×10^{10}	3.5×10^9	12.7	6.89
	2.7×10^{10}	2.43×10^{10}	*	-	6.92
Spearwood sand	1.14×10^4	3.05×10^4	3.82×10^3	62.6	5.88
	1.36×10^3	3.025×10^4	5.78×10^3	95.5	5.89
	1.1×10^6	3.00×10^6	3.80×10^5	63.3	5.93
	1.04×10^6	2.74×10^6	3.40×10^5	62.0	5.95
	1.4×10^8	2.90×10^8	3.00×10^7	51.7	5.91
	1.74×10^8	2.56×10^8	1.64×10^7	32.0	5.94
	2.13×10^{10}	2.53×10^{10}	8.00×10^8	15.8	5.99
	2.23×10^{10}	2.26×10^{10}	*	-	6.00

Legend

* - no adsorption detected.

a - soil test and extract values were compared statistically (t-test) in cases where only marginal differences in counts were obtained.

Note: Lower figures in each group indicate replicate experiment.

Table 6b: Adsorption of S. adelaide to Bassendean and Spearwood sands at four cell concentrations using alkaline column leachates and fresh soil.

Soil	Soil test supernatant (unadsorbed cells/mL)	Soil extract (cells/mL)	Cells adsorbed/g	% Cells adsorbed	pH of soil test supernatant at 2h
	2.19×10^4	2.13×10^4	*	-	7.77
	1.11×10^5	9.24×10^4	*	-	7.50
	2.40×10^6	1.75×10^6	*	-	7.49
Bassendean sand	3.06×10^6	1.90×10^6	*	-	7.50
	2.25×10^8	1.75×10^8	*	-	7.64
	1.99×10^8	1.95×10^8	*	-	7.51
	1.94×10^{10}	1.95×10^{10}	*	-	7.67
a	1.36×10^{10}	1.54×10^{10}	3.6×10^8	11.69	7.83
	2.3×10^4	2.34×10^4	*	-	6.67
	7.6×10^4	1.1×10^5	6.8×10^3	30.9	6.60
	2.07×10^6	2.08×10^6	*	-	6.63
Spearwood sand	1.7×10^6	1.45×10^6	*	-	6.70
	1.54×10^8	2.11×10^8	1.1×10^7	27.0	6.72
	1.2×10^8	1.66×10^8	9.2×10^6	27.7	6.65
	1.51×10^{10}	1.93×10^{10}	8.4×10^8	21.8	6.90
	1.4×10^{10}	2.05×10^{10}	1.3×10^9	31.7	7.00

Legend as Table 6a.

Table 6c: Adsorption of S. adelaide to Bassendean and Spearwood sands at four cell concentrations using acidic column leachates and leached soil.

Soil	Soil test supernatant (unadsorbed cells/mL)	Soil extract (cells/mL)	Cells adsorbed/g	% Cells adsorbed	pH of soil test supernatant at 2h
	8.4×10^3	2.21×10^4	2.74×10^3	62.0	4.81
	a 2.08×10^4	2.48×10^4	8.00×10^2	16.1	4.80
	4.90×10^6	9.8×10^6	9.8×10^5	50.0	4.60
Bassendean	1.85×10^6	2.78×10^6	1.86×10^5	32.9	4.62
sand					
	6.2×10^7	1.11×10^8	9.8×10^6	44.1	4.80
	1.75×10^8	2.88×10^8	2.26×10^7	39.2	4.78
	7.3×10^9	1.07×10^{10}	6.8×10^8	31.8	6.70
	2.33×10^{10}	2.37×10^{10}	*	-	6.61
	2.76×10^4	3.97×10^4	2.42×10^3	30.5	5.70
	1.70×10^4	2.41×10^4	1.42×10^3	29.5	6.11
	7.78×10^6	1.09×10^7	6.42×10^5	28.6	5.60
Spearwood	2.08×10^6	2.19×10^6	*	-	6.05
sand					
	5.4×10^7	1.19×10^7	*	-	5.90
	1.30×10^8	1.22×10^8	*	-	6.04
	5.4×10^9	8.9×10^9	7.00×10^8	39.3	6.90
	2.5×10^{10}	2.03×10^{10}	*	-	6.85

Legend as Table 6a.

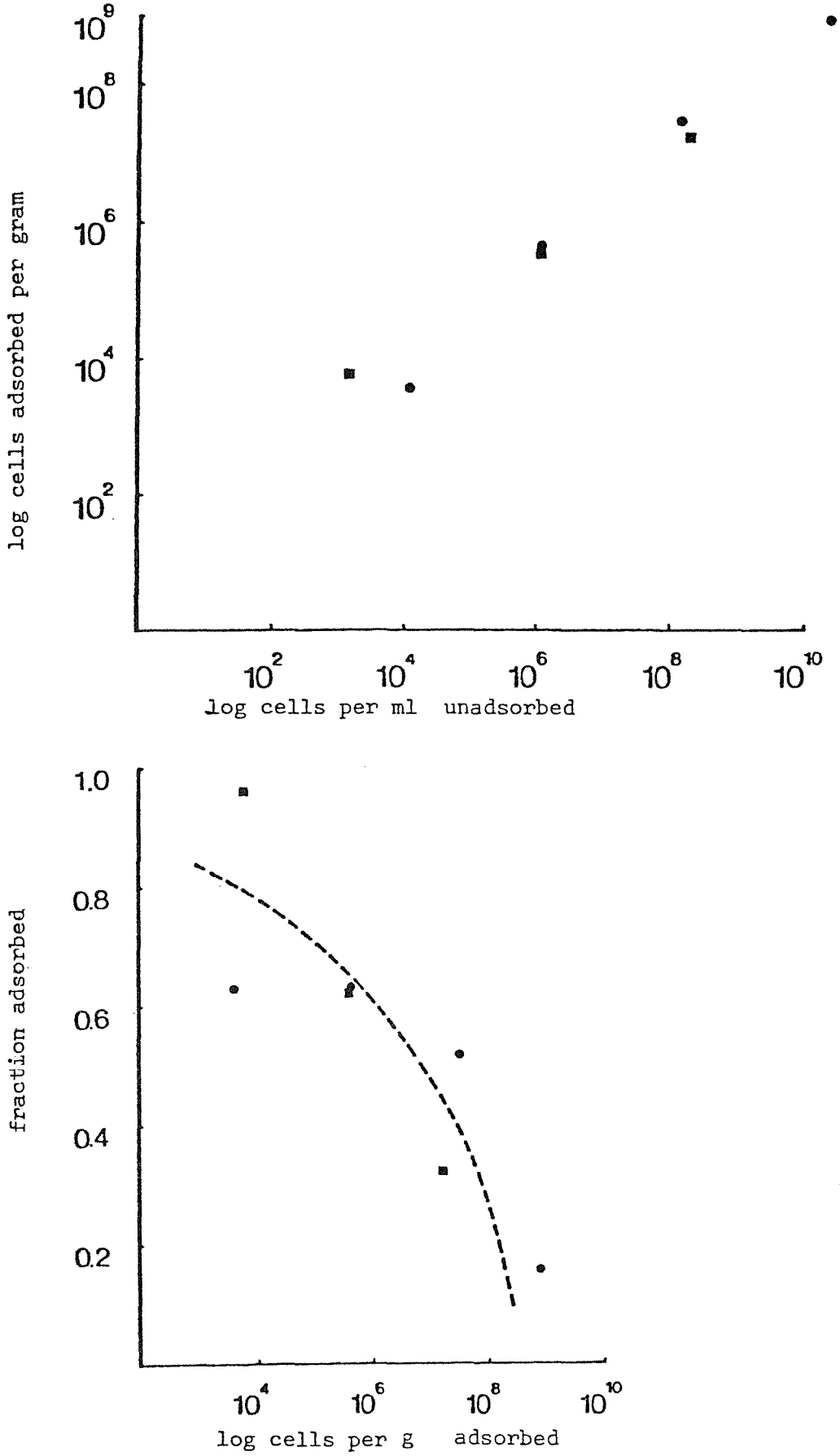


Figure 24a The effect of cell concentration on the adsorption of S. adelaide to Spearwood sand employing fresh soil and distilled water.
Different symbols indicate duplicate experiments.

