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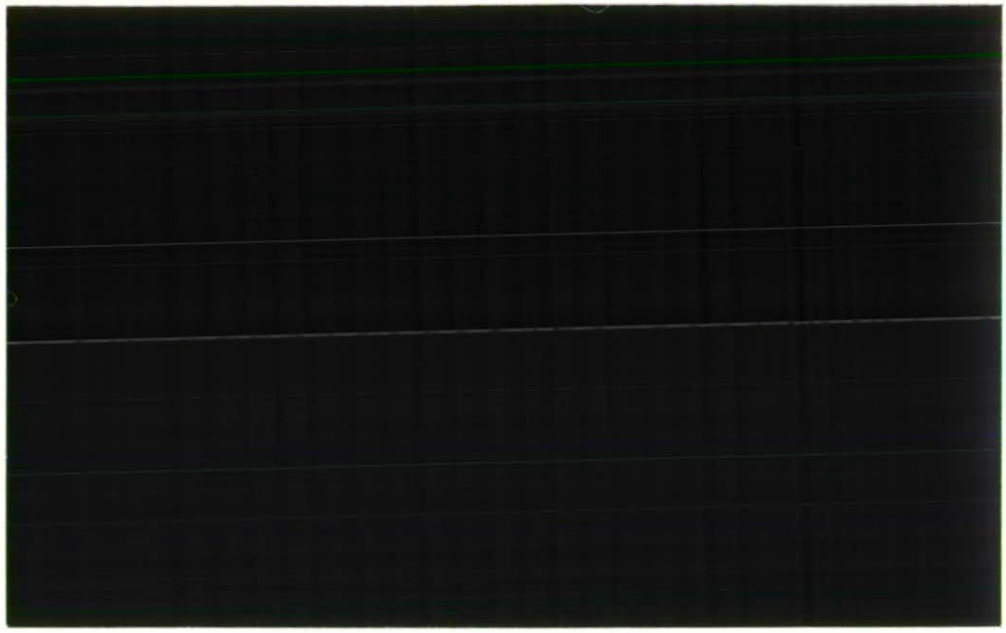

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MODELLING BACTERIAL CONCENTRATIONS

IN STREAMS

DRAFT Literature Review: The Sources, Survival, Transport and Modelling

of Faecal Coliforms in Catchments and Rivers.

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Executive Summary

This literature review gives a generalised overview of the key influences on the transport and survival of faecal bacteria, namely the faecal coliforms and *E.coli*, in catchments and drainage waters. Bacterial survival is influenced by factors of physical, chemical and microbiological nature. The relative influence of the different factors depends on the catchment compartment under consideration. On catchment surfaces, where bacteria become adsorbed to soil particles and plant surfaces and may persist for many months, survival is largely determined by moisture, sunlight and extremes of temperature. In the sub-surface zone where short-term environmental variations are attenuated and light cannot reach, metabolic influences related to nutrient supply and the effects of competition and predation by the natural microbial community dominate the rate of die-off. In surface waters, adsorption of bacteria to solids leads to removal from the water column and into the more stable bed sediment environment where continued accumulation can produce a large reservoir of bacteria. Disturbance of the bed sediments releases bacteria into the water column. The importance of hydrological factors stems from the link between rainfall and bacterial transport from non-point sources and the increased bacterial concentrations produced during high flow events. In a river channel the net transport of bacteria is determined by both bacteria suspended in the water column and those deposited and released from bed sediments. The bulk of modelling studies have focused mainly on bacterial die-off dynamics, although a small number of studies utilise a more deterministic or process-based approach to modelling bacterial transport dynamics, examples of which are reviewed.

1.Introduction

1.1 Bacterial Water Quality.

Bacterial water quality became an issue in Britain's growing industrial centres following outbreaks of typhoid and cholera in the late 19th century. These were largely caused by drinking water becoming contaminated with raw sewage. This contamination stemmed from the dual role of lowland rivers in built up areas, that is, their use for both water supply and effluent discharge. The engineering solution to this problem was quickly derived; separate sewage and water supply functions. To achieve this the uplands were utilised as sources of potable water, their catchment areas being free from human contamination. Further population growth, has resulted in the renewed role of lowland rivers for water supply, supported by vastly improved water treatment facilities. Such water works, however, are of limited capacity and bacterial and chemical water quality has further deteriorated as upland water bodies are more frequently used for recreational purposes, and as pressure on previously unused or little used upland areas grows for intensification of livestock farming and alternative outlets for sewage sludge are sought encouraging its use as a fertiliser.

Prediction of bacterial water quality in rivers to assist in planning and policy decisions is now mandatory given an improved understanding of the health risks posed by faecal contamination, the likelihood of increasingly stringent water quality standards, tighter discharge consents and the pressure to develop land for agriculture, settlement or recreation. Such predictive capacity can only be achieved by continually improving our knowledge of the many complex processes that influence bacterial water quality. Only then can these relationships be quantified to form empirical and/or dynamic representations of processes in computer simulations capable of aiding the process of water resource management.

1.2 Pathogens and Indicator Bacteria.

"The significance of the presence of *E.coli* in aquatic environments is that the water in question has been contaminated with faecal material from warm-blooded animals" (Dufour, 1977).

Human and animal faecal materials contain a wide variety of pathogenic organisms, bacteria, parasites and viruses, such as *Salmonella*, *V.cholerae*, *Shigella*, *Poliovirases*, *Leptospira* and *Cryptosporidium*. These pathogens and other organisms are associated with a variety of illnesses ranging from gastro-enteritis, typhoid, cholera, bacterial dysentery,

food poisoning (Stainer et al., 1984) to skin complaints such as "swimmers-itch", ear, eye, nose and throat infections. Illnesses may be contracted as a result of direct contact during recreational water sports activities or of direct ingestion of organisms in contaminated drinking water.

Since pathogens are only present in small numbers it would be difficult and costly to test for them individually. The concept of indicator bacteria has been derived to give an indication of the degree of contamination by sewage or material of a faecal origin and so the likelihood of the presence of pathogenic organisms. In general the minimum requirement for an indicator species is that it must be a biotype that is prevalent in sewage and excreted only by humans or warm blooded animals . In addition it should be present in greater abundance than pathogenic organisms, more resistant to various disinfectants than pathogens, incapable (or at least not more capable than pathogens) of proliferation and quantifiable by simple and rapid laboratory procedures (Kott, 1977; Cabelli, 1978; Dutka, 1973).

The main indicators used are Total Coliforms (TC), Faecal Coliforms (FC), *E.coli* and Faecal Streptococci (FS). There is, however, no ideal indicator; "the best indicator is the one whose densities correlate best with the health hazards associated with a given (preferably several) type of pollution." (Cabelli, 1978) and there is much debate about the use of different indicators.

The coliform group was first chosen as an indicator of bacterial water quality, based on the work of Escherisch, who in 1885, identified *Bacillus Coli* as being characteristic of the faeces of warm-blooded animals (Dutka, 1973). The group comprises; *Escherischia, Klebsiella, Enterobacter,* and *Citrobacter* types, however, not all are associated with faecal contamination. The faecal coliform index was developed from an attempt to select only those types specifically of faecal origin; *Escherischia* and *Klebsiella.* Positive FC results, however, have been recovered from samples of waters with no evidence of faecal contamination, such as waters receiving industrial effluents containing high levels of carbohydrates. This is attributable to Klebsiella species, whilst 90 to 97% of all coliforms found in the faeces of humans and warm-blooded animals are *E.coli* (Dufour, 1977). *E.coli,* therefore, remains the most widely-used and accepted indicator for faecal pollution (Findlay et al., 1990).

Faecal Streptococci, on the other hand, have been found to have a better survival rate than FC and coliforms, and in certain cases give a better indication of probable virus content in lightly contaminated waters, often being the only indicator of contamination of a faecal nature at distant sampling sites. Thus, the absence of FC (or *E.coli*) may not necessarily mean the absence of pathogens, although the levels of pathogens present may not present a high risk of infection, ratios of viruses to bacteria, for example, being approximately $5:10^5$ for FC and $2.5:10^4$ for FS (Cohen and Shuval, 1973).

The ratio of FC to FS, is also a useful determinant and may provide information about the origin of a particular discharge. This ratio, in man, is generally greater than 4.0 (table 1.1) and less than 0.7 in other warm-blooded animals, see (Geldreich and Kenner, 1969). There are, of course, exceptions to this rule. In gull faeces the ratio of FC to FS are found to vary between 80 to 580 (Gould and Fletcher 1978). Environmental factors would, also, significantly change the populations with time and distance from the source or point of discharge, therefore, FC/FS ratio may only give reliable information relatively close to the source of the bacteria (Geldreich and Kenner, 1969).

Source of Faeces	Ratio FC/FS
Humans	4.4
Cat	0.3
Dog	0.02
Rodents	0.04
Cow	0.2
Pig	0.04
Sheep	0.4
Duck	0.6
Chicken	0.4
*Herring Gull	87
*Lesser Black-backed Gull	340
*Common Gull	585
*Black-headed Gull	135

Table 1.1: *Ratios of Faecal Coliforms to Faecal Streptococci in various warm-blooded animals (after Geldreich and Kenner, 1969; and *Gould and Fletcher, 1978).*

Other indicator organisms that are of interest for investigative purposes include *Clostridium perfringens* (CP), which can be used as an indicator of point source contamination in waters already affected by non-point source inputs. CP concentrations are lower in most warm blooded animals and the soils they graze than in man, and so elevated CP levels, in the presence of non-point source contamination, would indicate sewage pollution (Sorensen et al., 1989). *Rhodococcus coprophilus*, has been used to trace pollution by farm animals, because of its ability to survive longer than FS and FC, but is inadequate as an indicator of recent or current pollution (Oragui and Mara, 1983).

E.coli and FC, are often referred to as the thermotolerant coliforms, this is because the test the test for their presence is undertaken at a higher temperature than that for TC. The test for TC requires the bacteria to ferment lactose, with gas production within 48hrs at 35 degrees celsius. The test for FC requires the characteristics of TC and the ability to ferment lactose and produce gas at 44.5 degrees celsius. The test for *E.coli* has a longer incubation time and requires extra confirmative tests. FC, however, is 90-97% *E.coli*. FC therefore, is often taken to approximate to E.coli. The use of the term thermotolerant includes FC and *E.coli* and avoids the ambiguity which may surround the use of the two latter terms.

1.3 Sources and Inputs of Indicator Bacteria to catchments.

There are a wide variety of sources of indicator bacteria to catchments and surface waters inputs may be categorised as either point or non-point sources (table 1.2).

Source of Faecal contamination.	Nature of Input.	Typical values, all in FC.
1.Non-point sources.	Diffuse inputs to the catchment surface, transported to surface waters by hydrological processes.	
Natural Inputs	Faeces of indigenous fauna e.g. rodents (FC per gram)[Geldreich and Kenner, 1969]	2.10^6
Avian	Inputs from nesting or roosting bird colonies. (FC per gram faeces)[Standridge, 1979]	3.10^7-4.10^8
Enhanced	Agricultural; use of farm animal wastes as fertilizers on arable and pasture land, dirty water reuse schemes for irrigation/disposal, intensive stock grazing. Sewage sludge disposal; application to arable land and forestry as fertiliser. Recreational; directly from contact water sports, contamination of rivers draining catchments supporting enhanced recreational activity [Geldreich and Kenner, 1969].	10^9 per 100ml [Evans and Owens, 1972] 10^7 per g human faeces.
2.Point sources.	Localised, easily quantifiable sources, such as direct piped discharges to a water body.	
Sewage works	Constant piped discharges of treated domestic (examples, FC/100ml) and industrial wastes [Cohen and Shuval, 1973; Menon, 1985].	10-10^6 final 10^9 raw
Industrial effluents	Discharges from food processing industries; poultry/meat/potato plants, fruit/vegetable canneries etc [Menon, 1985].	10^2-10^7 per 100ml
Urban runoff, storm drainage	Combined sewer storm overflows[1], storm drains[2], contamination from urban runoff[3], street gutters[4]. Storm induced inputs may result in extremely high "first-flush" concentrations due to scour of material in pipes. Storm sewerage may be outdated and operate before sufficient dilution by receiving water can be achieved. (All values per 100ml) [[1&2]Burm and Vaughn, 1966 (in [3&4]Geldreich et al., 1968)]	[1]10^7 [2]10^6 [3]10^3-5.10^4 [4]10^2-10^4
Landfill sites	Evidence exists showing that leachates contain faecal indicators and that bacteria can survive within sites for long periods [Blannon and Peterson, 1974].	10^6 per 100ml (In waste 10^5-10^7 per g)
Land drains	Discharge of bacteria in leachate from agricultural land with artificial drainage [Evans and Owens, 1972].	10-10^5 per 100ml

Table 1.2: *Sources and inputs of faecal contamination to catchments, with typical indicator bacteria concentrations.*

Non-point source inputs are essentially derived from the catchment surface and represent all the inputs, perhaps along a river reach, which cannot be measured at an identifiable point. Such sources are related to landuse practices,

such as livestock type and density, natural fauna, and recreational use. The main landuse practices producing high concentrations of bacteria in drainage waters are intensive stocking and the application of a variety of solid, semi-solid and liquid wastes from both farms and other sources such as sludges from sewage treatment works and wastes from food processing industries.

Farm yard wastes are applied directly, while fresh, or after some treatment such as settlement. In this case the liquid might be used for crop irrigation and the solids spread and ploughed into arable land or applied to pasture land as a fertiliser or for soil conditioning to increase organic matter content. Sewage sludge is also applied to arable land as are food processing wastes. A recent development in land disposal of sewage sludge has been its application to forestry as a direct result of the ban on dumping at sea.

Food processing wastes may be seen as a less likely source of faecal contamination. Considering the nature of the materials processed and end products, however, it is not unreasonable to expect similar concentrations of bacteria in these wastes as in faecal materials. Bacterial levels in vegetable processing wastes, for example, are attributed to the soils in which they were grown, the use of animal fertilisers, farm animals and poor quality irrigation water. In meat processing wastes, straw, blood, flesh, fat, offal and manure from the intestines of the slaughtered animals all result in similar bacterial concentrations to the faeces of farm animals (Geldreich and Kenner, 1969).

Recreational use of surface waters is not only a reason for setting bacterial water quality standards but may also act as a source of contamination, particularly in reservoirs used for water sports or in rivers draining catchments which support a recreational activity. Such inputs, however, may not be as significant as those previously mentioned and their effects may be more localised (Hendry and Toth, 1981).

Point source inputs are localised sources which are easily identified and quantifiable, such as piped discharges or septic tanks. Piped discharges include leachate from land drainage systems, sewage works outfalls, combined sewer storm overflows, industrial discharges and contaminated soilwater from sub-standard septic tanks.

Certain sources of faecal contamination may not easily be categorised as either point or non-point. For example, nesting or roosting bird colonies. A permanent water fowl population on the shore of a lake was found to be responsible for outbreaks of various diseases on a nearby recreational beach (Standridge et al., 1979), and this clearly represents a point source. Gulls, however, represent both a potential non-point and point source of contamination. Gulls feed on agricultural land, at landfills and sewage works and breed on moorlands, often using reservoirs for safe night-roosts. One herring gull may, for example, excrete around 18.10^6 FC in a day, 19.10^6 is typical for a man (Gould and Fletcher, 1978). As there is little diurnal variation in faecal deposition rates of gulls, it is quite possible that large

numbers of gulls, could lead to a deterioration in the bacterial water quality of water bodies used for night-roosting, as well as, streams receiving run-off from nesting sites.

Landfill sites represent a point source. Both open and closed sanitary landfills can, if not correctly designed or operated, pose a threat to the bacterial quality of ground and surface waters, as both wastes and leachates contain high concentrations of indicator bacteria (table 1.2)(Donnelly et al., 1981;1982). Gulls colonise many open landfills and presumably contribute to the bacterial concentrations therein, as well as consuming bacteria from the waste itself. Fish farms have been considered as a point source of faecal contamination. In a survey of effluents and water quality from various trout farms, the majority of the coliforms of faecal origin were traced to run-off from bird droppings on surrounding land (Niemi, 1985). This might be expected, fish are not warm-blooded and do not have a permanent faecal coliform flora in their intestines, in remote regions their faeces rarely contain FC, positive occurrences are related to contamination from wildlife (Geldreich 1970).

Clearly inputs from bird colonies, food processing wastes and landfills do not represent a widespread source of faecal contamination in the U.K., although they may be responsible for localised contamination problems. Urban run-off may have the potential to produce rapid bacterial peaks as a result of heavy rainfall. The major point-sources of faecal bacteria must be from domestic and agricultural effluents which contribute a continual background load. The major non-point source is wash-out from upland catchments, although in lowland areas the importance of inputs from storage within river bed sediments should be stressed (Section 3).

1.4 Bacterial Water Quality Standards in the U.K.

The only bacterial water quality standards that apply in the U.K. are the EC directives on drinking waters, surface water abstraction sites and bathing waters, see table 1.3. These standards apply to drinking waters at the point of use and abstraction and to bathing water on coastal beaches.

