DEVELOPMENT OF A CODE OF PRACTICE
FOR CO-DISPOSAL TO OBVIATE
INIMICAL ENVIRONMENTAL IMPACTS
OF GENERATED GASES AND LEACHATES

by

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DECLARATION

The experimental work described in this thesis was carried out in the Department of Microbiology and Plant Pathology (International Centre for Waste Technology, Africa), University of Natal, Pietermaritzburg, from October 1993 to May 1996, under the supervision of Professor Eric Senior.

These studies represent original work by the author and have not otherwise been submitted in any form for any degree or diploma to any other University. Where use has been made of the work of others it is duly acknowledged in the text.

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ABSTRACT

Despite its phasing out in numerous countries, such as Germany and the U.S.A., co-disposal of hazardous waste with municipal solid waste continues to be widely practised in South Africa. Co-disposal utilises properties and microbial activities in the refuse to attenuate the hazardous waste and thus obviate its environmental impact potential. All landfill operations require careful planning in not only site selection criteria but also the type and amount of various wastes accepted for disposal. It is clear, however, that the practice of co-disposal requires special precautions and management as the methods employed in the landfill operation determine to a large extent the environmental effects and, thus, the public acceptability of the operations.

Although co-disposal is not suitable for all industrial wastes the results of recent research efforts, conducted mainly in the U.K., have indicated that, when properly managed, co-disposal can be regarded as a safe and efficient disposal option for many hazardous wastes. Environmental awareness in many European countries ensures that numerous hazardous compounds are either recycled or recovered. Unfortunately, in South Africa the lack of similar concern has resulted in increased concentrations of toxic compounds being co-disposed on a regular basis. Since fundamental studies of this technology, pertaining to South African conditions, have been lacking laboratory models/microcosms were built to address this paucity.

Model. To effect the separation of species habitat domains of component species of growth rate-dependent interacting microbial associations responsible for terminal catabolic processes of the refuse fermentation, with retention of overlapping activity domains, and so facilitate examination of species in isolation without violating the integrity of each association, multi-stage models were constructed. The accidental overgassing of the culture with liquid petroleum gas (LPG) effected interesting fermentation balance changes which also emphasised the need for an Anaerobic Bioassay Test to assess the impacts of specific perturbants. Evidence of differential susceptibility of the component species to phenol was demonstrated in this study.

Microcosm. A total of 42 refuse packed single-stage glass column bioreactors were commissioned and subjected to phenol and/or anaerobically digested sewage sludge co-disposal. The effects of four different operational modes: leachate discard (single elution); leachate recycle; batch; and simulated rain on the co-disposals as well as refuse catabolism *per se* were examined.

The results of these studies indicated that protracted periods of adaption to phenol (1000 and 2000 mg l^{-1}) could have resulted from nutrient (elemental) limitation. Circumstantial evidence was also gained which indicated that the nitrate- and sulphate-reducing bacteria (SRB) were particularly sensitive to the added xenobiotic. Further, without the effective participation of the nitrate- and SRB the active and total fermentation of both the phenol and refuse components were depressed. It was also determined that the operating regime employed was a key factor in refuse degradation although with time, and especially following the phenol resupplementations, the operating conditions played a less significant role. In general, the single elution operated columns demonstrated increased phenol removal rates which were, unfortunately, coincident with low pH values and increased leachate residual phenol concentrations. Leachate recycle, on the other hand, unlike the batch operated columns, facilitated increased pH values and methane evolutions. The simulated rain columns were characterised by rapid washout of the added phenol as well as methanogenic precursors.

The sewage sludge co-disposal experiments, likewise, demonstrated that, depending on the sludge:refuse ratio, the operating regime was extremely important in optimising the refuse degradation processes although, in general, leachate recycle appeared to be the most favoured method of operation.

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CHAPTER 1

INTRODUCTION

1.1 The Nature of Hazardous Waste

The term "hazardous" waste has both proved difficult to define precisely and has no legal definition in many countries (Robinson, 1992). In legal terms, in the U.K., there are only two distinct waste types. The first, "controlled waste", comprises all household, industrial and commercial wastes, and, thus, includes most wastes with the exception of those from agriculture, and mining and quarrying. The second, "special waste", is waste which has potential to cause acute harm or injury to people directly exposed to it (Robinson, 1992; Watson-Craik, 1995). Special wastes were further defined in the U.K. Control of Pollution Act (Special Wastes) regulations (1980). When promulgated, these regulations were specifically aimed to provide tighter control for the transportation of wastes which could be hazardous to human health (U.K. Department of Environment, 1988). Further, a controlled waste is considered a special waste if:

a.   It consists of, or contains, any of the substances listed in Schedule 1 of the Control of Pollution (Special Waste) Regulations (1980) and, by reason of the presence of such substances, is:
    1.   Dangerous to life; and/or
    2.   Has a flash point of $\leq 21°C$; and
b.   It is a medicinal product available only on prescription (U.K. Department of Environment, 1988).

The U.S. Environmental Protection Agency (EPA) identifies as hazardous any waste that possesses any one of the following characteristics (Goldman, Hulme and Johnson, 1986; Miller and Miller, 1991):

a.   It exhibits ignitability, corrosivity, reactivity (explosiveness) or toxicity;

b.   It contains any of the toxic constituents named on published lists as having toxic, carcinogenic, mutagenic or teratogenic effects on human or other life forms; and

c.   It is listed on prescribed lists.

By 1994 the EPA had prepared a list of 363 compounds which it classified as hazardous. This list of organic and inorganic substances is subject to continual updating as new assays for hazardous materials are developed (Watson-Craik, 1987; Miller and Miller, 1991).