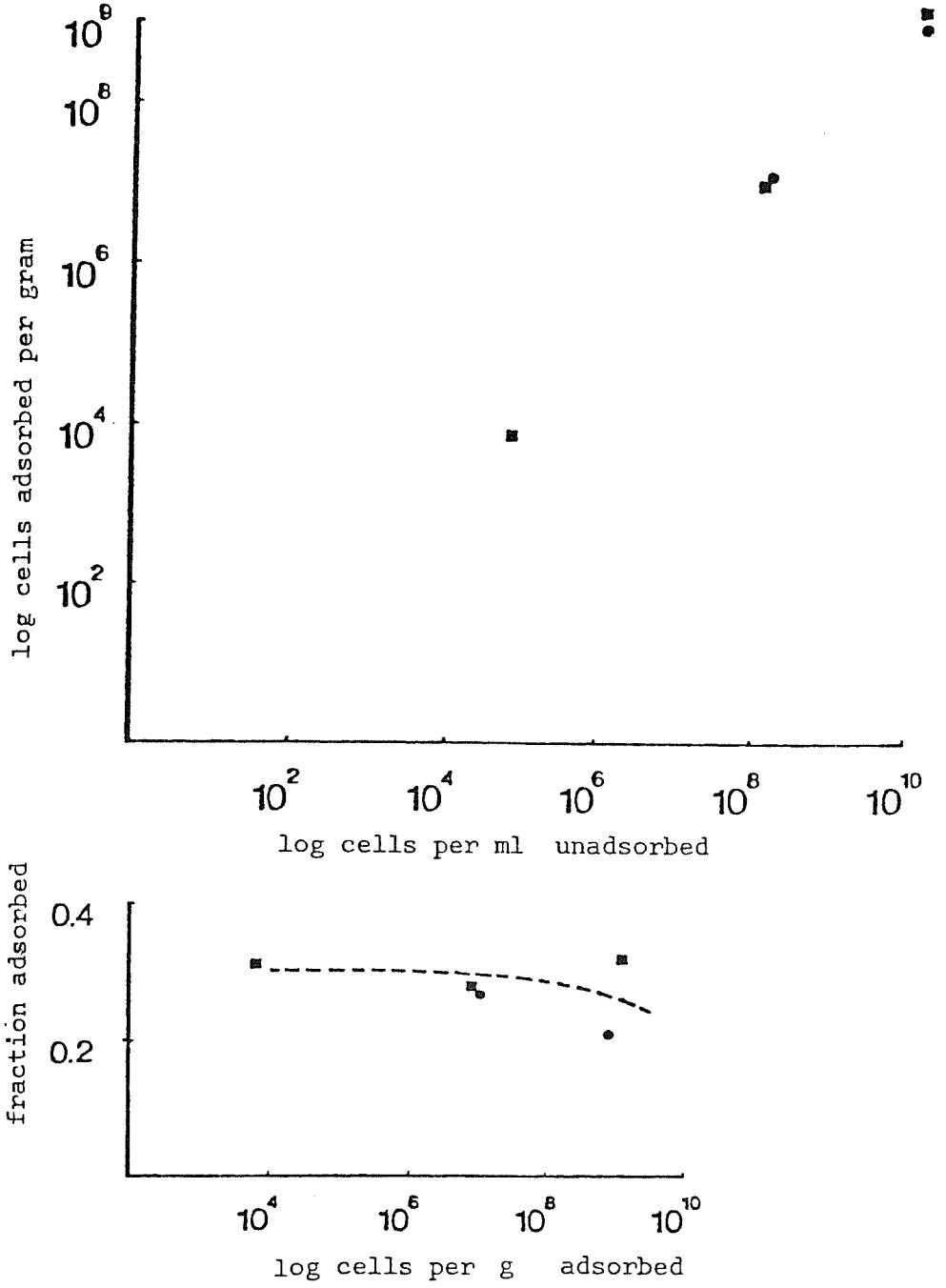


Figure 24b The effect of cell concentration on the adsorption of S. adelaide to Spearwood sand employing fresh soil and alkaline leachate.
Different symbols indicate duplicate experiments.

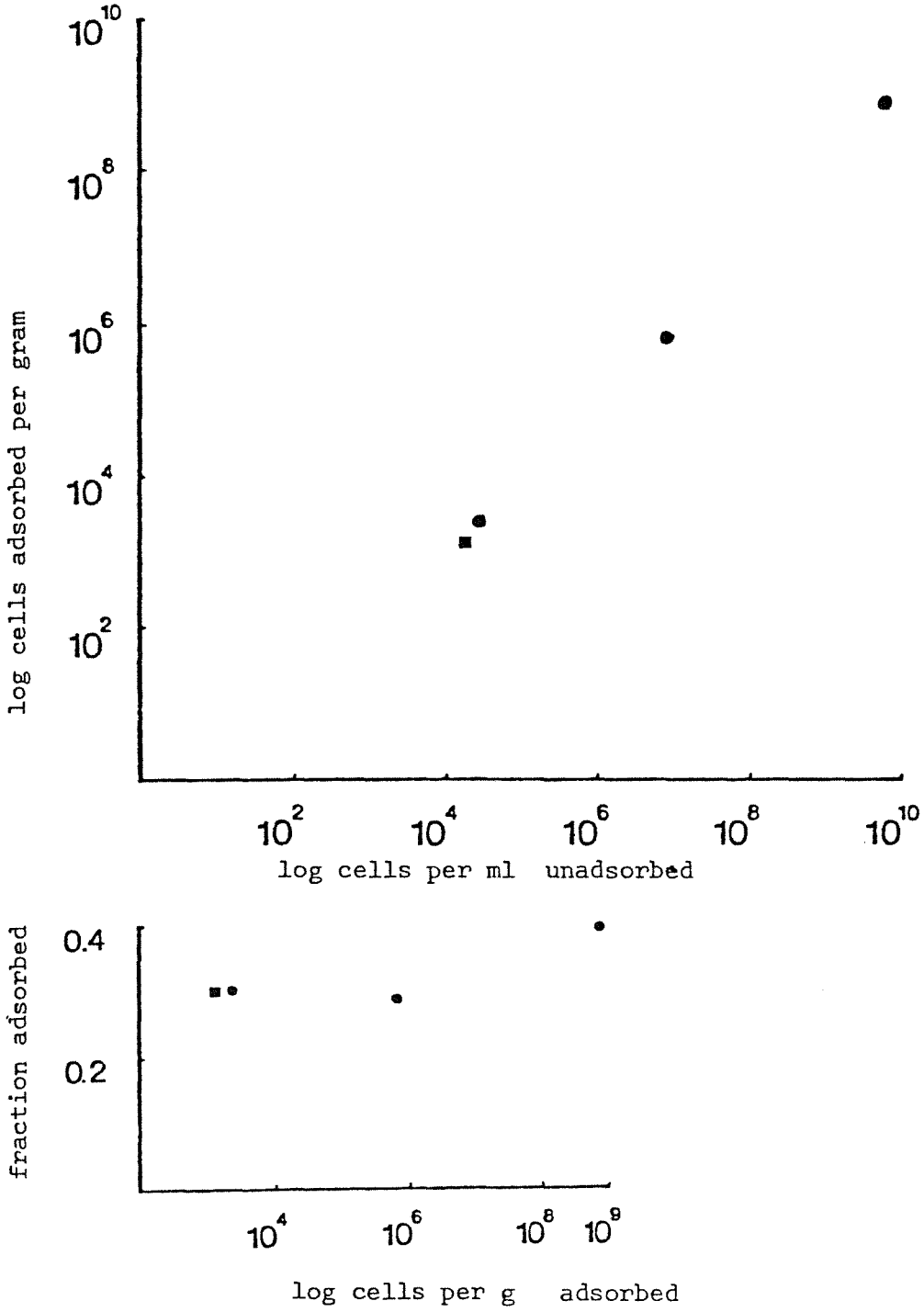


Figure 24c The effect of cell concentration on the adsorption of S. adelaide to Spearwood sand employing leached soil and acidic leachate.
Different symbols indicate duplicate experiments.