There are, no generally applicable bacterial water quality standards for rivers and other inland waters in the U.K. Historically, water quality in the U.K. has been described by chemical criteria. The National Water Council (NWC) classification system, currently assigns a reach to a particular class on the basis of BOD, dissolved oxygen and ammonia. These chemical criteria cannot adequately describe the ability of a reach to support life, for example, a pulse of toxic material discharged into a river could wipe-out the aquatic fauna. A sample taken shortly afterwards may show normal chemical quality and would not show the condition of life in the river, which would take far longer to recover.

Bacterial Water Quality Standards		
E.C. Bathing Waters Directive	Total Coliforms per 100ml	*E.coli* per 100ml
*Guide Level (Recommended) 80% of samples not greater than	500	100
*Imperative (Mandatory) 95% of samples not greater than	10000	2000
*Revised (Imperative) 95% of samples not greater than	????	1000
EC Surface Water Abstraction Directive Guide Level	5000	2000

Table 1.3: *U.K. Bacterial Water Quality Standards *(after Kay et al., 1990).*

The NRA has recently published its proposals for a system of Statutory Water Quality Objectives, SWQO's, which will be the first step to setting standards for non-bathing waters. The new system will minimise subjectivity in classification by setting clearly quantifiable standards for both chemical and biological quality which will be unambiguous for enforcement, secure a consistent nationwide approach to water quality management and avoid the likelihood of mis-classification. The system will include bacterial water quality standards. General classification, use-related classes and standards will be determined for each reach. The standards will, in general, be based-on, the current water quality, level of compliance with EC directives, current and desired uses, benefits both in quality and financial, requirements and options for achieving the objectives, consequences and costs of those achievements, and whether

the objectives are realistic in terms of the time-scale for achievement. Following consideration of all these factors, cost/benefit analysis of the various options and consultation with interested/affected parties, the recommended SWQO will be submitted to the Secretary of State for the Environment to be written into the statute books. The SWQO will need to include all the agreed use-related Classes and the applicable EC Directives with dates for implementation and achievement. The proposals suggested that the general classification and use-related classes for rivers and canals could be developed by Spring 1992 and the schemes implemented Spring 1993.

Meeting the new standards within a given time-scale may require reassessment of water quality management plans and the application of water quality models (NRA, 1991). The control of diffuse or non-point sources of pollution has been identified as one of the major challenges to water quality managers. Point-sources such as sewage effluent discharges or slurry tank effluents are easily identified and measurable and examination of their impact may be relatively straight-forward. Non-point or diffuse sources of faecal bacteria, on the other hand, are geographically widespread and very variable in nature due to the source of contaminants. It would, therefore, be difficult to develop a reliable and repeatable method for quantifying such inputs. Clearly, for the new standards to be implemented successfully, an adequate understanding of the concentrations of contaminants and their interactions with their local environment, will be required.

2. Influences on *E.coli* survival in the environment.

The survival of *E.coli* in the environment is influenced by many factors. These range from physical factors, such as temperature, moisture and solar radiation, chemical factors such as pH and nutrient availability, and microbiological factors such as competition and predation. These factors act both directly and indirectly, and identification of causality is not a simple matter. For instance laboratory studies may constrain many parameters to examine the effect of one. This may result in unrepresentative results because complex interactions are neglected. On the other hand, in situ, there may be variation in too many parameters at a time allow a clear picture to be drawn. To add to the complication different results may be observed in different environments, these may in turn vary diurnally or on a seasonal basis.

There are three main stores, or reservoirs, for *E.coli* in the environment. The land store, the water column and sediments. Settlement of adsorbed bacteria causes an apparent die-off in the water column. This is of course part of the larger topic of transfer and transport of bacteria between stores and through the catchment. This section will examine the various influences on survival and attempt to highlight the key influences in each bacterial store.

Characterising Bacterial die-off.

To allow comparison of the results of bacterial survival studies, two main descriptors are used in the literature, these are, the die-off rate coefficient k and T_{90} the time taken for a population to fall to 90% of it's initial size. The die-off rate coefficient k is comes from first order decay dynamics as described by Chick's Law:

$$\frac{N_t}{N_0} = 10^{-kt}$$

where, N_0 = the initial population, and

N_t = the population at time $t > 0$

$$N_t = N_0 .10^{-kt}$$

for, $k = 0$ there is no die-off,

$k < 0$ indicates growth, i.e 10^{-kt} is positive, and

$k > 0$ and increasing, die-off is more rapid.

Simple exponential decay of the bacterial population once outside the growth environment is assumed. The die-off rate coefficient, k, is the amount the $\log_{10}$ population falls per unit time, ie:

$$-k.t = \log_{10}\left(\frac{N_t}{N_0}\right)$$

$$-k.t = \log_{10}N_0 - \log_{10}N_t$$

$$\therefore k = \frac{\log_{10}N_0 - \log_{10}N_t}{t}$$

where, $t = t_0 - t_1$.

k can therefore be found graphically by plotting the $\log_{10}$ bacterial population against time and extracting from the y axis the two intercepts from the best straight line fit (figure 2.1), for one hour on the x axis. T_{90} is the time taken for the population to fall by one $\log_{10}$ cycle from the initial value. In practical applications there may be a time delay before a decline in the bacterial population occurs, in such situations k is the slope of the die-off portion of the graph, and T_{90} remains the total time from the beginning of an experiment to when the population has fallen by 90%.

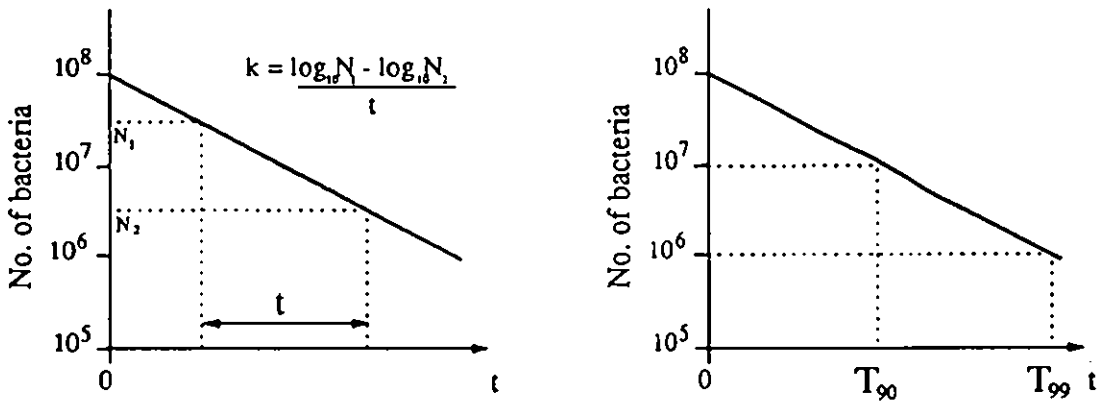

Fig. 2.1: *Graphical derivation of die-off rate k and 90% reduction time T_{90} assuming simple first order decay dynamics.*

2.1 Chemical and Microbiological influences on *E.coli* survival.

The Influence of Natural Microbial Communities on *E.coli* Survival

The balance of microbial communities is maintained by a complex array of inter-relationships between populations within the community. One of the best known consequences of these interactions is the elimination of alien populations (Verstraete and Voets, 1972). *E.coli* is not indigenous to the natural waters and is thus treated as alien or an *alien-prototype*.

A number of studies have attempted to isolate the sections of the natural microbial community which cause the most rapid die-off of *E.coli*. In tests on clean and polluted lake waters in Belgium samples were successively filtered and inoculated with *E.coli* cultured from natural strains and added to a nutrient preparation, the aim being to examine the effect of removing sections of the microbial community. The first step involved filtering-out algae and protozoa (5µm membrane) at the second step, bacteria (0.45µm), and finally remove bacteriophages and destroy all anti-microbial substances by autoclaving at 120°C for 10 minutes (figure 2.2). A bacteriophage is a virus whose host is bacteria. *E.coli* is known to be the host to a number bacteriophages (Singleton and Sainsbury, 1981). The anti-microbial substances, were referred to as *thermolabile toxins*, i.e. unstable, heat sensitive substances, which may have been excreted by other groups of the microbial community, such as algae. These substances were shown to be largely non-diffusable, and it was suggested that they could be a coliphage, a *Bdellovibrio* or a high molecular antibiotic.

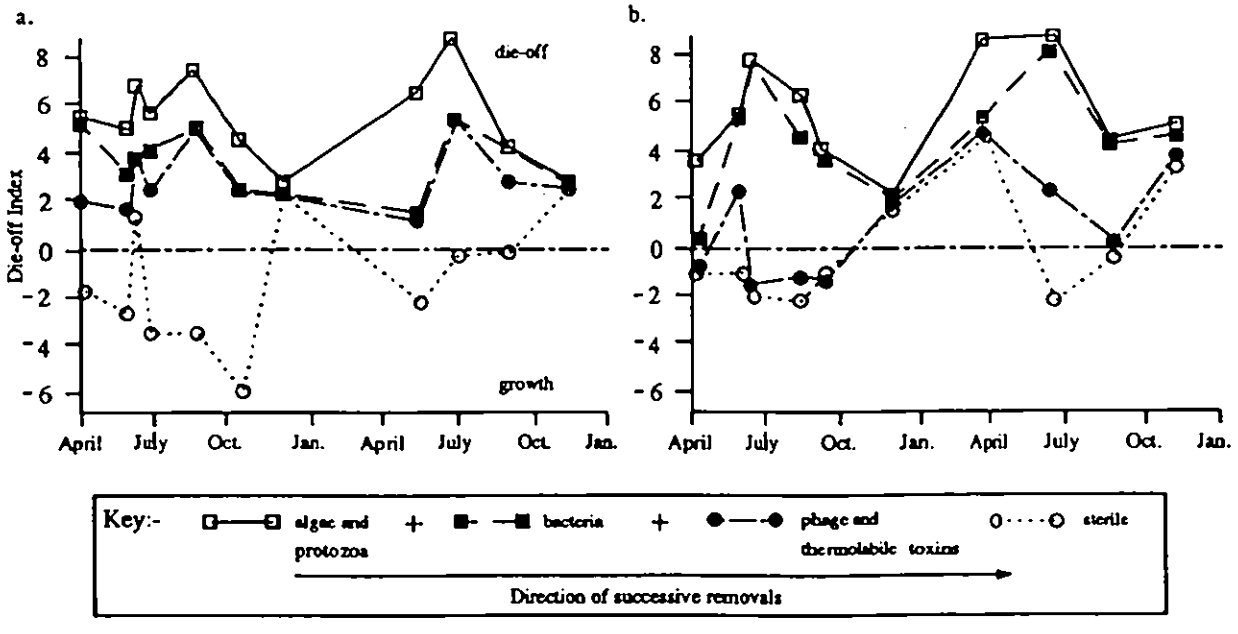

Fig. 2.2: *Die-off of E.coli in sewage contaminated (Coupure) and clean (Watersportban) water bodies, following removal of: protozoa and algae (dashed line); protozoa, algae and bacteria (dot-dash); filtration at 0.45µm and autoclaving (dotted line); raw samples (continuous). After Verstraete and Voets (1972).*

Further filtration (at 0.22μm), however showed that they were not *Bdellovibrios*, as these organisms measure at least 0.3- 0.4μm. A dilution experiment was finally used to show that both coliphages and a "potent" colicidal toxin were present in the water, however it was suggested that further research would be necessary to determine the exact nature and origin of these toxins. In general, successive reductions in die-off rate occurred with each removal, however there were marked differences between the clean and polluted waters. In the polluted water the greatest effect was noted by removal of bacteriophages and thermolabile toxins, and in the clean water indigenous bacteria had the greatest effect on *E.coli* survival. The ability of the microbial community in the clean water to reduce the *E.coli* population was greater than in the polluted water (Verstraete and Voets, 1972).

A more recent study discusses the role of *ultramicrobacteria*, which play a similar role in *E.coli* die-off to thermo-labile toxins. Samples typical of a clean west-midlands river and one polluted with organic effluent were collected above and below the outfall of a sewage works. These were successively filtered to remove fractions of the naturally occurring microbial community, firstly through Whatman No.1 papers, then 0.45μm Millipore membrane filters and finally sterilised by autoclaving. The filtrates and raw samples were then used in die-off experiments, results were expressed as T_{90} (figure 2.1) and die-off coefficient *k*. Die-off was termed *disappearance* because there was no evidence that *E.coli* actually died. Little difference was observed in die-off between the raw and Whatman filtered samples which were free of protozoa and suspended material, suggesting the minimal importance of these factors (figure 2.3). In the clean samples the removal of the majority of competing bacteria by filtration at 0.45μm had the greatest effect on survival. In the sewage contaminated sample, the removal of bacteria did not have as great an effect as the removal of phage and ultramicrobacteria, which were found on microscopic examination to be very motile (Flint, 1987). These results are very similar to those of Verstraete and Voets (1972) and suggest the dominance different microbial purification processes under different environmental conditions, namely that in clean freshwaters competition with indigenous bacteria is the dominant microbial factor whereas in sewage contaminated waters, phages, ultramicrobacteria and thermolabile toxins (which may be related to ultramicrobacteria and not known at the time of Verstraete and Voets (1972)) dominate *E.coli* die-off. The difference in the two waters may be the abundance of nutrients allowing *E.coli* survival without competition.

A variety of die-off data are reviewed and reported by Enzinger and Cooper (1976). *Bdellovibrio bacteriovorous*, a parasitic bacterium of other bacteria such as *E.coli*, has been associated with increased die-off of *E.coli* in natural waters. Coliphages have been found in sewage contaminated sea-water, however, this does not imply their activity in removal of coliforms. Phages have been shown to be ineffective in reducing *E.coli*, because they require an actively growing host and *E.coli* cannot usually meet this requirement once released into the hostile natural environment. Protozoa have been shown to reduce *E.coli* survival in sewage, mainly due to motile strains or *ciliates*. In a further series of experiments, with estuarine water, Enzinger and Cooper (1976) tested the effect of protozoa and bacteria

removal on *E.coli* die-off. In separate tests, protozoa were removed from samples by membrane filtration, and indigenous bacteria were removed by anti-biotics, anti-biotic resistant *E.coli* strains were therefore used in the bacteria removal tests. *E.coli* die-off was most rapid in the presence of the complete indigenous population and related to the development of predator populations. Die-off in the samples free of bacteria was as fast as with raw water. In the absence of protozoa die-off was much slower (figure 2.3).

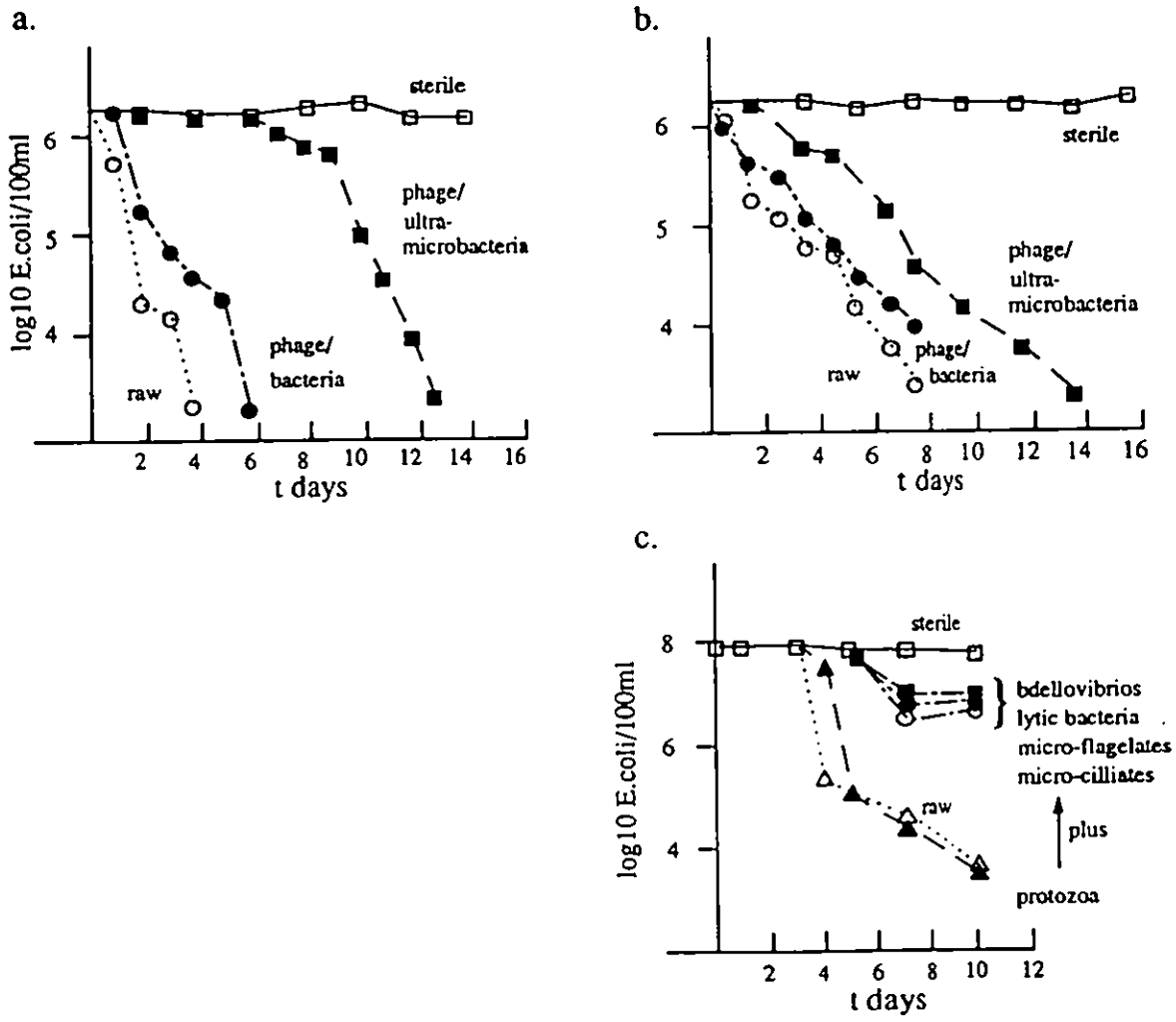

Fig. 2.3: *The influence of successive removal of sections of natural microbial communities on E.coli die-off in a. estuarine water (after Enzinger and Cooper 1976), b. "clean" river water, and c. sewage contaminated river water (after Flint 1987).*

A range of pore sizes were used in the filtration experiments, 0.22µm filtrates were generally free of natural organisms and *E.coli* numbers in these samples remained near constant. Only bdellovibrios were present in the 0.45µm filtrates, these caused a slight increase in die-off rate. Results for 0.8 and 1.2µm filtrates, were similar. These contained *Bdellovibrio* plus one or two other bacteria, capable of *E.coli* destruction. The 1.2 and 3.0µm filtrates both contained

numerous *micro*-ciliates and *micro*-flagellates. Only the 3µm filtrates contained protozoa and die-off was most rapid in these. The lag-time before the onset of die-off in these experiments, was probably due to the time taken for the protozoan population to reach sufficient density to effect a detectable removal of *E.coli*. Bacterial predators were not found to be a significant cause of die-off, protozoa exerted the greatest influence on survival, and have also been found responsible for bacterial decline in soils. Protozoan predation may also exert pressure on bacterial populations in other environments (Enzinger and Cooper, 1976).