A variety of parameters, such as the aquatic and mammalian toxicity, carcinogenicity, levels of production, reactivity, bioaccumulation and persistence, make a contribution to what is called the 'pollution potential' of xenobiotics. This measure is used to distinguish priority pollutants which are compounds which are thought to cause severe environmental damage. Every country decides on its list of priority pollutants. In the Netherlands, for example, diverse compounds such as ammonia, nitrate, cadmium, polycyclic aromatic hydrocarbons, polychlorinated biphenyls, asbestos, carbon monoxide, ozone and radon gas are listed. The European Community on the other hand has compiled a Framework Directive (76/464/EEC) relating to dangerous substances which includes two lists of dangerous substances (Leach, 1994a). List I, or the 'black list' of pollutants, is constituted by compounds which impose an immediate threat to humans and ecosystems. These compounds are illustrated in Table 1.1. List II, or the 'grey list', includes compounds that are considered to be less hazardous by virtue of being either less toxic or less persistent and include molecules such as ammonia and compounds of metals such as zinc, copper and lead (Leach, Middelbeek and Mijnbeek, 1994).

In 1990 the South African Department of Environment Affairs (DEA) commissioned, first, the Foundation for Research Development and, then, the Council for Scientific and Industrial Research (CSIR) to undertake a comprehensive study of hazardous waste and the legislation pertaining to it.

Table 1.1 Categories of 'Black List' Compounds as Classified by the European Community

Categories	Compounds
i	heavy metals, metalloids and related compounds;
ii	halogenated organic compounds;
iii	organic phosphorus compounds;
iv	organic stannous compounds;
v	persistent mineral oils and hydrocarbons from crude oil;
vi	other organic compounds;
vii	cyanides; and
viii	asbestos.

The aim of the investigation was to establish a strategy and action plan for the disposal of hazardous wastes in an environmentally friendly manner. In the absence of a formal regulatory system, South Africa does not meet the requirements of the Basle Convention. Since the net result is a reduction in international trade, steps must be taken to improve hazardous waste management in this country (Department of Environment, 1992f).

A final report (in five volumes) of the investigation was released in 1992 and encompassed the following aspects:

a. A situation analysis, including the quantities of waste generated and their hazard ratings;

b. Available technologies for hazardous waste treatment and disposal;

c. A proposed strategy, policy and regulatory system for hazardous waste management;

d. Legislation options; and

e. An impact assessment to compare different policy options for the regulation of hazardous waste.

The CSIR Report, and the subsequent Minimum Requirements for the Handling and Disposal of Hazardous Waste, proposed the following definition: *"hazardous waste" is any waste that directly or indirectly represents a threat to human health or to the environment by introducing one or more of the following risks:*

a.   *Explosion or fire;*

b.   *Infection, pathogens or parasites;*

c.   *Chemical instability, reactions or corrosion;*

d.   *Acute or chronic toxicity;*

e.   *Cancer, mutations, tumours or birth defects;*

f.   *Ecotoxicity or damage to ecosystems; and*

g.   *Accumulation in biological foodchains, persistence in the environment or multiple effects so that it requires special attention and cannot be released into the environment, or be added to sewage, or be stored in a situation which is either open to air or from which leachate water could be produced* (Department of Environment, 1992a; Anon. 1994b; Peckham, 1994).

Clearly, hazardous waste includes a wide variety of chemicals and waste materials and can be produced as a by-product of most human activities. Further, this waste can take any form (gas, liquid, solid or a mixture of these) and can contaminate any medium such as air, water or soil.

1.2   Quantities Produced

On a global basis, the World Health Organisation (WHO) estimated that before 1980 some 4 million chemicals had been either isolated from natural products or had been synthesised. Of these, some 60 000 were thought to be in daily use with 200 new chemicals marketed each year (Senior and Balba, 1984). This number has now risen to >2500 new chemicals each year (E. Senior, Personal Communication.) Unfortunately, despite estimates that between 60 and 90% of all cancers are of environmental origin, of which many have latency periods of >30 years, less than 20% of the identified "toxic" chemicals have ever been evaluated for their environmental impact potentials (Senior, 1991).

The total amount of hazardous waste produced by the countries of the European Community (EC) was calculated to be between 15 and 20 million tonnes in 1983 of which, approximately, 7 million tonnes were disposed of to landfill (Pearce, 1983). The United States of America is by far the greatest producer of hazardous waste, generating ten times as much hazardous waste as all of western Europe. In 1991, the U.S.A. total was officially estimated to be 500 million tonnes (Moyers, 1991). It has, however, been pointed out that figures such as these are not totally representative as more than 3 million tonnes of hazardous waste was shipped from the European industrialised nations to less developed nations between 1986 and 1988. For example, the West African nation of Guinea-Bissua hoped to receive US$ 120 million more than its total annual budget, by agreeing to store industrial wastes from other countries (Anon, 1991; Moyers, 1991).

South Africa is also seen by the international toxic waste traders as an ideal place for disposal. This increased interest in Third World nations results from the tightening of already stringent controls that govern the disposal of waste in European countries as well as the U.S.A. (Koch, Cooper and Coetzee, 1990). The now infamous Thor Chemicals plant at Cato Ridge is an example of this as the U.S.A. company American Cyanamid exports 10 tonnes of mercury waste each year to Thor Chemicals. Despite the detection, in water and soils around the plant, of the highest levels of mercury ever recorded in South Africa, and the subsequent suspension of all operations by the Department of Water Affairs, the company has restarted processing following design and construction modifications. The departments of Health, Environment Affairs and Tourism and Water Affairs have indicated that they have no objection to the continued importation of mercury by Thor Chemicals. The company and the government justified their actions by arguing that the shipments of mercury containing residues to the plant were classified as raw materials rather than toxic waste (Koch, Cooper and Coetzee, 1990).

Prior to 1990, very little was known about the total waste stream, and in particular hazardous waste, generated in South Africa. Therefore, the CSIR initiated a survey during which interviews were conducted with representatives of as many major waste generators as possible representing different sectors of industry. It was, however, clear from the outset of

the investigation that reliable data on current hazardous waste generation in South Africa would be difficult to obtain and even the costly survey planned was not going to give a very accurate assessment of the *status quo*. This was attributed to the fact that very few of the industries canvassed had any information at all about the quantities or composition of their waste streams.