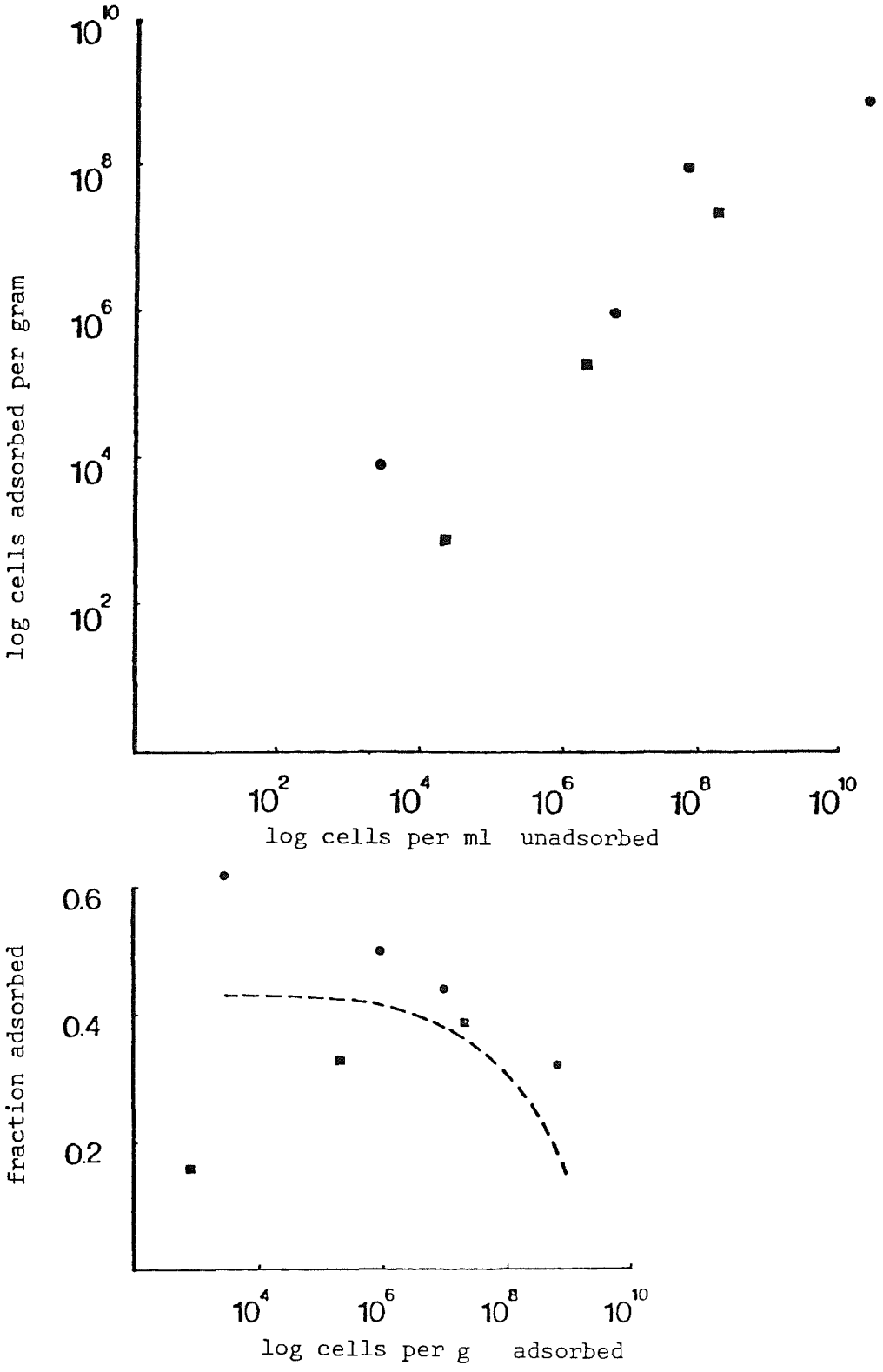


Figure 24d The effect of cell concentration on the adsorption of S. adelaide to Bassendean sand employing leached soil and acidic leachate.
Different symbols indicate duplicate experiments.

distilled water. When, however, acidic leachates were employed with leached soils, both soils adsorbed this particular bacterium. Considering Spearwood sand firstly, the data (Table 6c) show that, with one anomalous exception at an intermediate concentration, this soil adsorbed low levels of S. adelaide at all concentrations. As with alkaline leachate there was no apparent relationship between cell fraction adsorbed and cell numbers adsorbed (see Fig. 24c). The comparison between these experiments and identical experiments with distilled water and fresh soil suggests that the sorptive capacity was reduced in some way for leached soils. A further factor contributing to this conclusion is that pH values for both sets of experiments were similar. By contrast, there was a relatively high degree of adsorption of cells to leached Bassendean sand under acid conditions. A plot of cell fraction adsorbed vs. cells adsorbed (Fig. 24d) indicated that a possible relationship exists between these quantities. The fraction adsorbed at low cell concentrations for this soil was lower than for Spearwood sand under the same conditions although the probable cell concentration required to attain saturation was the same.

3.8.2 MS2 phage

Essentially similar experiments were conducted with MS2 phage. In general, the assumption was made that methods developed for bacteria would be appropriate and protocols for adsorption vs.time and adsorption vs.concentration were the same apart from preparation and assay of organisms.

Preliminary studies of virus adsorption to Bassendean and Spearwood sands suggested that the survival of the test organism was limited under the conditions of the experiments. Although survival in distilled water soil extracts was good, the survival in leached septic tank effluent was poor. Since subsequent experimentation involved the use of soil extracts, (and as such the virus was not exposed directly to leachates) the problem of virus viability was not felt to be serious.

It was pointed out that the adsorption of viruses to soils and other solids is time-dependent. Time-course experiments

Table 7a: Adsorption of MS2 phage to Bassendean and Spearwood sands at four concentrations using distilled water and fresh soil.

Soil	Soil test supernatant (unadsorbed PFU/mL)	Soil extract (PFU/mL)	PFU/g adsorbed	% adsorbed	pH of soil test supernatant at 2h
	a 2.48×10^2	2.8×10^2	0.064×10^2	11.4	4.35
	2.48×10^2	2.85×10^2	0.075×10^2	12.98	4.49
	1.85×10^4	1.78×10^4	*	*	4.48
Bassendean	1.68×10^4	1.13×10^4	*	*	4.61
sand					
	2.25×10^6	1.6×10^6	*	*	4.63
	1.98×10^6	1.43×10^6	*	*	4.52
	2.12×10^8	2.33×10^8	4.2×10^6	9.01	6.36
	2.38×10^8	2.68×10^8	4.4×10^6	8.46	5.98
	0.73×10^2	2.83×10^2	0.42×10^2	74.0	5.01
	0.26×10^2	2.50×10^2	0.45×10^2	89.6	5.05
	5.75×10^3	1.25×10^4	1.35×10^3	54.0	5.25
	2.2×10^3	1.1×10^4	1.76×10^3	80.0	5.37
Spearwood	5.0×10^5	2.15×10^6	3.3×10^5	77.0	5.23
sand	4.43×10^5	1.9×10^6	2.91×10^5	76.7	5.41
	8.83×10^7	2.53×10^8	3.29×10^7	65.0	6.16
	2.33×10^8	3.2×10^8	1.74×10^7	27.2	6.99
	2.45×10^{11}	5.38×10^{11}	5.86×10^{10}	54.5	6.49
	-	-	-	-	-

Legend as Table 6a.