Zerfas (1970), tested river water and found that removal of protozoa (by heating river water at 45°C for 1 hour) did not affect *E.coli* die-off. Raw, 0.45µm filtered, and heat treated water were tested. It is likely that the 0.45µm filtrate was free from protozoa, but it may have contained *Bdellovibrio* (Enzinger and Cooper, 1976), although, these only exert a small effect on die-off. In all samples growth occurred in the first two days (figure 2.4).

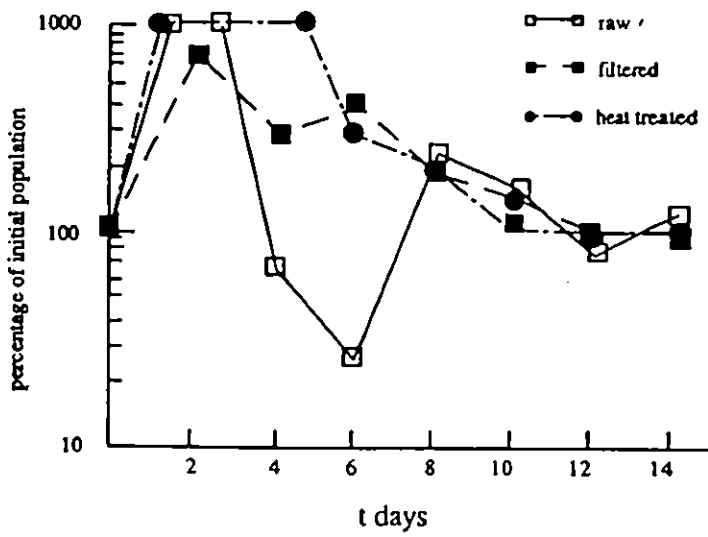

Fig. 2.4: *E.coli die-off in samples with indigenous biota present (raw), protozoa removed (filtered), and sterilised (heated) (after Zerfas 1970).*

Populations grew to well in excess of 5 times their initial sizes. The raw water samples undergoing the most marked die-off, as might be expected, falling to a fifth of the initial population at day 6. The filtered sample is unusual in that the peak *E.coli* population is small in comparison to the other samples. The population dies-off gradually towards day 8. In the heated sample, in which protozoa and other heat sensitive organisms are absent, there was rapid growth, the population obviously reaching a very large peak, and then falling to near stability after 8 days. Nutrients may have

been abundant in this sample, which was not filtered, and the heating may have released nutrients bound-up in organic matter. Net die-off probably resulted from nutrient depletion, followed by adjustment to suit nutrient availability in the new environment. In the raw water rapid die-off followed the initial growth period, possibly due to protozoan predation. The initial growth period resulting from the delayed development of the protozoan population (Enzinger and Cooper 1976). Following the die-off phase, the predator population probably suffered die-back, allowing recovery of the *E.coli* population, the two populations eventually reaching some balance. It is also possible that these results are an artefact of the sampling frequency, which could have been higher.

Clearly natural microbial communities can exert considerable stress on *E.coli* populations. In particular, protozoa, bacteriophages, ultramicrobacteria and thermolabile toxins, and the relative importance of the different groups of organisms may depend on the local environment. It would appear that in clean freshwaters the key microbial factor influencing *E.coli* survival is competition with bacteria, and in sewage contaminated waters, phages, ultramicrobacteria and heat sensitive toxins, whereas in estuarine waters protozoa have been shown to dominate. It is likely that similar behaviour occurs in soils and sediments, although these areas are not well documented.

The effect of pH

Low pH (3-4), generally has a detrimental effect on bacterial survival in soils and water (Yates and Yates, 1988). In-situ tests of FC survival in acid mine waters (pH at around 3) have shown very rapid die-off (2 hours for reductions from 10^6 to 100 FC.100ml^{-1}) compared to neutral waters (Carlson-Gunnoe et al., 1983). In dialysis chamber tests conducted at 10°C, *E.coli* die-off was slowest between pH 5.5 and 7.5 (figure 2.5) and die-off rate increased sharply either side of these values (McFeters and Stuart, 1972). In another study pH 5-6.4 promoted optimum survival (Cohen, 1922). In acid peat soils (pH 2.9-4.5) die-off occurs within a few days, whereas survival in limestone soil was upto several weeks. It is suggested that low pH not only affects *E.coli* survival ability but, also nutrient availability and antimicrobial agents (Cuthbert et al., 1955). High pH in eutrophic northern brackish waters is reported to cause a significant increase in die-off rate (Hirn et al., 1979). These results support the findings of McFeters and Stuart (1972).

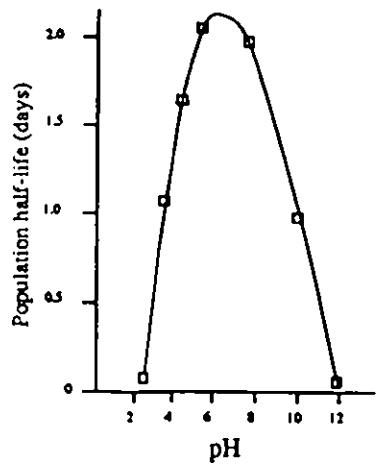

Fig. 2.5: *Effect of pH on E.coli die-off (after McFeters and Stuart, 1972).*

Some microorganisms have been found to absorb H^+ to such an extent that Na^+, K^+, and Ca^{2+}, within the cells, cannot be replaced. This inhibition of cation exchange causes the cell membrane to lose it's protective function by leaking K^+ and other compounds essential to the

normal functioning of the cell. The binding of heavy metals to the cell membrane may also cause death or injury by inhibiting the transfer of various metabolites in and out of a cell (McCalla, 1964; Singleton and Sainsbury, 1981).

Self-protection

This phenomenon has been observed in estuarine waters. This is the aggregation of bacteria, in the absence of sediment particles, to form an inner core protected from osmotic stress caused by saline waters. The outer cells being loosely attached by polymers or cations (Findlay et al., 1990). It is not known whether this effect plays a role in bacterial survival in fresh waters.

Nutrients

Nutrient levels and availability affect *E.coli* survival both directly and indirectly. Directly they affect bacterial metabolism, and indirectly, both the ability of the naturally occurring microbial community to compete and prey, and of bacteria to adsorb to particulates (Milne et al., 1991), which itself protects bacteria.

E.coli levels have been found to be correlated with phosphorous, nitrogen, and organic nutrients (Hirn et al., 1980), and enhanced survival has been observed in high conductivity water (McFeters and Stuart, 1972). In the absence of the natural microbial community, growth of enteric bacteria has been observed at temperatures above 10°C in low nutrient mountain stream water, and more rapidly in nutrient extracts from the stream bottom sediments. It was suggested that extensive growth may occur in bottom sediments where nutrients can be in high concentrations relative to free flowing water, in an environment less sensitive to diurnal effects than the overlying water body (Hendricks and Morrison, 1967). This may the case in the absence of the natural microbial community, however growth only occurs occasionally in raw samples, nevertheless the bottom sediment do offer conditions more favourable to survival (Verstraete and Voets, 1972).

Stephenson and Rychert (1982), suggested that organic matter (O.M) content many have a critical influence on the survival and/or multiplication of the bacteria in sediments. Burton et al. (1987), however, could not correlate O.M to *E.coli* survival in sediments, suggesting that this was due to the variable nature of organic matter and the multitude of other influences on survival.

Tests with sterile seawater showed that both nutrient supply and suspended solids enhance *E.coli* survival. These effects were found to be mutually exclusive. Maximum die-off occurred at 20°C and suspended solids (S.S) concentration of 12.5mg.l^{-1}. Nutrients were preferentially adsorbed to the particles, lowering nutrient availability and

the protection afforded to the bacteria by adsorption. The same nutrient concentration was used in all of these experiments. At increased suspended solids concentrations the die-off rate fell, presumably due to greater numbers of adsorption sites. Die-off was also less rapid at lower S.S concentrations, probably due to greater nutrient availability. At 5°C the effect of S.S addition was more marked. Die-off peaked at 5mg.l^{-1}, suggested a reduction in the ability of the bacteria to uptake nutrients at the lower temperature (Milne et al., 1991).

In tests using sterile samples, *E.coli* survival is enhanced with increasing nutrient supply and growth may even be possible. In raw waters growth is not often possible due to the influence of the natural microbial community but survival is enhanced in waters polluted with organic wastes compared to clean waters.

All of these results suggest that, ideally, the examination of influences on the survival of indicator bacteria should be undertaken in the presence of the natural microbial community, using samples representative of the water body under consideration.

2.2 Physical influences on *E.coli* survival.

As with microbiological and chemical influences on *E.coli* survival, physical influences act both directly and indirectly. Directly, higher temperatures may enhance bacterial metabolism or increase stress, solar radiation may damage organisms affecting the ability to reproduce, whilst in soils, lack of moisture may cause death due to desiccation. Indirectly, higher temperatures may enhance the antagonistic effect of the naturally occurring micro-community, reduce dissolved oxygen concentration, or enhance the drying out of soils.

Moisture

In soils, and on the catchment surface, moisture is perhaps the most important factor in determining bacterial survival (Van Donsel et al., 1967). Moist land areas contribute significantly greater numbers of faecal bacteria to surface waters than dryer areas, due to extended survival (Hunter and McDonald, 1991: Bagdasaryan, 1964), see figure 2.6.

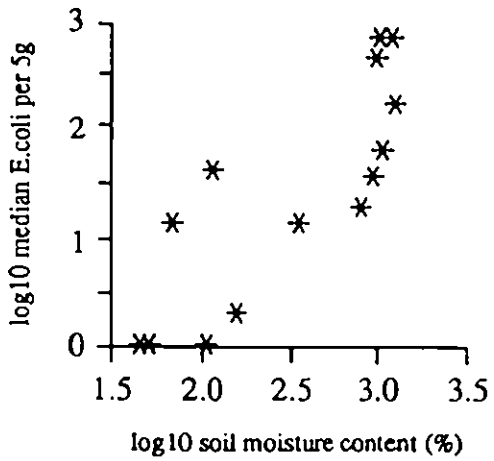

Fig. 2.6: *Scatter plot of log_{10} median E.coli count (per 5g) against log_{10} soil moisture content (% of dry weight) for a catchment in the Yorkshire Dales (after Hunter and McDonald, 1991).*

Bacterial survival has also been found to be greatly extended during periods of water-logging (Chandler et al., 1981). In one study, *E.coli* could be isolated from moist soil for 5 years after inoculation, and 10-40% saturation was found to be optimum for survival (Young and Greenfield, 1923). Moisture content and retention properties of the soil, are in-turn influenced by other factors which may indirectly effect bacterial survival, for instance, soil type and vegetation cover (Beard, 1940; Hunter and McDonald, 1991). Vegetation type is indicative of long-term soil moisture regime and could be used as a marker for zonation of a catchment into active and passive areas. Active zones being boggy/moist

areas where the bacteria survive for longest and which are active in run-off generation. Reductions in bacterial inputs to surface waters could be achieved by preventing livestock access to these active zones (Hunter and McDonald, 1991). Vegetation type and cover may also influence soil moisture conditions. Dense sward can afford protection from adverse environmental conditions, limiting the direct and drying effect of sunlight, wind and high temperatures (Chandler et al., 1981; Zyman and Sorber, 1988). Waste application may also influence moisture retention. Liquid animal wastes have high moisture content and good moisture retention properties (Crane and Moore, 1986). T_{90} values of 3 days and 25 days have been observed in sandy and loam soils, respectively. These results are attributed to the higher O.M content and better moisture retention of the loam soil (Van Donsel et al., 1967).

In another study, soil type was not found to be important in determining FC survival. The study lasted for three years and examined the persistence of FC in soils varying from clay to loam, in areas with rainfalls from 500 to 1000mm. Test plots were irrigated with piggery waste and subsoil profiles were taken at weekly intervals after application until indicator bacteria could no longer be detected. Further applications were then made. The average FC concentration of the applied slurry was $3.10^6.100ml^{-1}$ with mean dry matter content of 1.9%, the application rate approximated to 30mm depth. Overall T_{90} was 9 days for pastures and 11 days in top soil, these values were not significantly different at higher or lower waste application rates nor was die-off significantly different 6 weeks after application. FC concentration of the slurry was found to have a greater effect on soil concentrations than application rate. Of the 12 soil types tested, only one differed significantly (Chandler et al., 1981). The results suggest that such wastes not only supply vast numbers of bacteria, but, by maintaining moisture levels, provide conditions suitable for extended survival, and may, in time, increase soil organic matter content.

Soil/sediment Type

The moisture retention characteristics of different soil types is probably the most important characteristic in determining *E.coli* survival. Die-off in stream-bed sediments, however, will be determined by other factors unless periodic drying-out, resulting perhaps from over abstraction, occurs. Different sediment types may have differing organic matter content, nutrients and adsorption capacities. A study of *E.coli* survival in different freshwater sediments, under continuous flow laboratory microcosm conditions, has shown that the only sediment characteristic for which there was an apparent relationship with die-off was particle size. *E.coli* survived longer in sediments with at least 25% clay content (Burton et al.,1987), see table 2.1. These tests used reconstituted water and soil/sediment samples stored at 4°C for up to two weeks.

Clays and humic substances enhance bacterial survival by reducing the effect of solar radiation. Clay minerals have internal charge surfaces and hence very high surface area to volume ratio, resulting in high adsorption capacity. As

particle size increases, adsorption capacity falls and with it the level of protection offered to bacteria (Mitchell and Chamberlain, 1978).

Clay/silt/sand ratio	% Organic matter content	Total Kjeldahl Nitrogen (ppm)	Total Phosphoroûs (ppm)	*E.coli* survival rate k(hr^{-1}), *(1 · die-off rate)*
75 : 25 : 0	14.8	3.18	9.98	0.874
28 : 55 : 12	6.2	16.32	6.90	0.764
25 : 51 : 24	5.2	13.97	14.30	0.833
12 : 76 : 11	9.0	24.10	7.70	0.670
2 : 0 : 98	0.7			0.681

Table. 2.1: *Bacterial survival in different sediments (Burton et al., 1987).*

Dissolved Oxygen

Low dissolved oxygen levels favour *E.coli* survival (figure 2.7) and in well aerated water die-off is enhanced (Zerfas, 1970). The oxygen level may affect the ability of the indigenous microbial population to purify the water. Die-off being positively correlated with D.O content in sewage contaminated waters (Verstraete and Voets, 1972). Periods of ice cover have been shown to lead to enhanced bacterial survival as a result of oxygen depletion (Hirn et al. 1979; Gordon, 1972; Davenport et al., 1976).