As a first estimate of the quantities of hazardous waste generated in South Africa annually, a classification was made based on the definitions of the Basel Convention. This estimate indicated that the total hazardous waste stream amounted to 456,073 tonnes per annum. An alternative classification proposed by the CSIR, which was thought to provide a far more accurate estimate of the hazardous waste stream, showed an increased production of hazardous waste and is summarised in Table 1.2. This approach involved taking the universally accepted (but entirely qualitative) general definition of hazardous waste (see Section 1.1) and adding quantitative criteria to it to define the following five hazard groups of waste:

a. *High hazard waste* (Group 1) of first priority concern, containing significant concentrations of highly toxic constituents which are easily accessible, mobile or persistent in the environment and are bio-accumulative;

b. *Moderately hazardous waste* (Group 2) of second priority concern with highly dangerous characteristics which could be highly explosive, flammable, corrosive or reactive, or which is infective, or which contains significant concentrations of constituents that are potentially highly toxic but only moderately mobile, persistent or bio-accumulative, or that are moderately toxic but are highly mobile, or persistent in the environment, or bio-accumulative;

c. *Low hazard waste* (Group 3) of third priority concern, which is moderately explosive, flammable, corrosive or reactive, or contains significant concentrations of constituents that are potentially harmful to human health or to the environment;

Table 1.2 The Estimated Amounts of Hazardous Waste (Groups 1 to 3) Generated in South Africa (Department of Environment, 1992a)

Waste Types	Hazardous Waste	
	$t\ y^{-1}$	% of total
Air Emissions		
Inorganic	136. 725	7.2
Organic	204, 943	10.8
Wastewater		
Inorganic	1. 081, 876	57.2
Organic	34. 505	1.8
Mixed	3. 393	0.2
Liquids		
Organic	14, 606	0.8
Emulsions		
Organic	1. 840	0.1
Tars		
Organic	5. 005	0.3
Slurries		
Inorganic	24, 812	1.3
Organic	0	0
Sludges		
Inorganic	296, 412	15.7
Organic	975	0.05
Mixed	20. 846	1.1
Solids		
Inorganic	30, 066	1.6
Organic	36. 727	1.9
Slags	0	0
Ash	0	0
Tailings	0	0
Rubble/Spoil	0	0
Total	1, 892. 681	100

d.   *Potentially hazardous waste* (Group 4) which often occurs in large quantities and which contains potentially harmful constituents in concentrations that in most instances would represent only a limited threat to human health or the environment; and

e.   *Non-hazardous waste* (Group 5) which, at most, contains only insignificant concentrations of harmful constituents.

The criteria used for the hazard rating can broadly be divided into the following two categories:

a.   *Danger* criteria are those that relate to chemical and other properties of the waste which are either known or can be quantified with one or more relatively simple test procedures, and about which consensus is likely to be reached fairly easily. Characteristics of these criteria are explosivity and flammability as well as corrosivity and chemical reactivity; and

b.   *Toxicity* criteria relate to properties which are not nearly as simply quantified, for which easy and rapid tests are not available, and which require professional assessment. The criteria grouped broadly under this category range from acute and chronic toxicity to human populations (as estimated by mammalian toxicity data) to direct and indirect effects that different substances can have on natural physical, chemical and biological processes in the biosphere. Further, the infectious and health (carcinogenicity and mutagenicity) implications of any given chemical were also taken into consideration.

From Table 1.2 it can be seen that the total waste produced in South Africa is dominated by a few large and relatively non-hazardous streams. Some of these are effectively inert mining spoils and overburden some of which contain small traces of toxic components (gold mine tailings), or traces of leachable heavy metals (coal ash). These few, but very large, waste streams distinguish the South African total waste stream from that of most other countries. Further, it is shown that 59% of the hazardous waste stream is carried in wastewater with 93.6% of this figure accounted for by cyanide-containing effluents from gold mining (Department of Environment, 1992a). An example of the deleterious effects of mine tailings was highlighted by the *Weekly Mail* newspaper in July 1990 which investigated a river flowing through the old mine dumps in the Crown Mines

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suburb of Johannesburg and then through Soweto. It was found that the dumps were leaching a staggering amount of toxins into the stream which, as a result, contained (l^{-1}) 8 g SO_4^{2-}, 520 μg cyanide and 60 μg arsenic and had a pH of 2.58. Further, tests made by the Rand Water Board on toxins leaching into the Rietspruit river from adjacent mine dumps, revealed alarming concentrations of mercury, arsenic and sulphate. Unfortunately, toxic run off from old mine dumps is not confined to the Witwatersrand as a pristine dam in Barberton (Eastern Transvaal) supported no aquatic life due to the increased arsenic and cadmium concentrations (Koch *et al.*, 1990). Also of interest is the estimate in Table 1.2 that 18% of the hazardous waste stream is emitted to the atmosphere.

A further cause for the uncertainty in calculating the total hazardous waste stream was the discovery that in many sectors of industry waste materials have been stored, very often for reuse or for suitable treatment once the technology or facilities become available (Department of Environment, 1992a). These, so called, backlogs were recorded for many of the industries surveyed and included mainly sludges from the textile and leather industries as well as extremely hazardous pesticides and polychlorinated biphenyls (PCBs).

In most industrialised countries, such as the U.S.A., U.K. and Germany, a large portion of the hazardous waste generated is reclaimed through recycling by industry. In Germany, for example, the total quantity of industrial waste generated in 1975 was 119 million tonnes. This figure increased until, in 1980, it was 203 million tonnes. For the next four years there was a slight decrease in the amount of waste produced to about 200 million tonnes. This decrease was mostly due to active recycling and waste avoidance (minimization) policies which resulted in 16% of the industrial wastes being recycled in 1990 (Stegmann, 1990).

In comparison, South Africa maintained a national recycling average of 3.1% for the total quantity of refuse generated in 1990 (Verrier, 1990). The CSIR survey team reported that significant quantities of materials such as paper, glass and metals are recycled in South Africa but only very few companies practise hazardous waste avoidance (Department of Environment, 1992f).