Table 7b: Adsorption of MS2 phage to Bassendean and Spearwood sands at four concentrations using acidic column leachates with leached soils.

Soil	Soil test supernatant (unadsorbed PFU/mL)	Soil extract (PFU/mL)	PFU/g adsorbed	% adsorbed	pH of soil test supernatant at 2h
	0.85×10^2	0.25×10^2	*	-	6.08
Bassendean	4.25×10^3	5.5×10^2	*	-	6.13
sand+	4.5×10^5	7.4×10^4	*	-	6.20
	1.87×10^8	1.4×10^8	*	-	6.69
	0.125×10^2	1.25×10^3	2.48×10^2	90.0	3.55
	5.0×10^2	2.66×10^4	5.22×10^3	98.1	3.58
Spearwood	1.75×10^3	2.4×10^6	4.79×10^5	99.9	3.50
sand	4.12×10^4	1.18×10^7	2.35×10^6	99.7	3.61
	7.5×10^7	1.95×10^8	2.4×10^7	62.0	3.54
	3.82×10^6	1.23×10^8	2.39×10^7	96.9	3.81

+ - one experiment only.

Legend as Table 6a.

Table 7c: Adsorption of MS2 phage to Bassendean and Spearwood sands at four concentrations using fresh soils and groundwaters+.

Soil	Soil test supernatant (unadsorbed PFU/mL)	Soil extract (PFU/mL)	PFU/g adsorbed	% adsorbed	pH of soil test supernatant at 2h
	2.1×10^2	1.2×10^2	*	-	4.70
	1.98×10^2	1.23×10^2	*	-	4.66
	2.48×10^4	1.2×10^4	*	-	4.54
	1.625×10^4	1.7×10^4	*	-	4.63
Bassendean sand	1.68×10^6	2.00×10^6	6.4×10^4	16.0	4.72
a	1.60×10^6	1.70×10^6	2×10^4	5.8	4.90
	2.1×10^8	1.55×10^8	*	-	6.44
	2.55×10^8	2.30×10^8	*	-	6.51
	<5	1.2×10^2	*	-	6.39
	0.33×10^2	1.73×10^2	0.28×10^2	80.92	6.42
	1.83×10^4	1.25×10^4	*	-	6.47
Spearwood sand	2.00×10^4	1.25×10^4	*	-	6.48
	9.75×10^5	1.48×10^6	1.1×10^5	34.1	6.52
	8.25×10^5	1.07×10^6	4.9×10^4	22.9	6.49
	2.15×10^8	1.8×10^8	*	-	6.65
	1.5×10^8	1.3×10^8	*	-	6.59

+ - pH of natural groundwaters: Bassendean location pH 4.01; Spearwood location pH 6.48.

Legend as Table 6a.

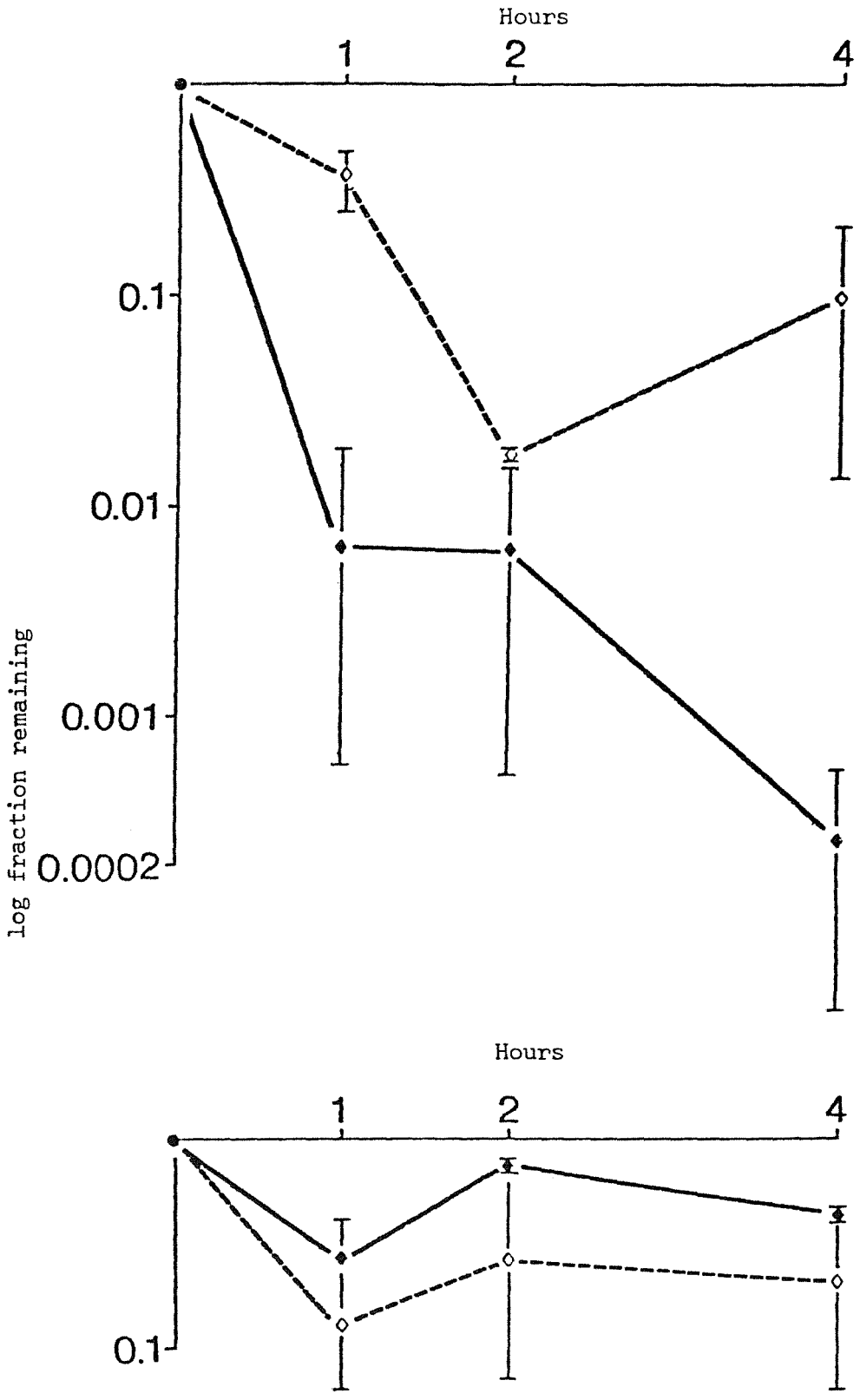


Figure 25a Adsorption of MS2 phage to Bassendean and Spearwood sands employing fresh soil and distilled water. Legend as Figure 23.
Upper curves Spearwood sand.
Lower curves Bassendean sand.