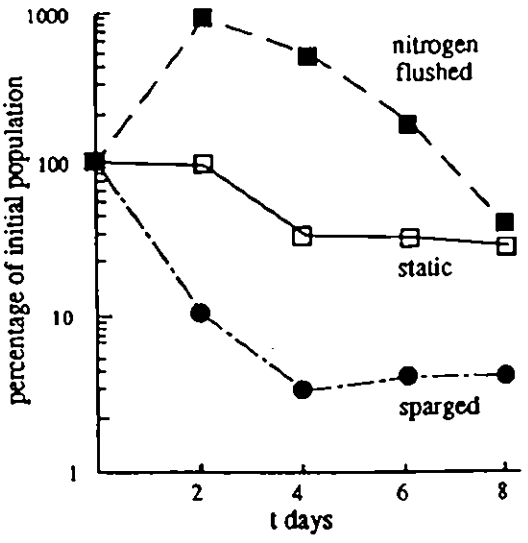

Fig. 2.7: *Survival of FC in raw river water at 20°C in air sparged, static, and N_2 flushed flasks (after Zerfas, 1970).*

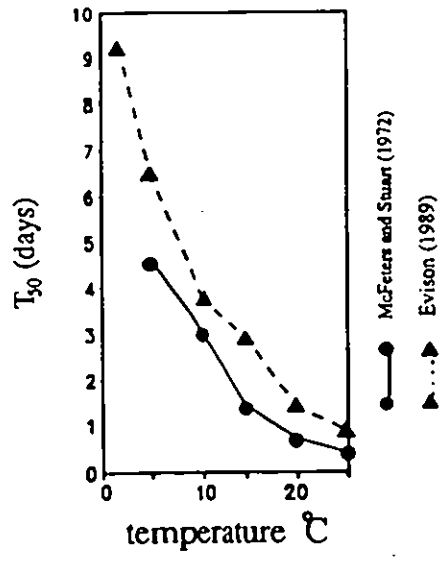

Fig. 2.8: *The effect of temperature on E.coli survival (after McFeters and Stuart (1972) and Evison (1989)).*

Temperature

Lower temperatures generally favour longer survival of *E.coli*. In eutrophic northern brackish water and in freshwater lakes, increased temperature causes greater predatory activity, enhancing *E.coli* die-off (Hirn et al., 1979; Verstraete and Voets, 1972).

Laboratory experiments using dialysis chambers inoculated with naturally occurring *E.coli* and using filtered stream water, tested the change in *E.coli* population half-life over a range of temperatures. The results showed bacterial die-off to be inversely proportional to temperature, from 5°C to 15°C (figure 2.8), above 15°C the effect of temperature increase was less marked than below(McFeters and Stuart, 1972). Kunkle and Meiman (1968) also found that water temperature was inversely related to bacterial counts, but could not separate temperature and insolation effects. Figure 2.8 also shows the results of Evison (1989), who used water from the River Coquet near the inlet to the Warkworth water treatment plant, a site free from industrial pollution with only slight sewage contamination. Samples were filter sterilised at 0.2μm, raw sewage was used as a source of *E.coli* and the effect of temperature was examined in the dark with samples inoculated with 0.25% raw sewage. The results show T_{50}, converted from the original T_{90} values assuming simple first order die-off. The results are very similar to those of McFeters and Stuart (1972) and Flint (1987) (not shown here). The slightly extended survival at each temperature may have been due to the experimental environment or the assumptions made in converting the data. McFeters and Stuart did not indicate the lighting conditions during their experiments and it may be possible that light reduced survival times.

In one mountain stream, increased FC concentrations were associated with higher temperatures (McSwain and Swank, 1977). Nutrient levels in this stream were very low, thus the natural microbial activity may have been insignificant. The metabolic benefits of increased temperature, therefore, out-weigh any detrimental effects resulting from enhanced activity of the natural biota.

Extremes of temperature, may promote rapid die-off. Elevated temperatures, which have already been demonstrated to enhance die-off, combined with drying conditions, are particularly detrimental as are freeze/thaw cycles (Crane and Moore, 1986).

In most U.K streams and rivers the effects of the natural microbial community will mean that at higher temperatures die-off of *E.coli* will be enhanced. Although complicating factors such as flow regime may affect bacterial input rates and can mean that such patterns are not immediately apparent.

Seasonal Behaviour

Interesting seasonal trends are observed in bacterial concentrations in different climatic areas. In northern brackish waters off Finland, temperatures range from 20°C in the summer with ice formation in the winter between January and April. Highest bacterial levels are observed during these periods of ice cover (figure 2.8) and in the autumn, with maximum survival at around 0°C (Hirn et al., 1979). Low temperature and dissolved oxygen tending to favour extended survival (Hirn et al., 1979; Gordon, 1972; Davenport et al., 1976).

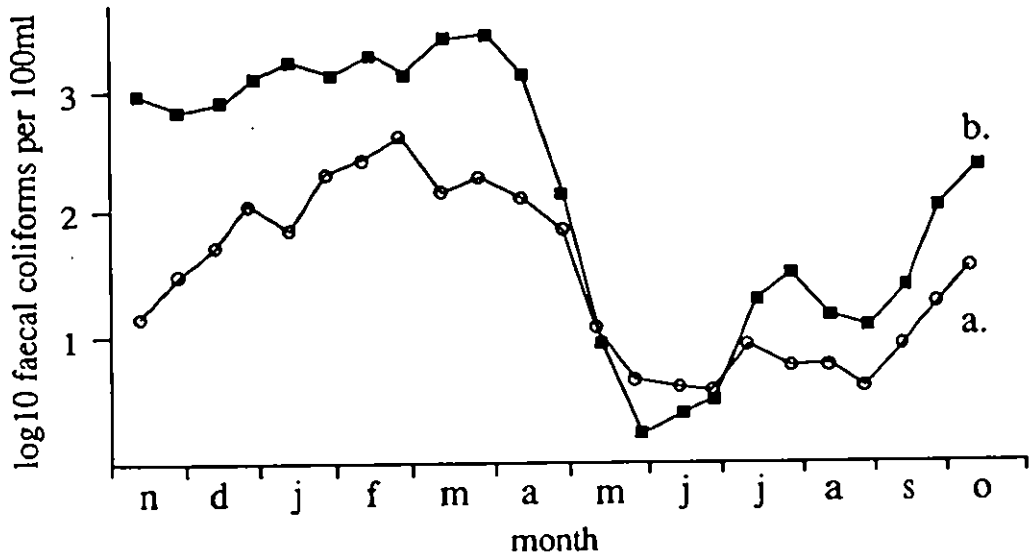

Fig. 2.8: *Seasonal variation of faecal coliform concentrations in a. a eutrophic bay (average depth 7m) and b. a very eutrophic bay (average depth 3m) west of Helsinki, Finland (after Hirn et al., 1979).*

In warmer climates, seasonal trends may be similar, but for different reasons. In Israel, survival was also found to be better in winter, this was probably due to stress induced in the bacterial population at the higher temperatures during the summer (Cohen and Shuval, 1973) although enhanced microbial activity may also have been significant (Verstraete and Voets, 1972; Zanoni et al., 1978).

Stream flow is an important factor when considering seasonal variations in bacterial concentrations. Hydrological conditions will influence bacterial input rates and transport within the channel and different dilution conditions may mask the true seasonal survival patterns. This may merely indicate the importance of the influence of transport mechanisms over die-off characteristics in determining stream bacterial concentrations.

Solar radiation

Solar radiation has a direct lethal effect on *E.coli*. Indirectly, it may raise temperatures, and in water increase photosynthetic activity in water thereby increasing dissolved oxygen supply and perhaps concentrations of toxins excreted by certain algae (Verstraete and Voets, 1972). It has also been shown to cause significant reductions of bacterial numbers on vegetation sprayed with liquid wastes (Crane and Moore, 1986). The ultra-violet component of sunlight has often been thought of as the main cause of bacterial die-off. Visible and near-visible light, however, also have a lethal effect on bacteria (Mitchell and Chamberlain, 1978; Fujioka and Siwak, 1985). Fujioka and Siwak, found that die-off was enhanced in samples stored in glass and polyethylene exposed to sunlight compared to those stored in the dark, it is not certain, however, whether these samples were sterile or not. Figure 2.9 shows T_{90} values for various intensities of light from a solar simulator, sample temperature was maintained at 15°C (Evison, 1989). Fujioka and Siwak tested FC die-off under typically sunny Hawaiian conditions, temperatures varying between 15-25°C. Observed T_{90} was between 1-2 hours in the light and a number of days in darkness. Similar results have been attained with containers suspended a stream at shaded and exposed sites, die-off occurring in only 2 hours in the exposed containers (Kunkle and Meiman, 1968).

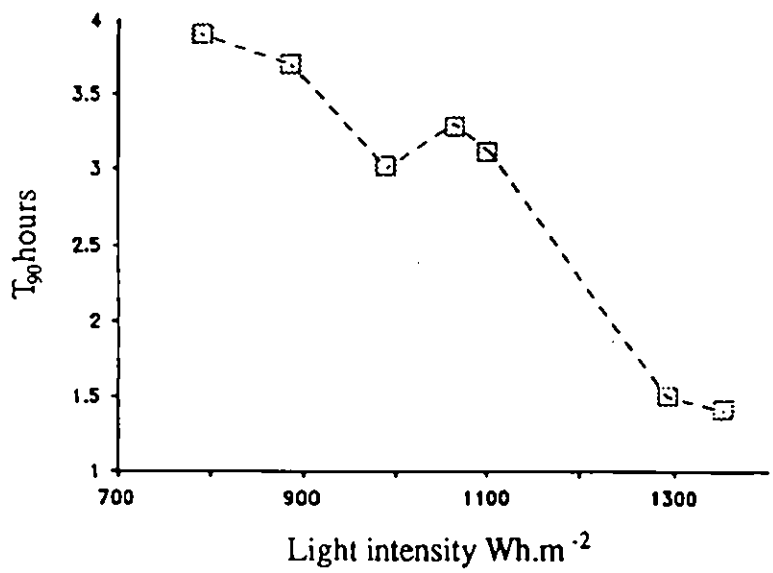

Fig. 2.9: *T_{90} for E.coli for a range of light intensities from a "solar simulator" (after Evison 1989).*

The penetration of U.V in water is relatively low, and will be effected by the clarity of the water. Waste pond studies have demonstrated how increased depth reduces coliform die-off rate, i.e the depth averaged die-off rate constant is inversely proportional to pond depth (Sariyaka et al., 1987). Verstraete and Voets (1972), showed that bacterial die-off due to solar radiation was greater in clean lake water, than in heavily sewage contaminated water. In both of these

cases solar radiation accounted for 40 to 50% of *E.coli* variation. Suspended matter such as clays and humic substances reduce the penetration of light, and may also afford protection by adsorption. (Mitchell and Chamberlain, 1978).

The effect of solar radiation will be dependant on the initial quality of the stream or water body receiving the bacterial load. In relatively clean, shallow waters, good light penetration will be possible leading to optimal die-off conditions; that is, the direct lethal effect of sun-light, adequate oxygen supply to the indigenous microfauna, and production of toxins by algae. Conversely, in poorer quality or deeper waters, such as those draining many of the U.K's urbanised and industrial areas, the benefits of sun-light may be minimal, due to poor penetration, protection by adsorption to suspended and deposited matter, and stressed micro-community.

2.3 Summary of the key influences on *E.coli* survival.

1. Moisture is the key influence on *E.coli* survival on the land surface and in soils. This overrides all other factors including temperature and solar radiation, although moisture content is influenced by these.

2. Vegetation type may be useful to indicate long-term moisture regime.

3. Temperature extremes are detrimental to bacterial survival, high summer temperatures combined with sunlight will enhance die-off considerably, as will frosts, although the lower winter temperatures and increased soil moisture will generally favour extended survival.

4. The effect of solar radiation will be dependant on the vegetation cover and to some degree by the aspect of the catchment surface.

5. In the soil matrix conditions may be much more favourable to bacterial survival; solar radiation will be excluded from all but the top few centimetres of soil, temperature variations will be increasingly damped with depth, and moisture levels will fluctuate less. The effects of the natural soil microcommunity may then be the most significant influence on survival, as a result of competition for nutrients, and predation by protozoa, coliphages and lytic bacteria.

6. Sedimentation of bacteria adsorbed to suspended matter is the primary means of removal of indicator bacteria from the water column, this mechanism has in the past been misconstrued as die-off in situ.

7. Suspended matter only influences survival in so much as adsorption affords protection to bacteria from the harmful effects of solar radiation and offers a transport mechanism to the more stable bed sediment environment.

8. In the water column, depth, clarity, and mixing will influence the effect of solar radiation.

9. Water quality will affect the ability of the indigenous microbial community to cleanse the water.

10. Sediment deposits are generally nutrient rich and favour bacterial survival.

11. Predation and competition for nutrients may be the greatest factors reducing bacterial survival in sediments.

die-off rate in sediments may be expected, these will most likely be in response to temperature, however, no observations have been made to this effect.

13. In eutrophic waters, such as lowland rivers, oxygen depletion may occur in the river bed sediments during the summer. Under these conditions low pH can develop leading to rapid die-off. A table of die-off rates for various enteric bacteria is given in appendix I.

3. Storage and Transport of bacteria in the environment.

Sections 1 and 2 of this review dealt with sources of faecal contamination and the influences on *E.coli* survival in catchments and surface waters, this section examines how the bacteria are stored within and transported through a catchment, from deposition on the land-surface to transport in the riverine environment.

On the land-surface, bacteria may remain locked-up in parent faecal material. Once released they can become adsorbed to soil and organic matter, and survive for long enough to provide a semi-permanent reservoir capable of contaminating the surrounding aquatic environment following transport by hydrological processes (Hunter and McDonald, 1991). Indeed, stored faecal bacteria have been shown to be released at least 4 months after being deposited (Evans and Owens, 1972). In lowland catchments, transport by water infiltrating the soil mass probably accounts for majority of bacteria reaching surface waters from non-point sources, although these may be insignificant compared to inputs from point sources or inputs from bacteria stored in the stream bed. In upland areas bacteria may be transported by surface runoff due to infiltration or saturation excess, as well as matrix and non-matrix soil through-flow, during baseflow conditions inputs may be provided by return flows and matrix through-flow. The relative importance of these pathways will, however, depend on the nature of the catchment, the antecedent soil moisture status and the rainfall duration and intensity. Bacteria may enter a water-body adsorbed to solids or free in solution. The behaviour of mineral particulates is determined by the flow characteristics and adsorbed bacteria may, for instance, be transported in scour/deposition cycles. Low density particulates such as organic solids may, however, remain in suspension at much lower flow velocities and bacteria adsorbed to these may be transported over large distances. This behaviour will depend on the supply of particulates and vary between upland streams and slow flowing lowland rivers.

3.1 The Adsorption of Bacteria.

Adsorption Theory.

The process of adsorption is very important in determining storage and transport of bacteria in a catchment. Bacteria, including *E.coli*, can for example, become adsorbed to sediment particles, rocks, plant and animal surfaces, and organic matter, at so called liquid/solid *interfaces* (Marshall, 1979). The solid surfaces at interfaces, for example a mineral sediment grain, have net negative charge, and as a result attract cations. These *counterions* effectively cancel-out the negative surface charges by occupying the charge sites. A double layer of ions forms at the interface, the surface then represents a plane of strongly adsorbed cations a few nanometres thick, known as the Stern-layer.

Beyond this layer the electrostatic forces decay almost exponentially, as does the difference in numbers of cations to anions. This zone is termed the diffuse layer (Marshall, 1979; White, 1979). Van Der Waals forces also act at interfaces and have a weak attractive effect which acts independently from electrostatic forces. Micro-organisms are themselves negatively charged and are repelled by the charge at an interface. The Van Der Waals forces, however, tend to attract them to the particles (figure 3.1.a).

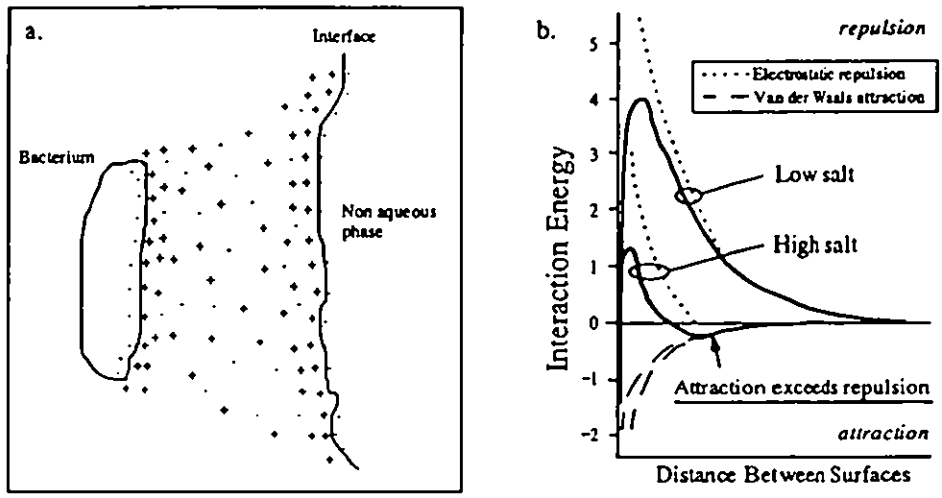

Fig. 3.1: *a. Schematic representation of the interaction between potentially overlapping cation clouds accompanying a negatively-charged bacterium as it approaches a negatively-charged interface, b. idealised curves showing the potential energy of interaction as a function of distance between a bacterium and an interface in solutions of different salt concentration (after Marshall 1979).*

In waters with very low salt concentrations the electrostatic repulsion exceeds the Van der Waals forces at all but the very closest distances from the interface. For adsorption to occur the microbe must actually make contact or collide with the surface (Figure 3.1.b). In most fresh-waters, however, the salt concentration is high enough to reduce the outer limit of the diffuse layer, allowing the Van Der Waals forces to exceed the repulsion forces at this boundary. This is because there are adequate cations to counter-act the negative charge at the solid surface. Closer to the interface the repulsion forces once more exceed the attractive force. The microbes rest at a distance where these forces are in equilibrium. Thus bacteria may become loosely adsorbed at an interface and may be desorbed by the application of a suitable shear force (Marshall, 1979). This loose adsorption was probably responsible for the observations of Grimes (1974), showing that the disturbance and relocation of bed sediments by dredging operations caused a release of adsorbed FC.