1.3   The South African Situation

In 1984 the U.S.A. Congress amended the environmental legislation and increased the federal role in solid waste regulation. These amendments (The Hazardous and Solid Waste Amendments of 1984) directed the Environmental Protection Agency to revise the criteria for waste facilities. At a minimum, these amendments require the EPA to develop enforceable standards for:

a.   Groundwater monitoring which is adequate to detect contamination;

b.   The location of new and existing facilities; and

c.   Corrective action.

Further, requirements proposed in the EPA's March 1987 draft also include locations, standards and leachate collection and removal systems (Pacey, 1989a).

At the beginning of the 1970s there were few specific controls for the disposal of industrial wastes in the U.K. Thus, there was little record of how wastes were being disposed of, or of their environmental effects. Therefore, the following legislation was enacted which provided a framework for effective regulation:

a.   The Deposit of Poisonous Wastes Act 1972 (subsequently replaced by the Special Waste Regulations, 1980) introduced a system of compulsory prior notification of any transport of difficult wastes. This began to generate national data on the production, movement and disposal locations for industrial wastes; and

b.   The Control of Pollution Act 1974 created the Waste Disposal Authorities and introduced a system of site licensing. The licensing of existing landfills (about 3,500) was largely complete by the late 1970s (Robinson and Gronow, 1992).

In South Africa, the situation appears to be slightly more ambiguous. Stander (1992), for example, reported that there is no all inclusive national policy which governs refuse disposal in South Africa. The result of this is a fragmented strategy with no all embracing legislation governing all dimensions of refuse disposal. In particular, the question of hazardous waste disposal has received little attention (Lombard, Botha and Rabie, 1992; Stander, 1992). Further, the CSIR report revealed that an assortment of provisions dealing with waste on land is to be found scattered among at least 37 Acts of Parliament, 16 Provincial Ordinances and numerous local by-laws (Lombard et al., 1992; Department of Environment, 1992d). The possibility exists that this number could increase even further following the new political dispensation in South Africa and the formation of new provincial governments. Also, the CSIR survey team concluded that, by 1992, of the 547 existing landfill sites only a minority had been subjected to state control and that industrial waste was frequently disposed of in sites unsuited for the purpose. Further, it was attempted to calculate a simple mass balance of inputs and outputs although, in most cases, the waste generators concerned had never previously carried out such a procedure and had no figures available on waste generation. Also, it was found that very few waste generators had any information on the composition of the waste or of the real costs of treatment and disposal (Department of Environment, 1992a).

As a consequence of this unsatisfactory situation, the Department of Water Affairs and Forestry has endeavoured to upgrade the standards for waste disposal in South Africa with the publication and enforcement of the Waste Management Series.

In South Africa, legislative powers for the prevention of environmental pollution, as a result of disposal of refuse in landfill sites, are vested in the Minister of Water Affairs and Forestry (Ball and Bredenhann, 1992). In 1992, the Department of Water Affairs and Forestry (DWA/F), as custodians of South Africa's water resources, instigated a proactive

programme to ensure that all refuse disposal is done in an environmentally acceptable manner. In this context DWA/F formulated a set of Minimum Requirements for refuse disposal facilities. The Minimum Requirements represent a means of distinguishing between the acceptable and the unacceptable so that refuse disposal in South Africa can be raised to an appropriate standard. This document is a "living document" in that it will always be subject to validation and updating (Ball and Bredenhann, 1992).

The Minimum Requirements for the Handling and Disposal of Hazardous Waste (1994) accepts that improved standards will inevitably result in increased costs. Consequently, the Best Practicable Environmental Option (BPEO) approach has been adopted to provide affordable environmental protection. This is achieved by the promotion of a waste management process which comprises three basic steps: waste avoidance; correct classification; and minimum requirements for the safe handling, treatment and disposal of hazardous waste. Further, it was recognised that costs have, in the past, been externalised as social and environmental costs as a result of the absence of standards. It is now envisaged that these costs will be internalised in accordance with the polluter pays' principle. An Integrated Environmental Management approach has also been adopted throughout which implies advance planning for all aspects of waste management. As a part of this planning approach an environmental impact assessment is essential before any landfill is issued with a permit (Anon, 1994a).

The first part (Volume 1) of the Waste Management Series, produced by the Department of Water Affairs and Forestry, was published in 1994 and thus far comprises.

a.   *Document 1* - Minimum Requirements for Waste Disposal by Landfill.

b.   *Document 2*: Minimum Requirements for the Handling and Disposal of Hazardous Waste; and

c.   *Document 3*: Minimum Requirements for the Monitoring at Waste Management Facilities.

The Minimum Requirements are enforceable nationally by means of the site permitting system, and by the provision in the Environmental Conservation Act of 1989 which states that no person shall discard waste or dispose of it in any other manner except at a permitted waste disposal site. The regulatory system, in the near future, will be further enforced by means of the registration of generators and transporters of hazardous waste.

In *Document 2* a procedure is described whereby any waste product can be identified and classified as hazardous by comparing it to the South African Bureau of Standards (SABS) Code 0228 which uses the International Maritime Dangerous Goods Code as its base. The disposal option can then be determined by placing the waste in one of 9 classes according to SABS 0228. The detail of the nine classes and all the relevant subdivisions have been described (Anon, 1994b) and will not be further discussed here. Waste which has been classified as Class 6 (poisonous and (toxic) and infectious substances) must further be rated according to a hazard rating which takes into account the acute and chronic toxicity and its environmental fate (Anon, 1994b) In this way, 50 examples have been given which extend the SABS Code 0228 which, when complete, will provide an easy and convenient means of identifying and classifying substances for waste disposal. It is interesting to note that according to this classification, wastewater effluents, such as sewage, are not classified as hazardous waste streams for disposal purposes because they are regulated and controlled by the Water Act (Act 54 of 1956). Similarly, radioactive waste is regulated and controlled by the Nuclear Energy Act (Act 92 of 1902) and the Hazardous Substances Act (Act 15 of 1973).