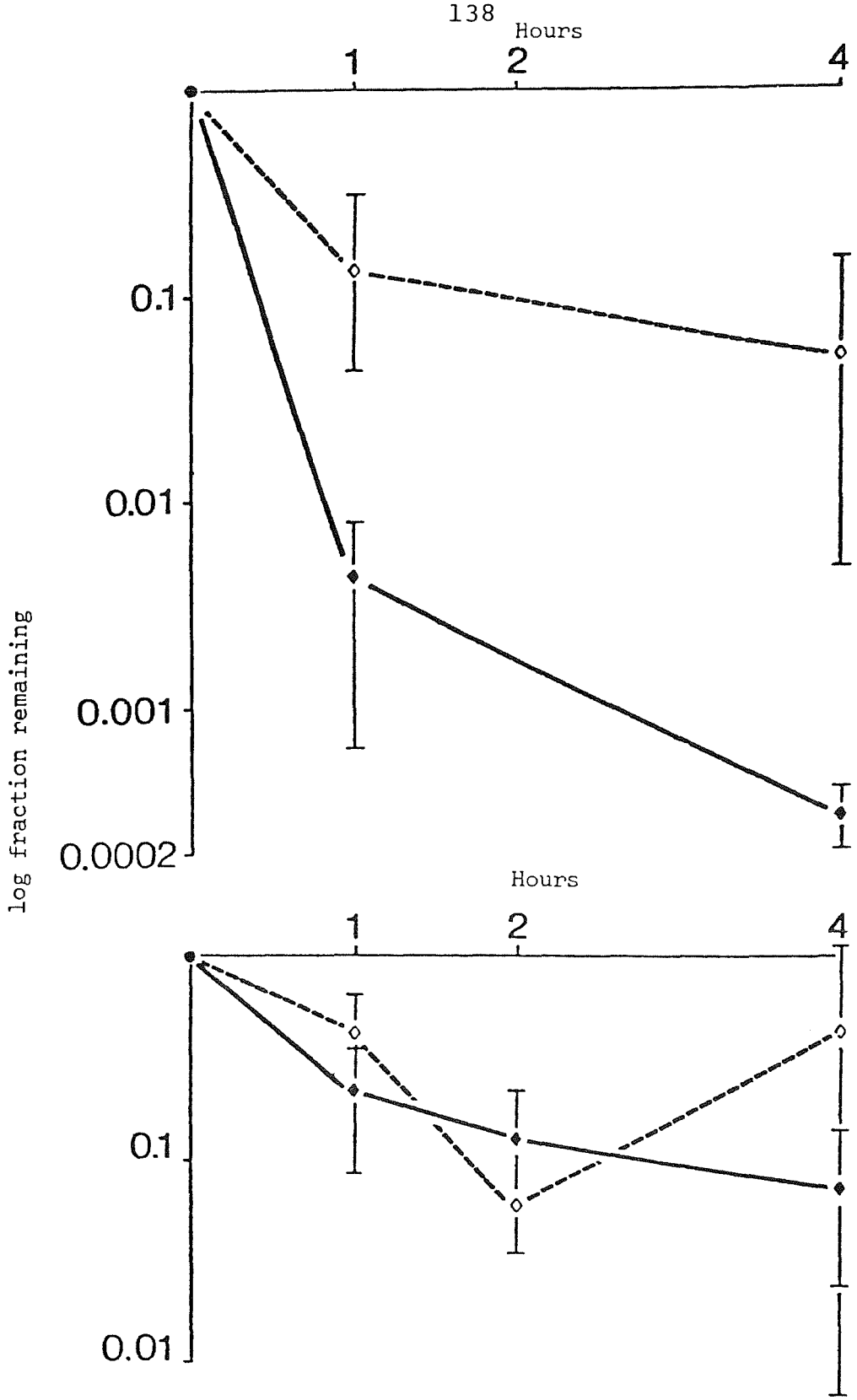


Figure 25b Adsorption of MS2 phage to Bassendean and Spearwood sands employing leached soil and acidic leachate. Legend as Figure 23.
Upper curves Spearwood sand.
Lower curves Bassendean sand.

were also conducted with MS2 phage in order to determine at what point an equilibrium occurred. Results of experiments with both fresh and leached soils are shown in Figs. 25a and 25b. It was evident that whilst fresh Spearwood sand adsorbed the virus, fresh Bassendean sand did not. Leached soil samples behaved similarly, but there was some indication that for incubation times in excess of four hours, Bassendean sand may adsorb the test virus. As with S. adelaide, an incubation period of two hours was selected for further studies designed to examine the effect of concentration on adsorption. The effect of MS2 phage concentration on adsorption to Bassendean and Spearwood sands using distilled water, acidic leachates, and two groundwaters is shown in Tables 7a, b and c respectively. At the time these experiments were conducted, alkaline leachate was no longer available.

The data were plotted as virus adsorbed per gram versus unadsorbed virus per mL in Figs. 26a and b. Again, these plots are in the form of adsorption isotherms. With distilled water as suspending fluid, MS2 phage adsorbed to Spearwood sand but not to Bassendean sand (Exceptionally there was low order adsorption at concentration extremes for this soil). It was evident that, compared with the test bacterium, the virus was more effectively removed from the supernatant fluid. The relationship between virus fraction adsorbed and virus per gram adsorbed (Fig.26a) showed that some 10^{12} PFU per gram might be adsorbed. Unlike the bacterium, larger proportions of lower concentrations were adsorbed. Both of these findings are consistent with the comparative sizes of the test organisms. However, with this consideration in mind, the degree of adsorption was of a low order. A contrasting pattern was obtained with acidic leachate and leached soil. Under these conditions Bassendean sand did not adsorb the virus. Leached Spearwood sand (Fig.26b) adsorbed much greater fractions of virus than fresh sand.