Adsorption to Different Substrates.

Adsorption occurs at many different liquid/solid interfaces, for example, mineral sediment particles, to rocks, plant and animal surfaces, plankton and biological debris (Marshall, 1979). In some upland catchments with low sediment loads, the stream bed can serve as a source and recipient of bacteria to and from the water column. In the absence of sediment, this may be due to interactions between the bacteria and the zoogloeal film which forms on the stream bottom. Thus, a relationship may exist between bacterial numbers and the water/stream-bed contact surface area due to interactions between the bacteria and the zoogloeal film which forms on the stream bottom (Morrison and Fair, 1966).

The adsorptive capacity of mineral sediment particles is determined by the surface area to volume ratio. The greater this ratio the larger the density of charge sites for adsorption. Clay minerals have excellent adsorptive properties, silts, fine sands and coarser sands have successively lower adsorptive capacity (Marshall, 1979). In sandy sediments the actual particle size distribution is important. Unlike clays, which have internal charge surfaces available for adsorption, only the outer surface of a sand grain is available for adsorption. Thus, the smaller the mean particle size the greater the availability of adsorption sites in a given volume of sediment. The degree of adsorption also varies between species of bacteria (White,1979), and is affected by other factors such as pH, the degree of saturation of adsorption sites, and flow regime. Adsorption increases with reduction of pH below 8.0, the addition of divalent cations and, in soils, with decreasing soil moisture (Bitton, 1980).

3.2 Transport of *E.coli* into surface waters.

Bacteria are transported from the land-surface by hydrological processes. The main routes of transport are in surface and sub-surface flow. The sub-surface flow component comprises soil matrix through flow and macropore or bypass flow (non-matrix through flow).

Stream flow may be separated into two components, quick and slow flow, on the basis of the time response of the flow to an impulse, such as an intense rainfall event. The relative importance of these components is dependant on local hydrology, topography and climate. As bacteria can exist free in solution or adsorbed to sediment particles, quick-flow may contain bacteria in both phases. The slow-flow component is more likely to contain bacteria in free solution because slow-flow processes tend to promote deposition of suspended material.

Transport in soils.

Two of the main transport mechanisms for bacteria within soils are matrix through-flow and by-pass or macropore flow, sometimes referred to as non-matrix through-flow. Experiments with disturbed and un-disturbed soil columns have shown that macropore transport is not an important pathway for bacterial transport in disturbed soils. In undisturbed soils, however, transport via macropores can result in significant contamination many tens of metres from the source of bacteria (Smith et al., 1985). Knowledge of current land-use may therefore be quite significant in accurately determining bacterial inputs from agricultural land in upland catchments, because changes in land-use will alter the hydrological response of the catchment to rainfall and hence patterns of bacterial transport. During frequently occurring or short duration rainfall events, however, the sub-soil transport of bacteria will be dominated by matrix through-flow resulting in no significant transport beyond a few metres (Germann et al., 1987). Other authors have found that bacterial transport is enhanced in the saturated zone (Hagedorn et al., 1981).

Hunter et al., (1992) have recently examined the relative contributions of inputs from overland flow, matrix through flow, and non-matrix through flow to stream bacterial numbers in an upland experimental catchment in Yorkshire. Medians of FC concentration, FC load and flow rate were used in this study because bacterial data generally exhibit a log-normal distribution and the arithmetic mean over-emphasise the upper values of the distribution. Matrix through-flow was found to produce a small stream bacterial loading in comparison to non-matrix through-flow and overland flow, due to low concentrations and low flows. It was suggested, however, that as the bulk of matrix through flow input occurs at or below the stream surface, this flow mechanism could account for a large proportion of the water input to the channel, but could only input relatively small numbers of bacteria due to the capacity of the soil matrix to filter them out. Mechanisms which retain bacteria, such as filtration, are described in the literature as deposition mechanisms, those which dislodge trapped bacteria are referred to as entrainment mechanisms, the latter being of least importance when considering bacterial transport (Hornberger et al., 1992; Corapcioglu and Haridas, 1985). The main deposition mechanisms, ie. restrictions to bacterial transport in the soil matrix, are: straining or filtration in the contact zones of adjacent pores; sedimentation in the pore spaces, and; adsorption (Corapcioglu and Haridas, 1985). Organic material can limit the extent of bacterial transport in soils, in the form of a mat composed of bacteria or extracellular polymers. Such mats form an integral part of septic tank drain fields, acting as fine filters to strain out organisms (Yates and Yates, 1988). Hunter et al. (1992) observed FC loads of 3 and 5 orders of magnitude higher than matrix through flow from non-matrix through flow and overland flow, suggesting that matrix through flow is not a significant contributor of base flow bacterial inputs to the stream channel. The low frequency of zero values of bacterial concentration at non-matrix through flow sampling outlets, suggested that those sites were end-points of extended macropore systems. Field observations showed that water and entrained bacteria from the catchment surface on the land close to the stream, reached the non-matrix through flow sampling sites via root systems and non-biological voids. It was suggested that most of these voids occurred near to the soil surface where plant root density

is greatest.

On land used to test the effect of piggery slurry application in Australia, low levels of FC, 1 per 100ml, have been recovered at depths of 1.1m and 1.25m in fine sandy clay soils. The greatest subsoil contamination, occurred for several weeks during water-logging, after winter rains. The contamination was rapid and probably due to transport via contraction cracks (macropores) which developed during the previous summer. In general contamination was higher in the moist soils, and the number of organisms in the effluent was found to have a greater influence on top soil bacterial concentrations than the rate of application (Chandler, Farran and Craven, 1981).

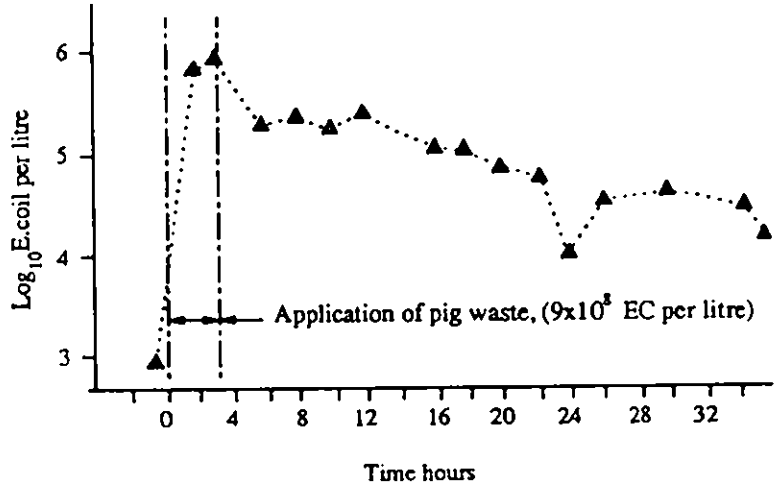

Fig. 3.2: *Response of E.coli concentration in water draining an extensive land drainage system to an application of piggery waste (after Owens and Evans, 1972).*

Land-drainage schemes present a rapid transport pathway for bacteria from a catchment surface. Evans and Owens (1972) studied the *E.coli* response of an extensive land drainage system to an application of pig effluent to the overlying pasture (figure 3.2). The variation in *E.coli* concentration in the land-drain water in the absence of fresh inputs of faecal material was also examined (figure 3.3) and was found to be affected by flow-rate, the number of bacteria in the soil or on the vegetation and the rate of application of slurry. The relationship between flow and *E.coli* concentration accounted for 77% of the observed *E.coli* variation, but was not valid while large volumes of applied slurry remained on the land surface. Figure 3.2 shows the rapid response of the system to the application of waste, discharge was monitored continuously throughout this test and found not to be affected by the application. After the very high concentration peak, the bacterial concentration returned to near background concentration within a matter of days. It is possible that this rapid peak and recovery to near background concentrations may represent the time taken for the bacteria in the waste water solution ,to reach equilibrium with the available adsorption sites within the soil structure. In a later study Evans and Owens (1973), examined the relationship between flow in the land drain and

the concentration of viable bacteria (figure 3.4). These results suggested that at very low discharges bacterial numbers rise, probably due to lack of dilution by soil water. Although it was suggested that this observation might have been due to an artefact of the data as a result of too few samples at low flows (Evans and Owens, 1973). With increased discharge the bacterial concentration fell presumably due to dilution, at the point when the pore water velocity is sufficient to entrain bacteria or perhaps even dislodge particles the bacterial concentration rises steeply as greater numbers of bacteria are desorbed.

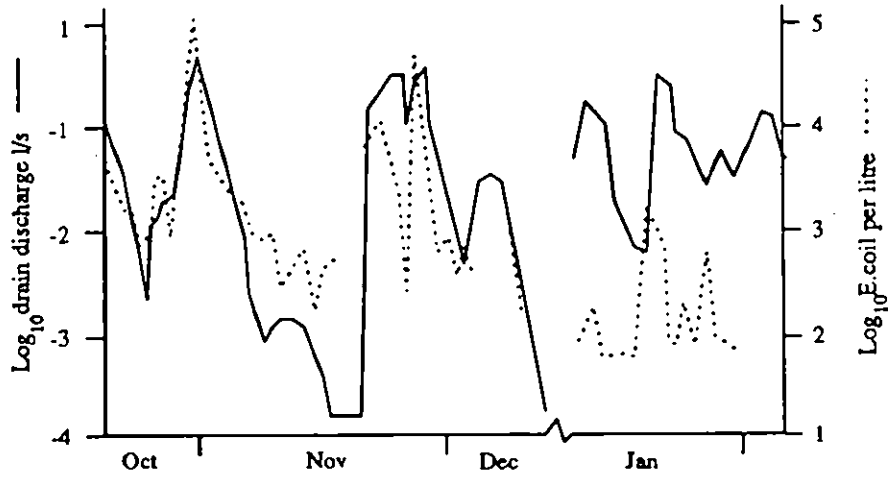

Fig. 3.3: *Variation of E.coli concentration in water draining an extensive land drainage system in the absence of recent inputs of faecal bacteria (after Owens and Evans, 1972).*

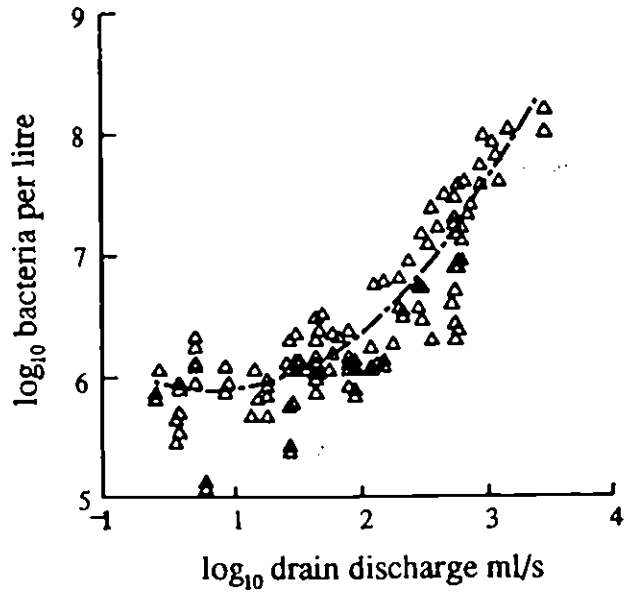

Fig. 3.4: *Curve showing relationship between viable bacteria and land drain discharge (after Owens and Evans, 1973).*

Rainfall Run-off

Bacterial counts in receiving waters, resulting from non-point source contamination, will be highly dependant on local hydrological characteristics (Kunkle, 1970). The proportion of overland and sub-surface flow generated by a particular rainfall event will be dependant on a great number of factors including catchment characteristics, such as soil type, antecedent moisture status, vegetation cover, elevation and the presence of impermeable surfaces, ie. bedrock, and the characteristics of the rainfall event, including duration and intensity and spatial extent, and the local climatic conditions which may influence evapotranspiration rates.

Kunkle, (1970) studying upland catchments with permeable soils, found that bacterial contributions from areas away from stream margins were small compared to those derived near channel and grazing had minimal impact when carried-out away from the stream margins. Upland areas contributed little or no overland flow during storms, most storm runoff originating on saturated areas which built-up along channel edges. It was therefore suggested, that due to the runoff processes, bacterial contamination is probably more a function of activities in and around the stream channel than of basin-wide land use. Hunter and McDonald (1991) found that moist areas, where overland flow was preferentially generated, contributed significantly higher numbers of faecal bacteria to surface waters than dry areas.

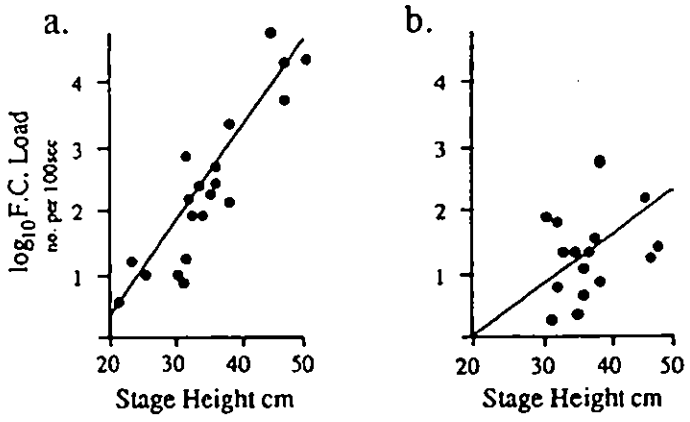

Fig. 3.5: *Scatter plots of $\log_{10}$ faecal coliform load against stage height for inputs from a. overland flow and b. non-matrix through-flow (after Hunter et al. (1992)).*

Hunter et al. (1992), in their study of FC inputs to an upland Yorkshire stream, found that the FC load contributed by overland flow was 5 orders of magnitude higher than matrix through flow and 2 orders of magnitude higher than non-matrix through flow. Overland flow was a major contributor to stream bacterial load during both base and stormflow conditions. Seeps, springs, return flows and protostreams all contributed to overland flow. *Protostreams* occur where subsurface and surface flows combine to produce a defined and recognisable channel. Protostreams were recognised as being extremely important in the transport of bacteria from the catchment surface to stream-bed

sediment store as was the identification of surface water contributing areas which often comprise significant parts of upland heathland. The bacterial input rate was suggested as being largely determined by rainfall conditions, positive correlation was found between FC input load and stream stage height from both overland and non-matrix through flow sites (figure 3.5). In the stream there were strong positive relationships between stage height and FC concentration and hence load. At a quarter of the overland flow sites, however, there was a significant negative relationship between FC concentration and stream discharge, it was explained that this may be due to depletion of the local land store caused by rising flow and increasing bacterial removal, resulting in a dilution of FC concentration. FC *load* at these sites, however, still increased with rising flow. It was suggested that areas prone to depletion may have been those subject to continual water movement and hence bacterial removal. Considerable variation was found in the median FC load values for overland flow, this reflected the hydrological processes influencing the flow to a particular sampling point. Flows were derived, for example, from bacteriologically pure near channel return flows, or from highly concentrated flow in protostreams.

Hunter et al. (1992) suggest that the relative importance of inputs from catchment and bed sediment bacterial stores, may depend to a great extent on the location at which the stream is sampled (figure 3.6). For their sampling location it was indicated that erosion in the channel would predominate, resulting in the likely dominance of land surface over bed-sediment inputs, and that a clear relationship might be expected between bacterial input from the land store and stream bacterial concentration. McDonald et al. (1982), during controlled releases from an upstream reservoir, showed bacterial inputs from the bed sediment store to be dominant over land based inputs, however, the finite nature of this store was also demonstrated.