1.4   Treatment Methods

There is a wide range of technologies available for the treatment of hazardous waste and most treatment technologies can be readily adapted and modified to the requirements of a particular waste stream. The main objective of treating a waste is to either destroy or reduce the toxicity of the harmful components and, therefore, to minimise the potential inimical impacts on the environment. A further benefit of waste treatment is the possible

recovery of materials during waste minimisation and/or recycling programmes. Available technologies for the treatment of hazardous waste can be conveniently classified as:

a. Thermal treatment;

b. Chemical treatment;

c. Physical treatment; and

d. Biological treatment (Watson-Craik, 1987; Knoll and Winter, 1987; Department of Environment, 1992b; Behrmann and Hall, 1992).

1.4.1 Thermal Treatment

Thermal treatment can involve any one or a combination of the following (Lord, Ahrens, Tworeck and Rabie, 1983; Cope 1983a; Miller and Miller, 1991; Lombard *et al.*, 1992; Department of Environment, 1992b):

a. Sterilization. Normally sterilization is effected by raising the temperature to 120-140°C for up to 30 minutes. Any potentially infectious materials, especially waste from hospitals, should be sterilized if they are not to be incinerated. In sewage treatment plants, heat treatment is sometimes used to condition and sterilize raw or activated sludge;

b. Pyrolysis. This is thermal degradation of molecules and organic material in the absence of oxygen. The process is usually carried out at atmospheric pressure. Incineration always includes pyrolysis as a first step, and the resulting products are then ignited and burned. Pyrolysis itself is not always fast and may need an extended residence time. This is particularly important for polychlorinated biphenyls, pesticides and other chlorinated hydrocarbons. Polychlorinated biphenyls, with their >200 components, can be more safely pyrolysed for 10 seconds at lower temperatures (1100°C) than for 1-2 seconds at much higher temperatures. If incineration oxygen (air) is present at too early a stage of pyrolysis some stable and inimical compounds may be generated. For example, in the

incineration of polychlorinated biphenyls, or chlorlignin from pulp bleaching, highly toxic substances (dioxins) can be generated. An emerging method that holds great promise is plasma arc technology. Solid or liquid wastes are pyrolysed into combustible gases by exposure to a plasma gas. The super hot plasma gas results from exposure to high-energy radiation and has localised temperatures ranging from 30 000°C to 50 000°C. The plasma arc can destroy the molecular structure of the waste without any traditional chemical reactions at all. A further benefit is that the process is compact and the development of mobile units is likely (Goldman, Hulme and Johnson, 1986).

c. Incineration. This involves the aerobic combustion of the waste material. Incineration has also been described as an ultimate disposal process when applied to certain wastes that cannot be recycled, reused or safely disposed of in a landfill site (Batstone, 1989). When burning harmless material, it also provides a cheap support flame for material which is not capable of self-sustained combustion. Hazardous waste normally has to be incinerated at extremely high temperatures, with a residence time of 1-2 seconds, in order to break down the thermally stable compounds and the problematic secondary materials formed. The greatest difficulty in operating an incineration facility is to meet the stringent standards required by air pollution permits (Egarian and Platt, 1985). In the U.S.A., for example, federal standards require destruction, by incineration, of key organic species at the 99.99% level. This includes not only the original organic material but also all "daughter" products of incineration (Lederman and La Grega, 1981). Despite this, internationally, after landfilling, incineration is the most preferred treatment/disposal option for organic hazardous and toxic wastes (Willetts, 1983a). Further, legal requirements in the U.S.A. restrict the range of hazardous waste that can be landfilled and increase the cost of landfill disposal. This has made thermal treatment more financially competitive and, indeed, the sole option for certain wastes.

Not only are there very few incinerators capable of burning hazardous waste in South Africa but, as can be seen in Table 1.3, incineration is also economically extremely unfavourable (Department of Environment, 1992b).

Table 1.3   Relative Costs of Different Disposal Options Utilised in South Africa

Technology	Relative Costs
Waste disposal at regional attenuation sites	1
Pre-treatment and co-disposal in containment facility	4-12
Fixation and co-disposal in containment facility	5-15
Pre-treatment and disposal in secure isolation facility	20-70
Incineration	100-200
Encapsulation and placement in containment facility	200-250

Unfortunately, incineration has often been viewed as a "magic wand" which can quickly and conveniently dispose of any hazardous material. In reality, however, the successful operation of a hazardous waste incinerator plant is as strongly dependent on its operation as the actual design of the facility. Incorrectly operated, or applied to the wrong type of waste, incineration could lead to the generation of other secondary wastes which could be even more hazardous than the original waste fed into the unit (Department of Environment, 1992b). For example, wastes which contain inorganic salts and/or halogen compounds demand high energy scrubbers for their removal from the exhaust stream (Willetts, 1983a). According to Linde (1994), the Air Pollution Control Section of the Cape Town City Council has always adopted a policy to discourage waste incineration as far as possible. Despite the possibility of toxic emissions, incineration of waste is an extremely expensive option and, therefore, it is doubtful wether significant use will be made of this option in South Africa in the near future.

Selected hazardous wastes are being burned extremely successfully as fuels in more than 25 cement kilns in the U.S.A. There are a number of advantages to

cement kilns including the high temperatures (up to 1500°C) that are required for the manufacture of cement clinker, the long residence times (up to 6 seconds), the automatic scrubbing of noxious gases such as hydrogen chloride from the gas stream by the alkaline conditions in the kiln, and the savings in conventional fuels such as coal (Anon, 1994b; Jones, 1994).

1.4.2 Chemical Treatment

Chemical treatment is, generally, used more to reduce the toxicity of hazardous components than as a means of disposal *per se* (British Medical Association, 1991). Specific chemical processes include the following (Goldman *et al.*, 1986; Batstone, 1989; Miller and Miller, 1991; Anon, 1994b):

a.   Neutralisation. Acidic effluents, for example, are mixed with alkaline wastes (lime or caustic salts) to adjust the pH to near neutral;

b.   Precipitation. Removal of metals as their hydroxides renders process wastewater, after settlement, suitable for discharge to sewer;

c.   Oxidation-Reduction. These reactions are frequently used to detoxify compounds. Oxidation, for example, is used to facilitate the removal of cyanide from metal plating wastes. Reduction reactions are particularly important in the treatment of chromium wastes (Cope, 1983b; British Medical Association, 1991; Miller and Miller, 1991; Department of Environment, 1992b); and

d.   Electrolysis and Hydrolysis. In the former, an electric current is passed through a solution and the positive ions (cations) go to the anode where they are reduced and the negative ions (anions) go to the cathode where they are oxidised. Hydrolysis involves the addition of the elements of water to an organic compound (Anon, 1994b).