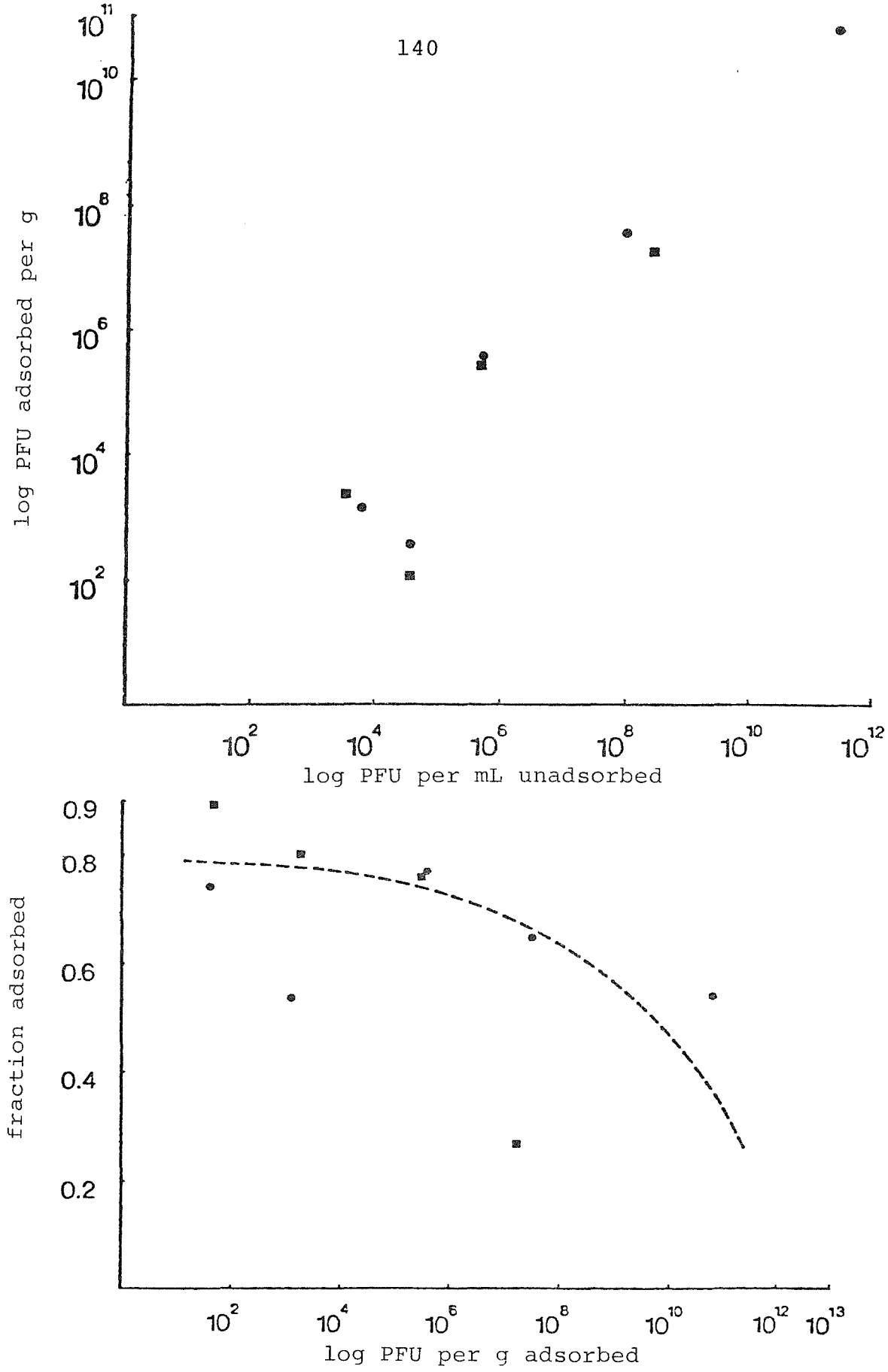


Figure 26a The effect of concentration on the adsorption of MS2 phage to Spearwood sand employing fresh soil and distilled water.
Different symbols indicate duplicate experiments.

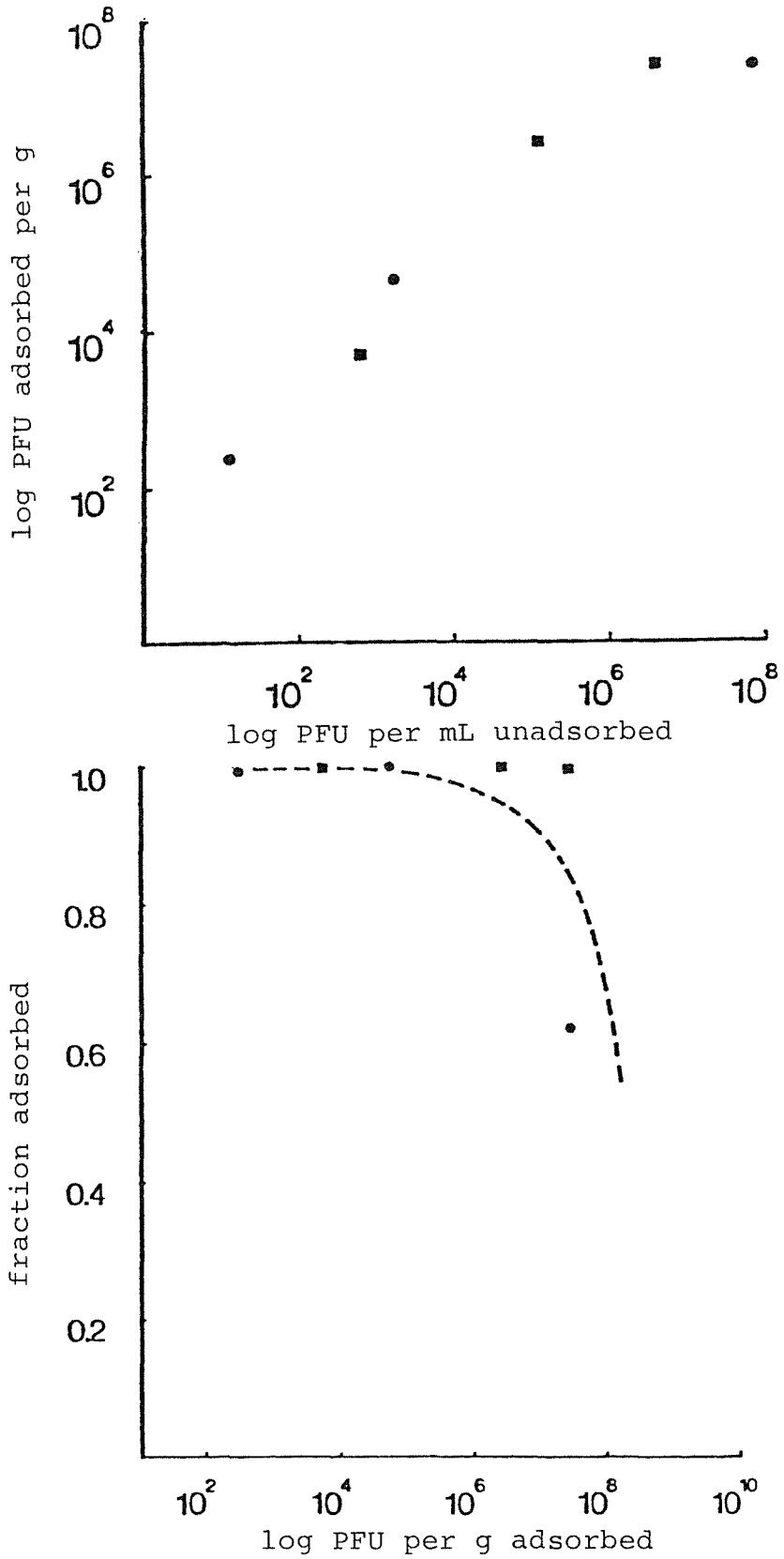


Figure 26b The effect of concentration on the adsorption of MS2 phage to Spearwood sand employing leached soil and acidic leachate.
Different symbols indicate duplicate experiments.

Whereas fresh Spearwood sand was apparently capable of adsorbing substantial quantities of virus, the leached soil was not. The isotherm obtained for the latter material was not a straight line and at the higher concentrations the data points tended to flatten. Taking the same data and comparing virus fraction adsorbed with virus per gram adsorbed (Fig. 26b), it is evident that although higher initial concentrations were adsorbed, the soil material was saturated at lower titres. By extrapolation, a saturation value might be between 5×10^8 and 10^{10} PFU per gram.

Two natural groundwaters were also studied and no consistent adsorption occurred (Table 7c). In other experiments, Bassendean sand, even when acidic, did not adsorb the virus and the results with acidic groundwater were the same. It is possible also that the tendency of the second natural groundwater (removed from a well in limestone strata underlying Spearwood sand) to exhibit a reaction approaching neutrality may have been responsible for the lack of virus adsorption under these circumstances.

4.0 DISCUSSION

4.1 General

Sewage disposal has become a sophisticated technology in most industrialized nations and the possibility of treatment of wastewater to a very high standard exists. It is technically possible to convert raw sewage to potable drinking water within one plant.

Yet despite this sophistication, the most primitive and simple sewage disposal methods are still in use in some of the world's richest nations. In the USA for example, Cooper and Resek (1977) estimated that in 1970, 16.6 million housing units used on-site disposal systems and this number was growing by half a million annually (Patterson et al. 1971). Although more sophisticated designs are available nowadays, it is quite likely that a large proportion of existing septic tank systems in the USA and elsewhere are little different from the design of Philbrick (1883). These statistics may appear as a surprising contradiction to the popular belief that such primitive disposal systems are a thing of the past. The reasons for large and increasing numbers of on-site disposal systems are economic and compounded, at least in the USA, by geographical considerations.