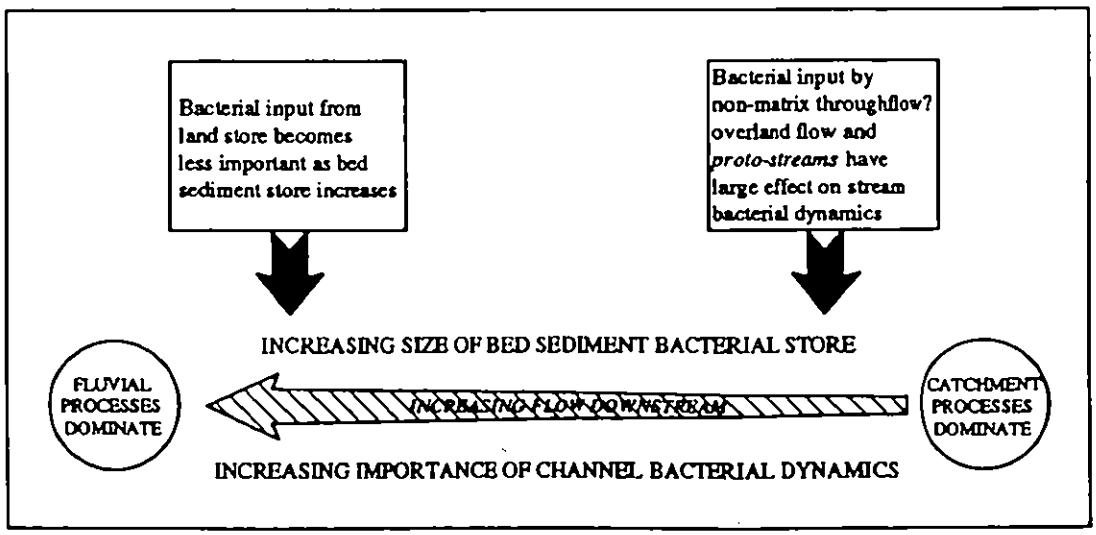

Fig. 3.6: *The relative importance of bacterial input rates from the catchment land store and the channel sediment store to stream bacterial dynamics, with distance downstream (after Hunter et al., 1992).*

3.3 Transport and Storage of *E.coli* in the Aquatic Environment.

In the aquatic environment bacteria mainly exist in two phases, freely suspended in the water column, and adsorbed to solid materials. Bacterial transport in stream and river channels, therefore has two main components; the transient movement of materials stored in the stream bed or deposited on the banks, and the continuous movement of freely suspended bacteria, and those adsorbed to low density solids, with the water body.

It is accepted that adsorption to particles, deposition, storage and subsequent resuspension is one of the major processes of bacterial transport in river channels. Because of the possibility for extended survival and resuspension, bed sediments have the potential to lower the quality of overlying waters when disturbed (Matson et al.,1978). This may, for instance, occur as a result of a recreational activity or scour during the rising limb of a hydrograph event. *E.coli* densities, in sediments, for example, may be up-to 1000 times higher than in the water column (Van Donsel and Geldreich, 1971; Matson et al.,1978; Stephenson and Rychert, 1982). The transport of freely suspended bacteria and those adsorbed to low settling velocity particulates, will be a continuous process associated with water flow. In lakes and reservoirs the potential for transport of sediment bound bacteria, beyond the main body of water, is likely to be limited.

Adsorption and Settlement.

On entering a water body *E.coli* can become adsorbed to particulate matter or may already be adsorbed to particulates. Sediment bound bacteria may settle out into bed sediments, or be maintained in the water column, depending on the nature of the particle and the flow characteristics. Flow velocity is an important factor determining the transport and deposition of a given particle in an open channel (Matson et al., 1978). The initial phase of the bacteria, ie. in free solution or adsorbed, will determine their initial behaviour in a reach.

Experiments to assess whether the adsorption of bacteria onto estuarine silts and marine muds takes place in the water column or at the bed, have shown that 20% of FC are adsorbed immediately onto particles in suspension (Milne et al.,1986). Effective adsorption relies on the availability of sufficient sediment with high adsorptive capacity (Weiss, 1951). When bacterial numbers entering a reach exceed the availability of adsorption sites, the excess bacteria will remain in free solution. This was demonstrated in a study of bacterial concentrations in water and sediment up and downstream of a sewage effluent discharge. Upstream of the discharge point there was statistically significant correlation of bed to water column bacterial concentrations. Downstream, however, no such relationship was found. It was suggested that the upstream concentrations were in equilibrium, and that downstream the sediment was probably saturated with respect to bacteria (Matson et al., 1978).

E.coli in sewage effluent may already be adsorbed to low settling velocity particles such as organic matter. These particles will remain in suspension at much lower flow velocities than mineral particulates, for instance in estuary water. FC deposition was found to be a function of time and the FC deposition rate was directly proportional to SS concentration. SS deposition from the top 30mm of sewage final effluent, however, was not a function of time and no significant alteration in concentration occurred after 3hr. When effluent was mixed with estuary water, deposition of FC was no longer a function of time and the deposition rate exhibited no correlation with SS. Tests between the different particles showed that bacteria were just as likely to become adsorbed to effluent particles as to estuarine silts and muds, the difference in deposition was therefore attributed to the settling characteristics of the particles. The experiments were carried out with concentrations of between $5x10^4$ to $7x10^4$ *E.coli* per 100ml and it was suggested that at other concentrations different behaviour may be observed. This might also be the case under competition with indigenous and other microorganisms of faecal origin which may compete for adsorption sites (Milne et al., 1986).

During stable flow conditions sediment and water bacterial numbers have been observed to achieve a relative steady-state (Matson et al., 1978; Morrison and Fair, 1966).

Resuspension of sediments.

Resuspension of sediments may be caused by hydrological processes such as storm flows, or in shallow waters, by other disturbances caused, for instance, by a bather, a passing boat, or wind wave action. In all cases there might be sufficient bacteria in the sediments to lower the bacteriological quality of the overlying water. The observed effects may depend on the bacteria source and sediment characteristics.

Increased coliform concentrations during storm discharge in mountain streams have been found to be a function of suspended sediment concentration (McSwain and Swank, 1977). Stream bed flushing is generally accepted as a major contributor to high bacterial concentrations, associated with rising stages and larger flows (Kunkle and Meiman, 1967; Morrison and Fair, 1966). The flushing potential of the first peak in any flood, especially after a prolonged dry period, is likely to exceed that of later peaks (Elder, 1978). Seasonal accumulation has been observed in the bed sediment store, low flows during summer being favourable to sedimentation. During the higher flows of winter, with a certain amount of scour and minimal deposition, net transport within the bed may result (Streeter, 1934). But the relative significance of bacterial inputs from channel bed sediments and the catchment surface may depend on the location at which the stream is sampled, inputs from the bed sediment store will generally increase in significance with distance downstream (Hunter et al., 1992).

McDonald et al. (1982) undertook a series of experiments to determine whether increased bacterial concentrations during storm events were the result of washout from the catchment surface or scour of bed sediments. The study site

was in an upland Yorkshire catchment, downstream of a small reservoir. Controlled releases from the reservoir were used to generate artificial hydrograph events in order to test the hypothesis that increased bacterial numbers during hydrograph events were a result of soil-matrix throughflow, non-matrix throughflow and overland flow. According to this hypothesis, the artificially produced hydrograph would not cause an increase in the bacterial concentrations in the stream, and may even reduce levels by dilution, given that the reservoir water, derived from depth, contains low bacterial levels. The hypothesis was found to be false, marked bacterial peaks coincided with the hydrograph peaks. The peak bacterial concentration also increased with distance downstream, with a slightly reduced peak flow, suggesting continued entrainment and accumulation of sediment bound bacteria as the flood wave propagated downstream (figure 3.7). The possibility of exhausting the stream bed sediment supply was also demonstrated during a prolonged release to provide water for a slalom canoeing event. High flow was maintained throughout the event and following the peak in bacterial concentration, which coincided with the hydrograph peak, the bacterial concentration began to fall, suggesting that the bed sediment store was finite and would eventually become exhausted (figure 3.8).

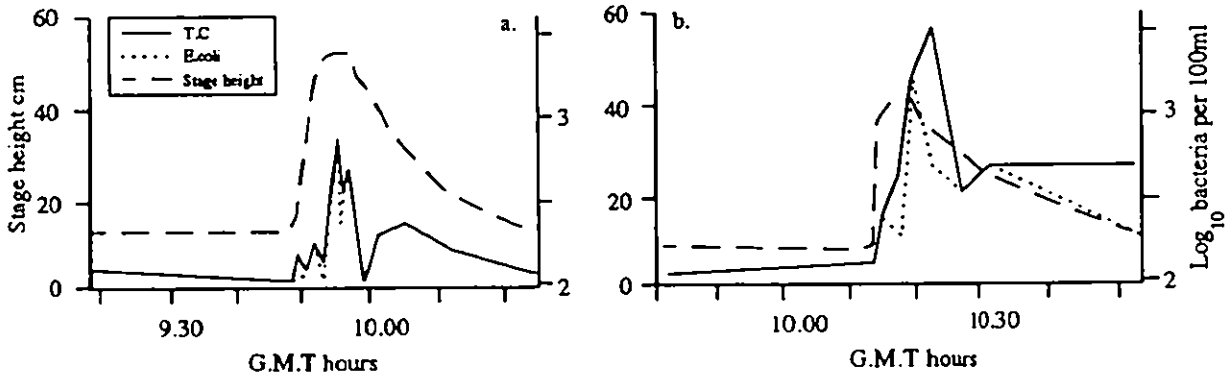

Fig. 3.7: *Propagation of artificially generated hydrograph with response of TC and E.coli concentration at locations (a) 400m, and (b) 2500m downstream of the hydrograph source (after McDonald et al., 1982).*

Immediately after a major natural hydrograph, that would have flushed the channel store of bacteria, a further controlled release was made, no bacterial peak resulted, but slight dilution did occur. It was noted that inputs from overland flow would have been removed during the passage of the peak and receding limb of the natural hydrograph (McDonald et al., 1982).

In Lake Houston, sediment scouring by wind-wave action was thought to be responsible for FC concentrations of up to two orders of magnitude higher at a depth of 1 foot above the bed than at the surface (Davis and Valentino, 1985). In Lake Michigan a net survey was carried-out of FC levels around the mouth of the Milwaukee River. Levels decreased

with distance from the mouth and generally decreased with depth, levels were not even increased in samples taken where bottom sediments were known to have been disturbed in the process of sampling. It was suggested that this might be due to a thermocline produced by the flow of warmer river water over colder lake water. No significant difference in levels during dry or wet weather was found, the main inputs of faecal indicators being sewage treatment

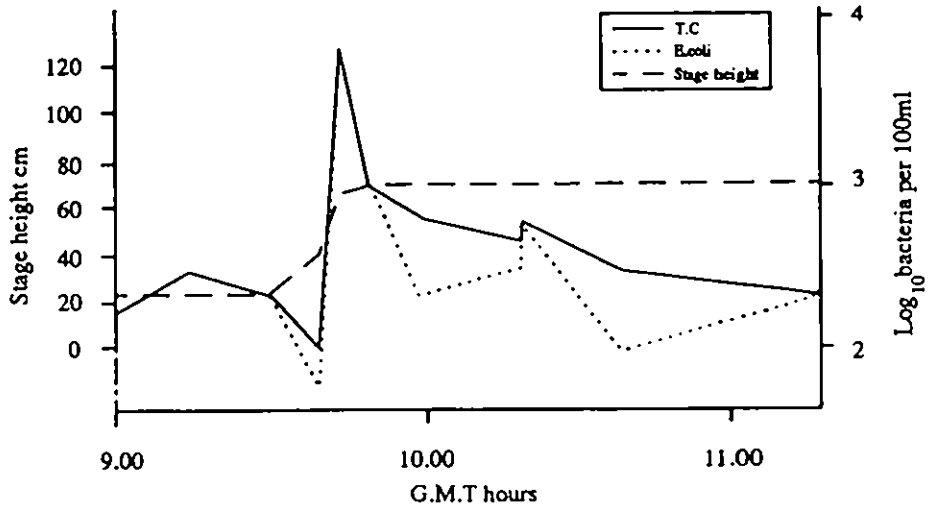

Fig. 3.8: *The response of TC and E.coli concentration to a step change in stage height (after McDonald et al., 1982).*

effluents, urban runoff and storm overflows (Zanoni et al., 1978). This suggests the absence of particles of sufficient settling velocity to supply bacteria to the sediments, and that due to slow flow velocities die-off occurs before the bacteria are transported far from the channel entrance. In DeGray Lake, Arkansas, on the other hand, storm flows were found to contribute considerable loads of nutrients, bacteria and suspended sediment. Turbid storm flows have been observed to travel the 32km length of the reservoir in only eight days. The average residence time of the reservoir is 1.4 years (Johnson and Ford, 1987).

The movement of solids in the stream seems to be the most important means of bacterial transport. These may either be continually suspended, or where they have higher densities be transferred between the water column and the bed sediment store by changes in flow velocity, or by other disturbances such as mans activities. As a consequence bacterial transport in free suspension can be modelled with a simple mixing model such as those mentioned in section 4.

3.4 Summary: The Key Transport Processes for *E.coli*.

1. Bacteria may become loosely adsorbed to solid phases including, for example, particulates, vegetation, stream-bed surfaces and other organisms.

2. Clay minerals have very high bacterial adsorption capacities and this characteristic decreases with particle size. Particle size distribution and mineral type may be important in determining the bacterial storage capacity of a given system.

3. Bacteria may survive on the catchment surface for long enough to provide a semi-permanent reservoir capable of contaminating the surrounding aquatic environment following transport by hydrological processes.

4. The hydrological characteristics of a catchment will determine the transport dynamics of *E.coli* from non-point sources.

5. Transport of bacteria from the catchment surface will be affected by landuse, non-matrix throughflow being negligable in disturbed soils. Non-matrix throughflow can transport bacteria rapidly over large distances.

6. Bacterial inputs from soil matrix throughflow are minimal due to removal mechanisms such as straining, adsorption and settlement.

7. Bacterial inputs from non-point sources and land-drainage schemes will correspond with rainfall events. Applications of waste generally result in short lived bacterial peaks.

8. In head-water catchments areas where overland flow is preferentially generated may be very important in the transport of bacteria to the stream channel. Such areas combined with macropore non-matrix through flow systems and protostreams may offer rapid bacterial transport via connected pathways over extended areas.

9. In a stream channel the bacterial concentration in the water at a particular time will be determined by the balance between the rate of bacterial input from the catchment surface and the input rate from sediment sources in the channel.

10. Bacterial inputs from the catchment surface will become less important than inputs from the stream bed sediment store with increasing distance downstream.

11. Flow hydraulics and sediment dynamics of the channel determine the dynamics of bacterial behaviour in the channel.

12. Bed-sediment storage is finite in nature and large flow events may be capable of depleting this bacterial source. The size of the bed sediment store may also vary on a seasonal basis, with net deposition in periods of low flow and vice versa.

4. Modelling bacterial transport and survival.

Two types of model commonly used in systems analysis, physically and statistically based, have been used for describing bacterial populations in a range of different environments. The physical approach uses exact mathematical relationships based on the understanding of the actual processes occurring in the system to relate bacterial concentrations to a set of environmental parameters or driving variables. Statistically based models present a probabilistic relationship between bacterial population and a given set of variables. These "black-box" models give no information about the physical mechanisms controlling the observed behaviour, although certain characteristics may have a physical interpretation. Statistical models are empirical and developed from field data collected from the system to be modelled, and as such must not be extrapolated beyond the scope of their calibration data. Physically based models may be developed from the perception and understanding the modeller has of the system behaviour and field data may then be used to verify the model structure and for calibration and validation. These process based models are capable of wide application and may be used for predicting future responses.

Models applied to water quality systems, and especially for bacterial applications, may include a combination of physical and statistical approaches. The approach may be determined by the scale of the system to be described and the breadth of detail required in the model output. The scale of the system to be described will in-turn determine the importance of the factors affecting the bacterial concentrations. In small scale systems the factors controlling the observed behaviour may be isolated more easily, and a physical approach adopted, for example, modelling water quality changes in a waste maturation pond. On the other hand, at the catchment scale, it may not be appropriate to attempt to account for every influence on bacterial survival or transport mechanism, especially since catchment physical characteristics are spatially heterogenous and in any case the effect of such influences may be smoothed-out with transport through the catchment. In this case only long term seasonal influences and the effect of rainfall events may be apparent in variations in bacterial numbers in a river channel. Relating stream flow characteristics to bacterial concentration may also rely on a combination of approaches. Some bacterial die-off models are based on time dependant first-order decay dynamics, where time can be substituted for distance in a river channel, given the flow characteristics. Others incorporate a die-off rate coefficient, composed of a number of terms accounting for environmental effects. These additional terms may in themselves be based on a statistical relationship or the physical understanding of the system.

In the literature, bacterial die-off and transport models are used for a range of applications including bacterial washout from faecal material, transport from waste applied to land, soil and groundwater models, river and lake models. Clearly, it is not in the scope of this study to examine every modelling application, only the approaches to modelling bacterial numbers in streams and rivers.

4.1 Lumped Bacterial Die-off Models.

The following section is largely based on a review of bacterial die-off modelling by Crane and Moore (1986).

Physically based models used to describe bacterial die-off are generally based on simple first order decay dynamics as given by Chick's Law (equation 4.1).

$$\frac{N_t}{N_0} = 10^{-kt} \ldots\ldots \text{ ie. } N_t = N_0 10^{-kt} \qquad \text{(Equation 4.1)}$$

The equation describes logarithmic die-off of a bacterial population (N) over a time period (0 to t), with constant die-off rate (k). Figure 4.1 shows form curves for bacterial decay from models based on first order dynamics. Curve 1 describes simple first order decay as given by Chick's Law (equation 4.1).