1.4.3 Physical Treatment

Physical treatment processes often either follow or precede some form of chemical or thermal treatment as physical treatments *per se* can not render a waste non-hazardous (Willetts, 1983b; Department of Environment, 1992b).

a. Sedimentation and Filtration. These methods are used to separate solids from liquids (Miller and Miller, 1991). Filtration is accomplished with microscreens, diatomaceous earth filters, sand filters and mixed media filters. Sedimentation on the other hand relies on gravity to remove suspended solids from an aqueous stream (Batstone, 1989). The addition of a flocculant, which increases the size of small suspended particles, is often used to increase the effectiveness in the sedimentation of particles (Anon, 1994b);

b. Absorption and Adsorption. Isolation of a gas component from other gases, liquids or solids can be effected by adsorption on and absorption into solids. Gases can also be displaced from liquids or removed by air stripping (Department of Environment, 1992b). For example, a commonly utilized method of removing H_2S and other trace compounds from gas streams is the use of activated carbon columns (Stegmann and Spendlin, 1989). The high efficiency of activated carbon is related to its high surface area and its affinity for certain, mainly, organic compounds (Schumacher, 1983); and

c. Distillation and Evaporation. Liquid distillation is a procedure that is often used to separate components of liquid waste streams into two or more fractions (Miller and Miller, 1991). Evaporation, on the other hand, is simply the vaporisation of a liquid from a slurry and, therefore, results in a more concentrated slurry and reduced volumes (Goldman *et al.*, 1986).

1.4.4 Biological Treatments

In microbiological waste treatments the hazardous wastes are catabolized or, better, mineralised. As a result, free energy is released and can be used, in part, by the microorganisms for their metabolic needs. Thus, in essence, complex waste molecules are gradually broken down through a series of chemical reactions (Howgrave-Graham, 1995).

The biotechnology of microbial attenuation originated in the activated sludge process for sewage treatment which was developed around the turn of the century (Miller and Miller, 1991). Unfortunately, as industries developed, the chemical structures of waste molecules became increasingly complex. Fortunately, microorganisms, with few exceptions, have been able to adapt to most new challenges which have appeared. This is possible because the genes responsible for microbial degradation of synthetic chemicals (xenobiotics) are located on extra chromosomal elements, the degradative or catabolic plasmids. These plasmids can be transferred from one bacterium to another and, thus, increase the catabolic potential of microbial associations (multi-species gene pools) towards different molecules (Müller, 1992; van der Meer, de Vos, Harayama and Zehnder, 1992).

For convenience, microbiological waste treatments can be classified into two main types, aerobic and anaerobic, although the two are often used in series, possibly in conjunction with physico-chemical treatments (Senior, 1990a).

Aerobic Treatments

Within certain limits, water authorities in the U.K. have been encouraging the discharge of industrial process water to public sewers since they believe that it is in the best interest for water pollution control (Harkness, 1984). Generally, the mixing of high-strength industrial wastewater with domestic sewage (in a ratio of 5:95 v/v) is acceptable although problems such as poor settling and clarification or sludge bulking can result (Senior, 1990a). These problems, together with the fact that there may not be sufficient capacity available for high volume discharges to sewer, have led to the development of

localised treatment plants on site. Fortunately, many wastes, such as, for example, landfill leachates, can be satisfactorily treated without involving the use of expensive plant equipment. Leachate treatment by recycle through the refuse mass, aerated lagoons and reed-beds are just three low-cost options (Doedens and Cord-Landwehr, 1989; Pohland, 1989a; Robinson, 1990; Robinson, Barr and Last, 1992).

Treatment plants can be designed for partial treatment prior to sewer discharge or for full treatment. For either option various treatment methods exist (Senior, 1990a).

Fixed Film Reactors

Trickling Filters

The traditional low-rate trickling filter is the most widely used fixed biomass process. The filter consists of beds of crushed rock (clinker, granite, basalt or slag) on top of which the liquid containing organic matter is sprayed. The mode of operation is counter current with a flow of air upwards. As the liquid trickles slowly through the high surface area bed, which has a high void space to facilitate the movement of air and liquid, the organic matter contacts the pre-established microbial film (Montgomery and Montgomery, 1994). Stone packings are presently being replaced with plastic media biological filters which have higher voidage and surface areas (Sarner, 1980).

Rotating Biological Contactor

The basic configuration comprises a series of discs (2-3 m diameter) mounted on a shaft which is driven so that the discs rotate at right angles to the flow of effluent. The discs, which are usually made of plastic, are located so that about 40% of their area is submerged at any one time. The process is, therefore, one of alternating absorption of pollutants and then of oxygen by the biofilm on the discs. It is important that the film is protected from rotational shear by limiting the speed of rotation to 0.5 - 10 revolutions per minute. During operation, excess biofilm sloughs off and settles to the bottom of the bioreactor (Forster, 1985).

Upflow Filter

The basic concept is that of a packed bed reactor (using stone or plastic media) in which there is an upward flow of liquid so that the support medium is totally submerged. Aeration is achieved either externally or by diffused air at the base of the reactor (Senior, 1990a).

Homogeneous Reactors

Activated sludge

The most common type of homogeneous reactor is the activated sludge process. In this treatment, the surfaces are provided by the organisms themselves which are closely linked in an association called a floc. These flocs can vary in size from small (comprising only a few bacteria) to a large floc which can include many millions of bacteria. The formation of heavy, rapidly settling, flocs is essential for the efficient operation of activated sludge treatment systems (Montgomery and Montgomery, 1994).