There are numerous wastewater disposal systems available, but as the level of sophistication rises, so too does the cost. A significant increase in cost per capita occurs as soon as there is a requirement for collection sewers capable of handling raw sewage. The costs of the planned sewerage installations in the Perth Metropolitan Area for the period 1981-1986 are some A$ 269 million (MWB 1981), and over 70% of this will be spent on reticulation and pumping stations. The treatment works costs account for under one third of the total. From this it will be apparent that if a population is dispersed the costs will rise further. In the USA for example, there has been a trend for populations to move into rural areas (Beale and Fuguitt 1975, cited by Kriessl 1978) and in many cases, these populations are

beyond the reach of sewerage.

The growth rate of populations is also a factor that must be considered here. Perth, for example, has the highest growth rate of any Australian capital city. Between 1966 and 1976 the city grew by 47%. Adelaide, a comparable city away from the main southeastern population centre grew by 18% in the same period. (Australian Bureau of Statistics 1966, 1976). The rapid growth of Perth has meant that sewerage installation has not kept pace. This is reflected by the peripheral distribution of septic tanks (Fig. 4). Houghton's analysis of the 1976 census shows the same pattern (Houghton 1979).

There has been a recognition for Perth that septic tanks with direct on-site disposal, or other systems intermediate between on-site disposal and the collection system, will be used permanently (Fimmel and Troyan 1981). The implication of this is clear. Potential or actual groundwater pollution caused by such systems must be controlled by design and/or appropriate siting. In turn this requires that groundwater pollution be defined and the fate of both chemicals and microorganisms in the soil be determined for the specific soils in question. This study has been restricted to the study of sewage microorganisms in two major sand soils largely because they occupy the largest land area, but also because they have the highest population density.

It has been argued that the safety of septic tanks with respect to water-borne infections cannot satisfactorily be determined by epidemiological methods, nor can experimentation be done, at least in an uncontrolled suburban area, by following the fate of deliberately introduced pathogens. A number of indirect approaches are available. Overall conclusions may be drawn by integrating the results of a range of studies covering the environmental parameters expected to influence microbial survival and movement.

4.2 Soil pollution by septic tank effluent in coarse sand soils.

At the commencement of this study there were only two known surveys where unsaturated soil receiving effluent had been examined; Caldwell (1938c) and Bouma et al. (1972). Both studies, especially the latter, indicated that no penetration of faecal microbes would be detected beyond about 0.50m. The technique of sample collection was by simple excavation and coring. Some use has been made in other field studies of ceramic tensiometers, but for the reasons discussed elsewhere and the difficulty of installation, these were not used here. There are some limitations to the technique used in this study; for example, there may have been some underestimation of FC counts because organisms were stressed. But it is reasonable to assume that the results obtained were a good indication of the comparative efficiencies of the soils, thus fulfilling the major aim of the survey.

Some evidence of groundwater pollution, presumably arising from septic tanks, was obtained in a preliminary study of so-called 'failed' systems. At sites 34 DUN and 24 WAL (Table 3), in Spearwood and Bassendean sands respectively, there were TC, FC and FS present in adjacent groundwater samples. At the first site the septic system was 25 m laterally from the well and although no information regarding groundwater movement was obtained, the indication was that well organisms originated in the septic tank. This suggests that organisms were able to migrate through soil. The area, it should be noted, has underlying porous rock and saturated flow therefore may have been possible. The well was securely covered and in good condition. Water abstracted from the bore spear was negative.

At the second site, the septic system was installed contrary to regulations (Health Act 1911-1978) and the bore was situated 2 m from the edge of the second soak well. From the analysis of the bore water it was evident that some

faecal contamination was occurring, although this may have originated in a neighbouring septic tank. Whether organisms were migrating through the soil directly or laterally on the surface of a hardpan and down the bore casing could not be determined. Presumably such pollution would not have occurred if the bore had been located further away.

The results of the preliminary survey also showed that on passage through two soak wells there was generally a reduction in bacterial counts. In many cases the first soak well was functioning as a sealed system (i.e. a septic tank) with no, or very little, infiltration.

The results of the more extensive survey of effluents and sub-soils have been discussed in Parker et al.(1981). Data from the additional five sites were similar. It is worth emphasising the observation that in this study greater numbers were found beneath septic systems compared with the findings of Bouma et al.(1972). Apart from the possibility of improved desorption, it is also possible that the mFC and m-endo media used in this study were more effective than the medium used by Bouma and his colleagues. The comparison cannot be taken further without an extensive evaluation of coliform media using field samples.

The major conclusion that can be drawn from the survey is that a coarse sand, devoid of clay, e.g. Bassendean sand, is not an appropriate soil for microbial removal. This is particularly so in view of the tendency for high water tables in certain areas. By comparison with soil and site factors that may be combined to determine the suitability of soils for septic tank operation (Wall and Webber 1970), Bassendean sand appears to fall within class 4 of a total of 5; class five being the poorest category. In terms of indicator concepts and bearing in mind the data of Grunnet and Olesen (1978) regarding the value of TC organisms as indicators, it would appear that bacterial (and presumably viral) groundwater pollution may occur in unsewered

suburbs on Bassendean sand where high seasonal water tables occur. In contrast, indicator counts, with the exception of FS, declined markedly in Spearwood sand with depth, and groundwater pollution is much less likely (a) because of efficient soil removal, but also (b) because of comparatively greater depths to the water table. Without the benefit of more extensive data, conclusions concerning Quindalup sand cannot be made, but on the basis of very limited FC data, the removal of bacteria was not as great as might be expected from a consideration of the high calcium content.

There is some indication in both sets of data for Bassendean and Spearwood sands that FS might be effective indicators of faecal pollution. These organisms, although less numerous, did not decline to the same extent as the coliforms, and for Bassendean they behaved very similarly to the TC at depths of > 0.35 m. This may be an indication of greater survival potential or a lesser degree of sorption.

In general therefore, some doubt must be cast on the accepted standard depth of soil (Wagner and Lanoix 1958) for proper microbe removal in the case of Bassendean sand. A depth of 1.2 m may be quite insufficient. However, a more extensive survey of bores, wells and surface drainage systems would be necessary to accurately delineate areas where septic tanks should be removed or prohibited in new developments. Such a survey may be thwarted by the simple problem of gaining access to suburban gardens, and not being able to undertake appropriate sampling. The study discussed in Whelan and Parker (1981) is an example. In this case, a larger number of wells would have been an advantage, but both fruit trees and lawn prevented this. However, the results of that survey did indicate that FC organisms were able to reach the groundwater, despite the fact that soil core analysis showed a decline with respect to depth suggesting no such migration.

4.3 Conclusions from the controlled laboratory approach to soil removal of sewage microbes.

4.3.1 Static studies

Once released into the external environment enteric bacteria and viruses exhibit die-off. The rate of die-off depends on the organism in question, temperature, moisture and the presence of protective materials and nutrients. It is very difficult to compare die-off rates for bacteria and viruses, but it is generally observed that enteric viruses have a better survival potential. Die-off is, then, one 'removal' mechanism. The adsorption or entrapment of organisms is another, and again it may not be meaningful to make direct comparisons between bacteria and viruses or even between different strains of the same type. Clearly, the two phenomena are not mutually exclusive, and in the study of one, the experimenter is obliged to control the other. In the field however, the two phenomena operate together and the extent of penetration of a bacterium or virus into the groundwater is presumably affected by each. The relative importance of each phenomenon may be hard to evaluate in the field situation. Furthermore, the results of laboratory experiments may be difficult to rationalise in terms of behaviour from field experiments.