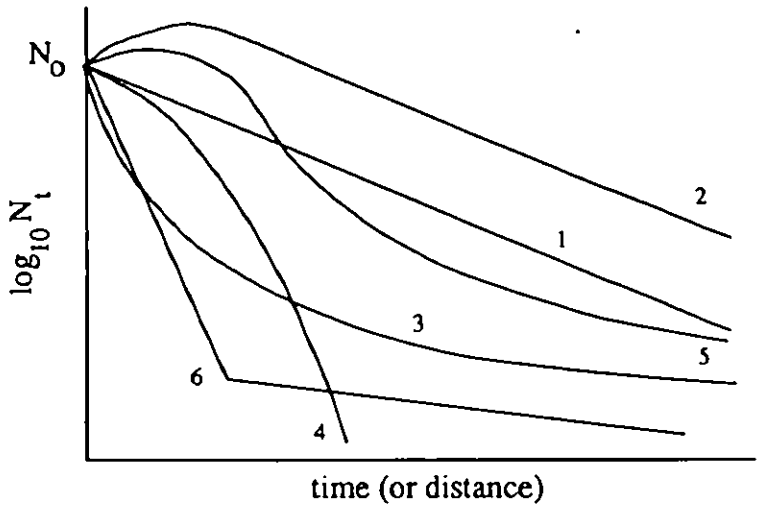

Fig. 4.1: *Bacterial die-off curves as predicted by models in the literature (after Crane and Moore, 1986).*

Bacterial die-off, however, does not always follow first order kinetics (Burn and McBean, 1986) and the onset of die-off often follows a time delay when the population remains static or may even grow. Die-off should be assumed to correspond to a summation of first order reactions (Verstraete and Voets, 1972). Equations 4.2 to 4.10 extend Chick's Law in an attempt to account for such delays, and combined die-off effects. Immediate and constant die-off of the entire population is described by equation 4.1. Equation 4.2 allows for a period of extended survival and growth until $t > t_d$, the time delay, after which decline commences. This is represented as curve 2 in figure 4.1. An initial increase in population may result from reduction in stresses due to dilution, lower levels of toxins, increased nutrient supply,

fewer antagonistic effects from other organisms and a change in oxygen level in the new environment.

$$\frac{N_t}{N_0} = 10^{-k(t-t_0)} \qquad \text{(Equation 4.2)}$$

Equation 4.3 is proposed for die-off of a population composed of a number of sub-groups with different tolerances to environmental stresses.

$$\frac{N_t}{N_0} = a.10^{-k_a t} + b.10^{-k_b t} \qquad \text{(Equation 4.3)}$$

A changing die-off rate may be observed over time as more susceptible groups die-off more rapidly leaving longer lived sub-groups such that the overall die-off rate takes the shape of curve 3 in figure 4.1. Equation 4.3 represents two bacterial sub-groups with a and b the proportions of the total bacterial population having die-off rates k_a and k_b (Streeter, 1934). This equation may be extended to give a complex series explaining a large number of sub-groups with different die-off rates (equation 4.4).

$$\frac{N_t}{N_0} = a\,10^{-k_a t} + b\,10^{-k_b t} + \ldots\ldots n\,10^{-k_n t} \qquad \text{(Equation 4.4)}$$

A number of other models in the literature produce a function similar in shape to curve 3 (equations 4.5 and 4.6), including the statistical model of Burton et al. (1987), see later.

$$\frac{N_t}{N_0} = \frac{1 - 10^{-k't}}{2.3\,k't} \qquad \text{(Equation 4.5)}$$

Where k' is the die-off coefficient.

$$\frac{N_t}{N_0} = (1 + nk_0t)^{-1/n} \qquad \text{(Equation 4.6)}$$

and in equation 4.6. k_0 is the initial die-off coefficient and n is the coefficient of retardation. $n>0$ for a type 3 curve, $n<0$ for type 4 (Phelps, 1944; Fair and Geyer, 1954).

$$\frac{N_t}{N_0} = 10^{-kt^{(-1/n)}} \qquad \text{(Equation 4.7)}$$

Similarly in equation 4.7, a non-uniformity coefficient is used, again with $n>1$ for a type 3 curve, $n<1$ for type 4 (Fair et al., 1971). Curve 4 describes a situation where a continually rising death rate occurs, perhaps in the presence of toxic compounds or as a result of chlorination. Frost and Streeter (1924) used a statistical approach to produce a formula giving function drawn as curve 5 and constants b, c, d and k, in equation 4.8 are empirically derived to fit the data.

$$\frac{N_t}{N_0} = \frac{b}{1 + (a + d)10^{kt}}$$ (Equation 4.8)

Burton et al. (1987) in a study of bacterial survival in different freshwater sediments developed a statistically based model. In this study the only sediment characteristic for which there was an apparent relationship with die-off was particle size, this relationship, however, was not built into the model. The general model form was;

$$y = A e^{-\phi \ln x}$$ (Equation 4.9)

from which the following equation was derived;

$$\ln C = a + b[\ln (t + 1)] + \varepsilon$$ (Equation 4.10)

$\ln C$ is the natural logarithm of the initial bacterial density, a is the intercept, b is the die-off slope, t is time and ε is the residual error.

These die-off models must be used in conjunction with hydrodynamic and mixing models. Alone, these simple models cannot be expected to give a reasonable description of bacterial concentrations in natural systems because of the influence of stream dynamics and catchment flowpaths in response to a changing rainfall input. Their use in applications to streams and rivers would imply that immigration and emigration of bacteria are insignificant, diffusion is insignificant, point sources represent the only input of bacteria and sinks, such as bed sediments, are insignificant (Mahloch, 1973). Simple die-off models are most applicable in rivers and streams receiving high concentrations of bacteria from point-source discharges. In this case, and under constant flow conditions, die-off is effectively time dependant and represents the sum of sedimentation, physical die-off and dilution processes. It is clear, however, that a model of this type would be unable to cope with the sediment and bacterial dynamics associated with a changing flow regime.

4.2 Water Quality Models incorporating the Advection-Dispersion Equation and Simple Bacterial Die-off Functions.

A number of water quality models exist which simulate a wide range of pollutants and water quality characteristics, including bacterial concentration. Applications of such models are wide ranging and include lake systems, estuaries and rivers. Because of their wide ranging application, such models do not always offer a detailed description of the processes relating to bacterial dynamics and often only include simple time decay models such as those discussed in section 4.1. Such models do, however, incorporate flow hydraulics using mass balance and momentum equations combined with principles of continuity and may thus offer a basis for more effective modelling of bacterial dynamics.

The Advection-Dispersion Equation (ADE).

The ADE is commonly used in water quality modelling and flood wave propagation. For pollutant transport the equation simply describes the change in concentration of contaminant by two terms, one for movement with flow, advection and one to describe the effect of mixing on the pollutant concentration. The model can be developed for one, two and three-dimensional transport, this choice will depend on the system to be modelled. In a ground-water system, for example, there may be transport in all three dimensions. In most rivers, however if it is assumed that the flow is well mixed through it's cross-section, a one-dimensional formulation will suffice;

$$\frac{dC}{dt} = -U\frac{dC}{dx} + D\frac{d^2C}{dt^2} \qquad \text{(Equation 4.11)}$$

where, x = distance downstream

C = concentration

U = mean flow velocity

D = dispersion coefficient.

It is assumed that there are no inflows or sinks and that the contaminant is conservative in nature, ie. it is not transformed by physical, chemical or biological reactions. Figure 4.2 is a graphical representation of how the ADE works, showing how a pulse of contaminant entering a point x_0 in a river at time t_0 changes through time. Figure 4.2a, shows how the model works if $U=0$, ie for dispersion only, as might occur in a still pond. Figure 4.2b shows the variation in C if $D=0$, for advection only. Clearly this is an unreal situation, but is used here to demonstrate the model. Combining the two effects gives figure 4.2c showing transport downstream and attenuation of the contaminant

concentration. The ADE is utilised in all of the following models and is applied to salinity variation, sediment and bacterial transport and flood-wave propagation. For flood-wave propagation the velocity term in equation 4.11 uses the kinematic wave velocity instead of mean velocity, because the front a flood-wave travels faster than the mean velocity.

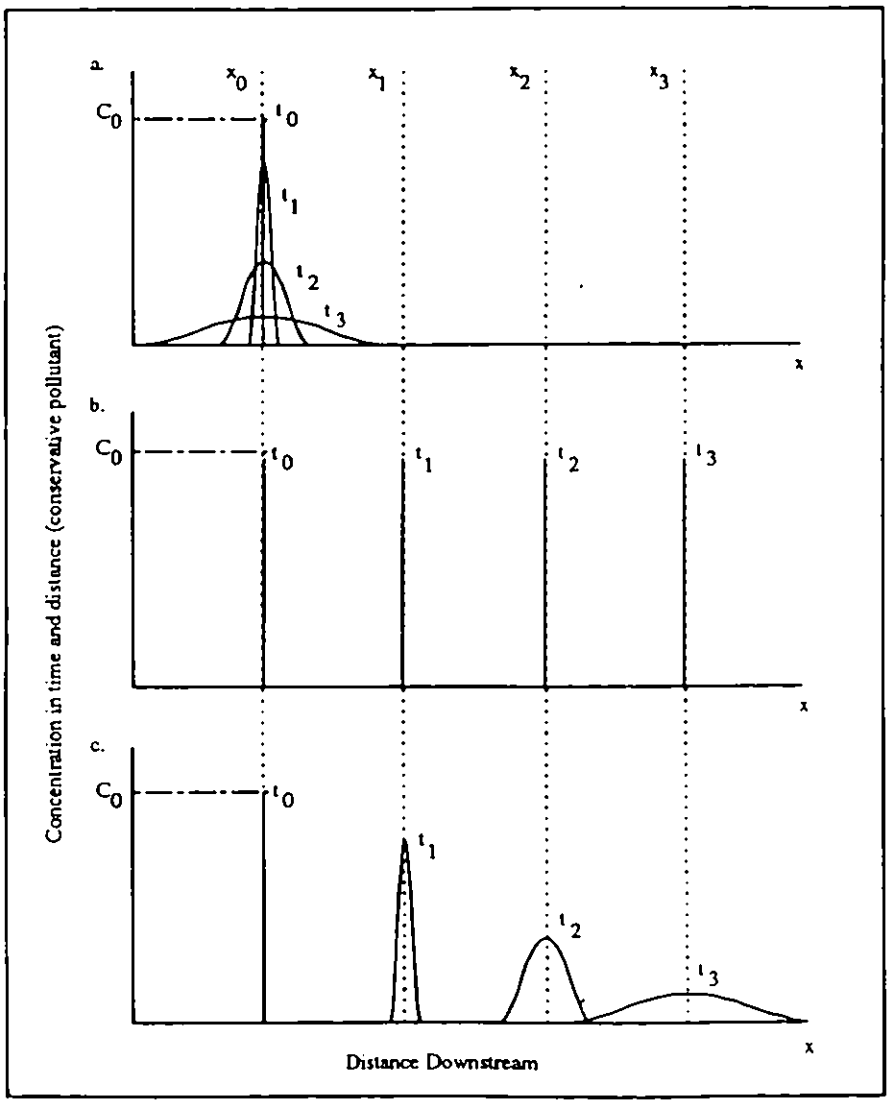

Fig. 4.2: A graphical representation of modelling the dispersal of a conservative contaminant under steady flow conditions using the ADE.

Estuarine Transport Model using the Advection-Dispersion Equation (ADE).

A physically based modelling approach has been applied to the dispersal and inactivation of bacteria in an estuary (Crowther, 1991; Wallis et al., 1989). The model is based on equations 4.12 to 4.13. The hydrodynamic model uses conservation of mass (or continuity) and conservation of momentum (equations 4.12 and 4.13, respectively).

$$(W + W_s)\frac{\partial h}{\partial t} + \frac{\partial Q}{\partial x} = \frac{\rho_L}{\rho} L \qquad \text{(Equation 4.12)}$$

and,

$$\frac{\partial Q}{\partial t} + \frac{\partial}{\partial x}\left[\frac{Q^2}{A}\right] + gAS_f + gA\frac{\partial h}{\partial x} + \frac{gAR}{2\rho}\frac{\partial \rho}{\partial x} = 0 \qquad \text{(Equation 4.13)}$$

where, Q = discharge (cumecs)

h = stage height (m)

t = time (sec)

x = distance downstream (m)

W = width of estuary at surface (m)

W_s = average width of storage areas (m)

L = lateral inflows per unit length (m2.sec-1)

rho = density (kg.m-3)

A = cross sectional area of flow (m2)

R = hydraulic radius (m)

and S_f = friction slope.

The model assumes that storage areas such as docks and basins do not affect the momentum balance in the estuary and that lateral inflows add no momentum to the flow. Storage areas tended to be long and narrow with small entrances, and hence poorly mixed and their effect on solute concentrations is ignored. The advection-dispersion equation (ADE) is used to model transport of a conservative solute, in this case salinity, and is extended to account for sediment and bacteria (equations 4.14 to 4.16, A is assumed constant to allow comparison with equation 4.11):

$$\frac{\partial S}{\partial t} = -\frac{Q}{A}\frac{\partial S}{\partial x} + D\frac{\partial^2 S}{\partial x^2} + \frac{L}{A}S_L \qquad \text{(Equation 4.14)}$$

and,

$$\frac{\partial C_s}{\partial t} = -\frac{Q}{A}\frac{\partial C_s}{\partial x} + D\frac{\partial^2 C_s}{\partial x^2} + \frac{L}{A}.C_{sL} + \frac{E-D}{A}.C_s \qquad \text{(Equation 4.15)}$$

and,

$$\frac{\partial C_b}{\partial t} = -\frac{Q}{A}\frac{\partial C_b}{\partial x} + D\frac{\partial^2 C_b}{\partial x^2} + \frac{L}{A}.C_{bL} + K.C_b \qquad \text{(Equation 4.16)}$$

where, S = salinity, and,

C_s, C_b = concentration of sediment and bacteria respectively,

D = dispersion coefficient,

E = ammount of sediment eroded from channel bed,

D_p = deposition rate,

Subscripts s and L refer to static storage areas and lateral inflows.

and, K = bacterial inactivation rate (s-1).

All of the different influences on survival are lumped into one value, K, and so sunlight and temperature effects are implicitly included since, K is derived from experimental results as a function of local salinity and suspended solids concentration. In simple terms bacterial concentration is modelled as a function of flow with first order decay.

Note that unlike the model proposed by Jenkins (1984), the influence of sediment dynamics on bacterial concentration in the water column is not accounted for (equations 4.15 and 4.16). This fact is surprising considering previous research undertaken by Milne et al. (1986, 1989, 1991) which recognises that sedimentation plays a role in bacterial inactivation. In equation 4.15, the terms for deposition and erosion, E and D_p, are based on the Krone formula, relating entrainment and deposition to channel bed shear velocity (u_*) and critical velocities of deposition and entrainment, v_d and v_e, respectively. Equations 4.17 and 4.18 show the functions used (Crowther, 1991):

for deposition $u_* < v_d$,

$$D_p = \left(1 - \frac{u_*^2}{v_d^2}\right) W_b v_s \quad \text{... for } u_* < v_d \qquad \text{(Equation 4.17)}$$

and for erosion $u_* > v_e$,

$$E = \left(\frac{u_*^2}{v_d^2} - 1\right) W_b \chi \quad \text{... for } u_* > v_e \qquad \text{(Equation 4.18)}$$

Where, chi is the erosion rate, W_b the bed width, and v_s the settling velocity. Not considered in this model is the fact that, for a given grainsize $v_e > v_d$ (linked to the hysteretic behaviour of sediment scour/deposition cycles), as the energy required to entrain particles is greater than that necessary to maintian their suspension (Hjulstrom, 1935). Therefore for $v_d < u_* < v_e$ the sediment will be maintained in transport ie. no net deposition or entrainment. This method of assuming either entrainment or deposition is a different approach to that of Jenkins (1984), which assumes that

deposition occurs continuously and the occurrence of either net entrainment or deposition depends upon the relative rates of erosion and deposition. In a very simplified application of equations 4.17 and 4.18 to bacterial transport, a further reduction of the deposition threshold velocity in equation 4.17 could be used to account for the release of bacteria from particulates when entrained in a turbulent flow, subsequent deposition occurring after the deposition of the particulates and as the bacteria re-attach to other solids entering the flow. Shear velocity, u_* is calculated from:

$$\left|\frac{Q}{A}\right| = \frac{u_*}{k} \log_e \left| 30.2 \frac{H}{k_s} \right| \qquad \text{(Equation 4.19)}$$

where k_s is the Nikuradse sand roughness coefficient, H is the depth, and k the von Karman constant for flow with sediment is 0.174.

A further development of these models might be the combination of equations 4.15 and 4.16 to account for sediment related bacteria as well as those associated with water flow.

River Transport Model (ADE)

The ADE has been frequently employed in water quality modelling, especially on large lowland rivers, for example the Tigris River in Iraq (Al-Layla and Al-Rizzo, 1989). The model uses a one-dimensional version of the ADE to simulate a steady-state decline in Total coliform count for 75km downstream of the "Saddam" Dam due to microbial self-purification of the river and described in the model by a simple first order decay coefficient similar to that of Crowther (1991)(equation 4.16). Figure 4.3 shows a profile of measured and simulated coliform concentrations.