The components of an activated sludge plant are the aeration tanks in which the biological oxidation of the waste takes place, the settling tanks for the recovery of the activated sludge, and a system of pipes and pumps to return the activated sludge to the inlet end of the aeration tank (White, 1978).

A completely mixed or plug flow activated sludge process, which can take between 4 and 24 hours to complete, depending on the type of wastewater, is often preferred to a fixed film reactor since the effects of shock loading may be minimised and a maximum organic loading rate may be maintained in all parts of the process. Often in the presence of excessive organic loadings, filamentous microbial growth may result which has poor settling properties. In addition, this phenomenon of "bulking" may be accompanied by the development of odours.

Immobilised Cells

Immobilisation of microbial cells represents the transfer of cells from the free state to a state in which they are confined in a defined region with the retention of catalytic activity and with the retention of viability so that they can be used repeatedly or continuously (Senior, 1990a).

Although immobilised cells carry out multi-enzyme reactions as easily as free cells, they are present in much higher initial biomass concentrations and, consequently, the reaction or processing times are much faster than with free cells.

Once immobilised, the major problem, particularly for gel immobilised cells, is the transport of reactants into the gel. This is limited by the double diffusion gradient which builds up, one in the gel matrix and the second from the gel to the cell. As a consequence of this diffusion barrier, the cells in a gel matrix can be faced with a multitude of localised micro-environments with differences in oxygen concentration, pH and substrate/product concentration. The net result is the production of a very heterogeneous population (Senior, 1990a). This immobilisation has certain advantages such as:

a.   The cells are prevented from contaminating the product or effluent;

b.   The stabilities of the cells are increased; and

c.   Immobilised cells can be evenly distributed throughout the reactor so ensuring an even supply of substrate to each cell (Cheetham, 1983).

Anaerobic Treatments

According to King, Long and Sheldon (1992), anaerobic processes have a number of advantages over aerobic treatments:

a.   High production of biomass which can generate saleable by-products such as methane;

b.   Excellent biomass retention;

c.   Minimal nutrient consumption; and

d.   Good resistance to high organic loadings.

Unfortunately, anaerobic processes have the major disadvantages of slower rates of reaction, odour production and a limited number of anaerobic pathways (King *et al.*, 1992). It is, however, becoming more common to design two bioreactors in series, with the first unit operated anaerobically (for example, to dehalogenate a compound) and the second reactor operated aerobically (to mineralise the resulting metabolic by-products) (Balba, 1993).

Anaerobic Bioreactors

The simplest anaerobic digester design is the septic tank and it is still used as a cheap device for treatment of domestic sewage, especially in rural areas.

Stirred Tank

These bioreactors have been used for domestic, agricultural and industrial wastewaters. These simple flow-through anaerobic digesters are, however, generally used for strong industrial wastewaters from the food and beverage industries. There is an increasing interest in the use of two-stage systems in which acidogenesis is isolated in the first vessel. Unfortunately, provision of the required solids retention time (SRT) in a suspended growth system often necessitates a very large reactor (Bitton, 1994).

Anaerobic Contact Process

To obviate the above size requirement, biomass recycle may be considered (Schink, 1988). Typical designs incorporate a primary reaction tank with the overflow connected to a settling tank which concentrates the sludge which is recirculated to the primary digester to maintain high biomass density.

Upflow Anaerobic Sludge Blanket (UASB)

This is a unique type of system that is compartmentalized and is capable of handling a wide variety of sludge characteristics presented by a given waste stream. Usually, there are three compartments, the sludge bed, the sludge blanket and the separation zone (King *et al.*, 1992; Lettinga and Hulshoff, 1992; Bitton, 1994).

The sludge bed lies in the base of the reactor and the waste is passed through this under minimal agitation. Uniform distribution of the sludge and waste ensures intimate contact and maximum treatment. This compartment accounts for about one third of the reactor volume and effects most of the treatment because it is the point of maximum contact between the waste, the bacteria and the nutrients. Rising gas bubbles produced during treatment serve as a natural mixing mechanism.

The second compartment is the sludge blanket which occupies about 60% of the reactor volume. The blanket contains highly flocculated sludge and minute gas bubbles which facilitate ideal mixing.

The separation zone accounts for the balance of the volume within the reactor. This area has a capture apparatus which collects the gas and releases any biomass which may be attached to the gas bubbles. Also, other solids are removed and the treated effluent is allowed to exit the reactor (King *et al.*, 1992).

These units are known for high removal rates of $\geq 90\%$, short residence times of the order of 3 hours, and conversion reactions that reduce organic loadings as well as generate methane gas. A further advantage is that they do not have any internal moving parts and are very energy efficient.

Upflow and Downflow Anaerobic Filters

Both these bioreactor types use a packing material which has a very high surface area to facilitate biomass attachment. The downflow system has the added advantage that

the biogas rising against the flow can aid effective distribution without the need for an expensive distribution arrangement (Howgrave-Graham, 1995; Hall, 1992).

Fluidised Bed

In these reactors a continuous fluid phase is passed up from the base through a bed of particulate medium (sand, gravel or plastic) which is then suspended in the column with an upflow stream of liquid at a linear flow velocity which must be greater than the settling velocity of the particulates (Forster, 1985; Hall, 1994; Leach, 1994b).

The efficacy of these units has been demonstrated at many locations internationally and they have been operated in aerobic, anaerobic and facultative anaerobic modes.

1.5   Waste Disposal Methods

1.5.1   Marine Disposal

Sea disposal of raw sewage, radioactive wastes, mine wastes and industrial effluents, including heavy metals and solvents, has often been thought to be the ultimate "dilute and disperse" option (Willetts, 1983b; British Medical Association, 1991). For example, in 1981 about 30% of the sewage sludge produced in the U.K. was disposed of to sea (Portmann and Norton, 1981). Reliance upon the natural marine environment to accommodate industrial pollutants and to dilute them sufficiently and to alter them chemically or biochemically to non-polluting forms is the basis, but increasingly false premise, of this practice. Marine disposal can be accomplished in a number of ways such as (Department of Environment, 1992b):

a.   Discharge via submerged pipelines;

b.   Incineration and dumping;

c.   Dumping from vessels and barges; and

d.   Encapsulation and burial in deep sea sediments (Department of Environment, 1992b).