The results overall of survival experiments with bacteria show that indicator organisms and S. adelaide declined in effluents and sludges, although in the latter there was some evidence that organisms survived longer. In soils (Parker & Mee (1982) the pattern was different and the initial rapid declines, followed in some cases by slower declines, were not seen. Instead there appeared to be in most cases some suggestion of oscillation between decline and regrowth In general the reduction over sixty four days was markedly less in soils than effluents. It appeared that when the combined results were analysed there was a difference between soils, although it is not evident that survival in one soil was greater than the other. When other analyses were done, no differences due to soil were apparent. From

this it is reasonable to conclude that observed differences between soils in field and column data are not likely to be due to intrinsic differences in soils causing different die-off patterns.

The second aspect of bacterial and viral removal from percolating effluent involves charge phenomena. As has been stated, bacteria and viruses are removed from percolating effluents by 'straining' and adsorption respectively. Since both bacteria and viruses are charged entities, adsorption may be an important mechanism for both. The overall results of soil comparisons for bacteria and viruses suggest this, although the degree of adsorption differed in some cases between S. adelaide and MS2 phage. A particular case is where leached soil was employed with acidic leachate, the conditions occurring in the soil 'column' beneath the septic tank system. Under these circumstances the bacterium was adsorbed, albeit poorly, to Bassendean sand whereas the virus was not. Conversely, the bacterium was adsorbed poorly, but the virus adsorbed well to Spearwood sand. It may be possible to explain these results in terms of pH differences. For reasons unknown, the pH values of soil solutions for Bassendean sand were, for the S. adelaide tests, generally less than pH 4.8, except at high cell concentrations. They were somewhat higher (pH 6-6.7) for MS2 phage tests. A similar pH contrast occurred for Spearwood sand, although in reverse. The contrast between fresh soils with 'early' alkaline column leachates and leached soils with acid leachates is apparent for both soils and both test organisms. For S. adelaide, both soils were poor adsorbents when fresh materials and alkaline leachates were employed, but when leached soils and acid leachates were employed the adsorption to Bassendean (effectively nil) remained unchanged, but Spearwood adsorbed cells. With the virus, the adsorption to Bassendean was nil in both cases, but under both circumstances there was adsorption to Spearwood. This indicated that if the soil has the capacity to adsorb in the septic tank environment then this can occur earlier

in the life of a system for viruses, and bacterial adsorption may occur only after the soil is leached by effluent.

The capacity of a soil to adsorb bacteria and viruses is important. Again, considerations of leached soils must be made because of the accumulation of effluent chemicals and suspended solids on surfaces competing or preventing access to micro-organisms.

4.3.2 Dynamic studies

It has been noted that it is more important to obtain reliable information about the movement of pathogens rather than indicators. Although various pathogens are present in septic tank systems, it requires exceedingly large surveys to obtain useful data (Senault et al.1965). The attempt to obtain, for example, salmonella counts with respect to depth in the same way as coliforms is impossible. There are two alternatives. One is to deliberately 'spike' effluents and thence follow the pathogen in the soil. This is unethical, particularly in suburban areas, as indeed might be the practice of using antibiotic-resistant bacteria (Hagedorn et al. 1978). The second is to use controlled columns. Provided they are constructed properly and operated correctly, large soil columns, particularly for coarse unstructured soils, are a valuable tool. The critique of Bitton et al.(1979) concerning proper column packing and operation is noteworthy in this regard.

In this study, the principal aim was to both design and operate a series of large columns so as to re-create in the laboratory the soil 'column' beneath a functioning septic tank system. For the large column studies pathogenic salmonellas as well as E. coli were selected because of their widespread occurrence in both clinical and external

water environments (SHLS 1977), and also because of their hardy nature when compared, for example, with the shigellas (Papavassiliou and Leonardopoulos 1978). MS2 phage was selected because of its similar morphology to enteroviruses, ease of handling and the accuracy of its quantitative determination.

At the outset, it can be asked what physical phenonema affect the movement of microbes in soil disposal systems. A number of papers have reported that flow rate is important in removal of bacteria and viruses. Wang et al. (1980) suggested that 'flow rate of water ... may be the most important factor in predicting the potential of virus movement into the groundwater'. These authors were studying a range of four soils. Generally, the slower the flow rates were, the better the removal tended to be. Green and Cliver (1974) similarly suggested that virus removal was lessened when sand columns dosed with septic tank effluent were saturated. FC and FS removal was improved when columns were dosed at flow rates of 5cm/day compared with 10cm/day (Ziebell et al. 1974). Vaughn et al. (1981) observed differences in removal over a wide range of rates and noted greatest removal between 12-24 cm/day compared with 1800-2400 cm/day. Evans and Owens (1972) showed that the numbers of E. coli and enterococci in a land drainage discharge were related to the flow rate of that discharge.

In the preliminary phase of this study the FC content of the leachates was clearly affected by flow rate (as high as 100 cm/day for Ba 2 and Sp 2). As these flow rates reduced, FC counts reduced. At the same time, other chemical and physical changes were occurring and these may have had an effect. However, during the second experimental phase it was possible to compare the removal efficiencies of Ba 1, Ba 2 & Sp 1, Sp 2. The slower rates of flow of Ba 1 and Sp 1 and better removal efficiencies strongly support the argument that flow rate is critical. Vital data for the same columns is different. When input concentrations of MS2 phage were > 10^8 PFU/mL and up to 10^{10} PFU/mL there was very little breakthrough of virus in

columns Ba 2 and Sp 2, but there was with the slower flowing Ba 1 and Sp 1 columns. This breakthrough was detected only at the commencement of the dosing and apart from column Ba 1 where virus was detected on three consecutive days, the virus appeared in the other column leachates only very briefly following dosing on day 306. There may have been carry-over of host cells leading to stimulation of protozoan predation, or poor intrinsic survival.

A second factor which may be interrelated with flow rate, for finer textured soils, is soil composition. The differences for these coarse sands would be of negligible importance as is indicated by the moisture characteristic curves. However, there were notable differences in bacterial and viral removal between soils when flow rates were similar, irrespective of column length. It was evident again that Bassendean sand was less efficient at purification.

Breakthrough of Salmonella spp. and E. coli was continual for Bassendean sand, but only intermittent for Spearwood, with the same input. The same soil comparison is not as strong for the virus. For the slower Ba 1, Sp 1 columns, virus breakthrough was similar, but for the faster flowing columns the breakthrough was different, Spearwood sand being a more effective "filter".

The removal of bacteria for both soils in 0.3 m columns was poor and soil differences negligible. But in the case of the MS2 phage, the removal through Spearwood (column Sp 3) was almost complete compared with Bassendean (Ba 3) where there was considerable breakthrough. The reason for this is not known.

The comparisons seen between the sorptive interactions of bacteria and the virus when leached soils were used were not apparent in these experiments.

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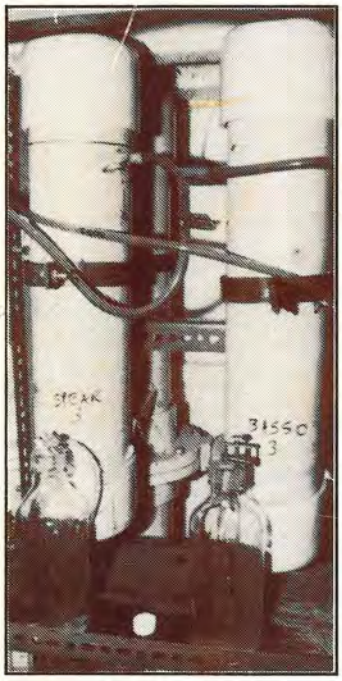


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