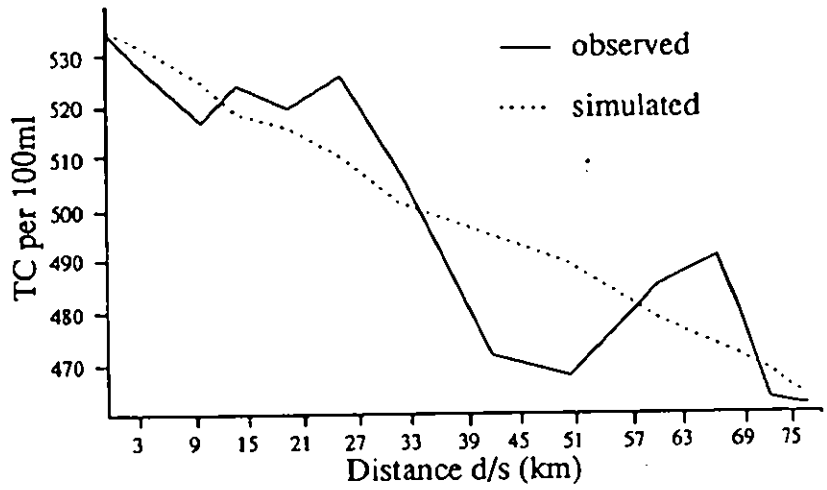

Fig 4.3: *Profile of simulated and measured coliform bacteria concentrations for the Tigris river in Iraq (after, Al-Layla and Al-Rizzo, 1989).*

It is aparent from figure 4.3 that whilst the general trend of coliform decay is effectively modelled, the model does not account for the more complex behaviour displayed by the observed data, indicating some inadequacy in either the model formulation or in its parameterisation.

Sediment Transport model.

The importance of sediment in determining the fate of bacterial contaminants in river and lake sytems has been stressed at all stages of this review. Historically, the behaviour of suspended solids in open channels has been described in a two dimensional context i.e vertical and horizontal. Consequently, a two-dimensional steady-state formulation of the ADE has been utilised to describe the transport and distribution of suspended solids and associated pollutants, including bacteria, in a steady turbulent river channel (Uchrin and Weber, 1979, 1980, 1981).

$$\frac{\partial C}{\partial t} = -U\frac{\partial C}{\partial x} + D\frac{\partial^2 C}{\partial x^2} + W\frac{\partial C}{\partial y} + D\frac{\partial^2 C}{\partial y^2} \qquad \text{(Equation 4.20)}$$

where, C = concentration of suspended solids,

x = horizontal channel distance,

y = vertical distance above bed,

W = particle settling velocity,

U = local time averaged velocity, assumed to be the same for ambient fluid and suspended solids,

and D = turbulent diffusion coefficient for particles.

If a uniform vertical distribution of particles is assumed an upstream boundary condition and an upper water surface boundary condition can be formulated, assuming no there is no transport through that surface. A generalised bottom boundary condition may be formulated to account for the possibility of complete settling or re-entrainment of particles given by:

$$D\frac{\partial C}{\partial y} = -tWC \qquad \text{(Equation 4.21)}$$

where t is a re-entrainment coefficient ranging between 0, for complete settling, and 1, for complete re-entrainment. W is determined as a function of hydraulic and particle characteristics using an empirically based analytical method. Particle settling velocity is treated as a randomly distributed function, generated from quiescent column settling data, which can be calibrated to account for turbulence. Given these boundary conditions and definitions, the two-dimensional partial differential equations (PDE) can be solved numerically. The resultant solutions can then be used to generate a rate law for specific water quality parameters of the form;

$$\frac{dC}{dt} = kC^n \qquad \text{(Equation 4.22)}$$

where t is the time of travel and k and n are constants relating to particle and hydrodynamic conditions. For uniform particle fall velocity, n has been found to approach unity.

This suspended solids model has been incorporated into a lake system model, which utilises a finite section approach, and applied to a recreational lake in South East Michigan in the United States. In application of this model bacterial removal from the water column, other than by sedimentation, ie. predation, die-off, etc., was lumped into a first order function parameterised from field data.

IH Quasar

The model QUASAR (Quality Simulation Along Rivers) has been developed to assess the environmental impact of pollutants on river water quality. The model has been applied to range of UK rivers such as the River Tawe to assess heavy metal pollution and the River Thames, to assess the movement and distribution of nitrates and algae (Whitehead and Williams, 1982, Whitehead and Hornberger, 1984). The model performs a mass balance of flow and water quality sequentially down a river system. The model structure takes account of inputs from tributaries, groundwaters and effluent discharges, and allows for abstractions, chemical decay processes and biological behaviour along the river.

Forecasting and planning information is generated for key locations along the river. The water quality parameters modelled are *E.coli*, nitrate, dissolved oxygen (DO), biochemical oxygen demand (BOD), ammonia, ammonium ion, temperature, ortho-phosphate, pH, and a "conservative" water quality parameter. To model these parameters the river is divided up into reaches. The reach boundaries are determined by points in the river where there is a change in the water quality or flow due to the confluence with a tributary, the location of a sewage treatment final effluent discharge, abstraction, or location of weirs. Water quality changes due to biochemical or chemical reactions are also considered by ensuring appropriate reach lengths (Piper et al., 1992). The flow models are derived from the Muskingum-Cunge flow routing method and give a good approximation to the advection-dispersion equation (ADE) (equation 4.23), for a full description of the application of this technique see Whitehead et al. (1979).

$$\frac{\partial Q}{\partial t} = -u\frac{\partial Q}{\partial x} + D\frac{\partial^2 Q}{\partial x^2} \qquad \text{(Equation 4.23)}$$

Where, Q = discharge

u = kinematic wave velocity

D = dispersion coefficient

The flow equations provide discharge data for the contaminant mixing models. For *E.coli* a simple first order temperature dependant decay model is incorporated (equation 4.24) into the continuously stirred tank reactor, or aggregated dead zone model (ADZ), below;

$$\frac{dC_t}{dt} = \frac{Q_t}{V}.U_{t-\tau} - \frac{Q_t}{V}.C_t + l_t - k.C_t \qquad \text{(Equation 4.24)}$$

Where, Q_t = discharge at time t

V = reach volume

C_t = bacterial concentration at time t

$U_{t-\tau}$ = is the upstream input to the reach allowing for travel time τ.

l_t = sources and sinks due to effluents, abstractions etc.

k = temperature dependant bacterial die-off coefficient (days-1)

The ADZ, or Continuously Stirred Tank Reactor, treats each river reach as a storage volume, or a series of buckets, the outlet of the first feeding into the next etc., each reach is stirred or mixed continuously, and the residence time of the water in each reach is given by, V/Q.

In this form the the model is clearly only capable of describing the distance decay observed downstream of a point-source input discharging large numbers of bacteria. It is, however, a good example of a water quality model which incorporates a flow model, a contaminant dispersal model and a simple bacterial die-off function. By modification to include sediment entrainment functions, QUASAR has the scope to include a more process-based formulation for describing bacterial transport.

4.3 Process based modelling of bacterial dynamics in upland streams.

A number of applications modelling bacterial survival and transport in the aquatic environment have been presented. Models tend to cover a specific aspect of bacterial transport or combine a number of sub-models describing non-point source transport, direct point-source inputs, receiving water mixing and transport, sediment transport, and die-off.

In a model developed to predict bacterial numbers in upland streams (Jenkins et al., 1984), water and sediment bacterial concentrations are represented by mass balance equations such that:

$$C = I + W - S - D \qquad \text{(Equation 4.25)}$$

and,

$$N = S - W - D \qquad \text{(Equation 4.26)}$$

where, C = concentration of E.coli in water,

N = number of E.coli in bed sediments,

I = input from land-surface,

W = washout from bed sediments,

S = loss to sediments (settlement) and,

D = net die-off as a result of environmental stresses.

Sediment behaviour is determined by the flow characteristics of the stream ie. turbulence and velocity as related to discharge. The on-set of washout is assumed to occur at some threshold discharge below which the washout term is zero. It is recognised that this may be a simplification of a more complicated fluvial process associated with sediment release following cobble movement and may also be sensitive to the adherence characteristics of the bacteria to the bed and/or sediment, but nevertheless, a sediment entrainment function utilising this threshold velocity or discharge concept are common in fluvial models (Graf, 1971). Non-point source inputs are perceived to be associated with "quick" and "base" flow runoff components. The baseflow component comprises, soil throughflow and groundwater flow, responsible for maintaining inputs during dry periods. The quickflow component resulting from rainfall events over the catchment, resulting in surface run-off and pipeflow. It is assumed that rainfall induced processes increase in intensity through the storm, discharge increases as a result, and input will, therefore, increase linearly with discharge. At the same time background and discharge related input will be diluted by the volume of flow. Input of bacteria to the stream from the surrounding catchment (I) is, therefore, described by the function:

$$I = \frac{I_B}{Q} + \frac{I_Q Q}{Q} = \frac{I_B}{Q} + I_Q \qquad \text{(Equation 4.27)}$$

Where, I_B = background input

I_Q = discharge related input

Q = discharge

Bacterial inputs from the catchment surface are assumed to enter the water store only, in the first instance. Transfer between the water and bed sediment bacterial stores is modelled as an internal function of the reach. Increased inputs raise the sediment store bacterial numbers indirectly by providing larger numbers of bacteria for sedimentation. It is suggested that this sedimentation effect is unlikely to remain linear as discharge increases, due the effect of turbulence which is assumed to reduce the rate of settlement. The model uses a threshold discharge value at which bacterial settling halved. Equation 4.28. shows the function used;

$$S = P_{set} \frac{Q}{1 + Q/P_Q} C = p.C \qquad \text{(Equation 4.28)}$$

Where, P_{set} = rate of settlement (proportion of total load settling in unit time)

P_Q = discharge at which P_{set} halves.

The model also accounts for changes in stream hydraulic characteristics with discharge, which will affect the rate of change of numbers of bacteria per unit stream bed area ie. as discharge rises the increased hydraulic radius allows bacteria to settle over a larger area (equation 4.29).

$$\frac{dN}{dt} = \frac{P_{set} \frac{Q}{1 + Q/P_Q} C}{v.w} = \frac{S}{v.w} \qquad \text{(Equation 4.29)}$$

Where, N = number of bacteria in the bed sediment store per unit bed area,

v = velocity,

w = channel width.

It is assumed that $v.w = Q/h$, where h is the mean flow depth, and that $h \propto Q^f$, where, $F = 0.5$, therefore;

$$v.w = \frac{Q}{Q^{0.5}} \qquad \text{(Equation 4.30)}$$

Substituting equation 4.29 into 4.30 and rearranging, gives;

$$\frac{dN}{dt} = P_{set} \frac{C}{1 + Q/P_Q} Q^{0.5} \qquad \text{(Equation 4.31)}$$

This modification causes a slight decrease in the number of bacteria settling into the sediment store once $Q > P_Q$.

Entrainment or wash-out of bacteria from the bed sediment store is assumed to follow a suspended sediment function based on a velocity/discharge relationship whereby;

$$\textit{Bacterial Load} = a.Q^b \qquad \text{(Equation 4.32)}$$

it is suggested that values for b fall in the range 2-3. Because bacterial entrainment occurs at some threshold discharge Q_T, W the bacterial washout is given by;

$$W = a(Q^2 - Q_T^2)N \qquad \text{(Equation 4.33)}$$

Where, N = the number of bacteria in the sediment store

Q_T = threshold discharge.

The empirical constant, a, represents the total bacterial wash-out for $Q > Q_T$ and is expressed as;

$$a = \frac{T_{H2}}{2.Q_T + 1} \qquad \text{(Equation 4.34)}$$

where, T_{H2} represents the proportion of the bacterial store washed-out at a discharge of $Q_T + 1$ $m^3.s^{-1}$. This allows a to be conveniently formulated in terms of perceived system behaviour or field observations. Equation 4.33 is further modified to account for the fact that washout was found to be a function of the size of the bed sediment store, ie. it is assumed the entrainment capacity of the flow limits the ammount of bacteria washed-out of the bed store. In equation 4.33, N is replaced by;

$$f(N) = \frac{N.\,Sat}{N + Sat} \qquad \text{(Equation 4.35)}$$

Where, Sat is the maximum value of $f(N)$.

$f(N)$ only approaches Sat for values $N>>Sat$ (figure 4.4).

The term for bacterial die-off D in equations 4.25 and 4.26, is based on the assumption that the net die-off follows simple first order decay dynamics;

$$C_t = C_0\, e^{-kt} \qquad \text{(Equation 4.36)}$$

Where, C_0 and C_t are concentrations at time $t=0$ and t, respectively, and

k = rate coefficient.

To represent die-off in terms of T_{90} equation 4.36 may be rearranged; at $t=T_{90}$,

$$C_{T_{90}} = \frac{C_0}{10} = C_0 e^{-k T_{90}}$$ (Equation 4.37)

and,

$$k = \frac{\log_e 10}{T_{90}} \quad and, \quad D = k C_0$$ (Equation 4.38)

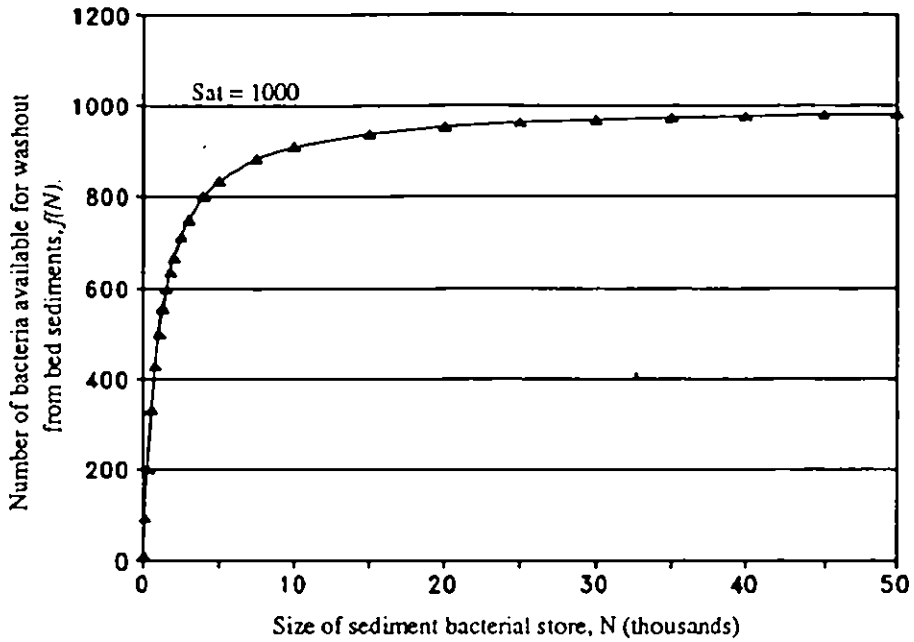

Fig. 4.4: *Plot of washout limitation function, for Sat=1000, as given by equation 4.18.*

The system equations for the model are derived by combination of equations 4.14, 4.15, 4.19 and 4.28:

$$\frac{dC}{dt} = -K_c.C - p.C + \frac{a(Q^2 - Q_i^2)N}{Q} \cdot \frac{I_s}{Q} + I_Q$$ (Equation 4.39)

$$\frac{dN}{dt} = -K_n.N + p.Q.C - a(Q^2 - Q_i^2)N$$ (Equation 4.40)

where, Kc, and Kn = Bacterial decay coefficients in water and sediment, respectively,

p = Settlement coefficient

In validation runs the model is shown to predict both timing and magnitude of bacterial peaks adequately, under a range of conditions. It is suggested, however, that further calibration and parameterisation is required to produce the

observed coincidence of bacterial peaks with respect to the hydrograph peaks (Jenkins et al., 1984). The model has only been tested at the site for which it was developed and has received little sensitivity analysis. On the other hand, use of this model is simple because only discharge and inflow data are required as driving variables. Conversely several parameters must be calibrated and initial values of N and C are required. Both of these tasks may prove difficult to acheive. Nevertheless, this model provides a detailed process-basis for bacterial dynamics upon which to build.

4.4 Summary of Approaches to modelling bacterial dynamics.

1. Simple time/distance dependant first-order bacterial die-off models can be modified to account for the influence of different factors on bacterial survival.

2. These can only be used to any affect when modelling point-source input discharges of large numbers of bacteria in relation to the background concentrations in the receiving water and under steady flow conditions.

3. Existing water quality models often incorporate simple bacterial die-off functions within hydrodynamic models and contaminant dispersal or mixing models, however, modification to include more process-based equations would be required to widen the applicability of such models to bacterial transport dynamics in rivers.

4. If applied on a reach by reach basis, large quantities of data describing input and output flows, bacterial concentrations and hydraulic characteristics are required and a large number of parameters require calibration. This tends to limit the usefulness of these models for wide geographical application and for operational management of surface waters.

5. Process based models require minimal data but have a large number of parameters which must be specifically calibrtated at each site.

6. Process based models represent the most attractivs way forward. Work is necessary to calibrate/validate the models and to assess spatial variation in calibrated parameter values such that these models can be applied across wide regions.

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