In general, however, the practice of marine disposal is declining and is being actively discouraged by International Conventions (Wu, 1987). This is exemplified by, for example, the U.K.'s commitment to cease ocean dumping by 1998 (Sinclair, 1994). In the U.S.A., New York City has, following an intense microbial study, ceased all disposal of sewage sludge at sea. The city is now using a combination of landfilling and other beneficial uses as alternatives (Anon, 1994d). This is also as a direct result of the Ocean Dumping Ban Act of 1988 in which the U.S.A. Congress amended the Marine Protection, Research and Sanctuaries Act of 1972 to prohibit all dumping of sewage sludge or industrial waste in the ocean after December 31, 1991 (Bastian, Farrell, Granato, Lui-Hing, Pietz and Southworth, 1992). Japan, however, is expected to continue dumping industrial and sewage sludges into the Pacific Ocean even after the amended annex of the London Treaty on Industrial Waste comes into effect in 1996. As a result, Japan has come under severe criticism from the Organisation for Economic Co-Operation and Development (Anon, 1994e).

At present, South Africa does not dispose of waste by means of incineration at sea or dumping from vessels. There are, however, 63 pipelines situated around the coast of South Africa, discharging a total of 760 Ml d^{-1}. Of these pipelines, 22 discharge sewage, 31 discharge industrial effluents and 10 discharge mixed effluents. In total, the pipelines account for 85% of the total marine discharge (Department of Environment, 1992b). According to Russell, K.S. (1992), marine outfalls have been scientifically proven to be a safe and viable alternative for sewage disposal because discharges are safely dispersed in the sea. Discharging by pipelines which extend for ≥ 1 km out to sea provide rapid mixing and high dilution of pathogens to very low levels (Russell, K.S., 1992). Of the 63 pipelines in South Africa, only 8 are longer than 500 m (Department of Environment, 1992b). It is to be expected, however, that international pressure against all forms of ocean dumping will soon manifest itself in South Africa and result in a decreasing dependence on this disposal option.

1.5.2 Land Treatment

Land treatment can justifiably be considered a disposal option as once the waste has been emplaced no further disposal is needed as the treatment allows for the simultaneous treatment (attenuation) and disposal (final storage) of the waste. This treatment, often referred to as land farming, comprises the mixing of sludges into the top surface of the soil and exploits the natural capacity of the soil and microorganisms to degrade and attenuate the added compounds. The method was developed for refinery sludges in the U.S.A. and the land is not subsequently used for agriculture (Batstone, 1989). This method, therefore, differs from the practice of applying sewage sludge to farmland for use as a fertiliser (see Section 1.8.12). The application to land of sewage sludge is a growing practice and is shown in Table 1.4 for the U.S.A. It is of interest to note the decrease in the relative percentage of the total sludge mass disposed of to the marine environment (Kama, 1992).

Table 1.4 Estimated Percentages of the Total Sludge Mass Disposed of by Different Methods in the USA (Kama, 1992)

Method	1976	1978	1981
Land Application	25	31	42
Landfill	26	29	15
Incineration	35	22	27
Ocean Disposal	15	12	4
Other (Lagoons)	-	6	12

Included in the definition of land treatment is the process of composting (Wilson, Parr, Taylor and Secure, 1982). In aerobic/thermophilic composting, biological reactions occur at temperatures above normal which makes this technique a favourable degradation system for hazardous wastes.

1.5.3 Immobilisation, Solidification and Encapsulation

These processes can also be considered as disposal options as more emphasis is placed on isolating the waste from the environment than treatment *per se*. To isolate a toxic

liquid waste from the environment the process of immobilisation or chemical stabilisation is often used which converts the waste to a more chemically stable or immobile form (Anon, 1994b). Solidification or cementation, on the other hand, involves the full or partial bonding of the organic waste by the addition of a supporting medium (binder). The chief affect of this is to eliminate the leaching of pollutants and hazardous constituents (Tittlebaum, Seals, Cartledge and Engels, 1985). For example, the low-level radioactive treatment plant at Pelindaba first subjects the waste to chemical co-precipitation which is then followed by solidification and compaction before disposal (Hambleton-Jones, 1994).

Encapsulation usually involves the storage of waste in sealed containers which, in turn, are encased in reinforced concrete. In general, highly toxic and carcinogenic wastes which require safe disposal are encapsulated (Department of Environment, 1992b).

1.5.4   Underground Burial

The complex problem of burial waste site selection has been a subject of extensive study in recent years, not only for hazardous and toxic wastes but also for radioactive wastes (Miller and Miller, 1991). Clearly, detailed geological investigations must be undertaken to determine suitable sites for these wastes, as exemplified by the longevity of radioactive wastes (> 300 years) before the isotopes have fully decayed (Hambleton-Jones, 1994). Notwithstanding these concerns, deep geological disposal (> 200 m) of high-level radioactive waste is currently the most favoured method for a number of countries (Maré and Keenan, 1992).

1.5.5   Incineration

Incineration has been regarded as both an option for hazardous waste treatment and for disposal and has been previously discussed (Section 1.4.1).

1.5.6 Landfill

Despite the diversity in treatment and disposal methods, most wastes, both hazardous and non-hazardous, are disposed of by landfilling (Jolley, 1992). In South Africa, landfill is still the major treatment/disposal option for hazardous waste (Department of Environment, 1992b). In 1987, in the U.K., landfill accounted for no less than 83% of all hazardous wastes while marine disposal, chemical and/or physical treatment and incineration accounted for 8.7 and 2%, respectively (British Medical Association, 1991).

From a hydrogeological point of view, landfill sites have been characterised as follows (Senior, 1986; Senior and Balba, 1990):

a. *Containment Sites (Class 1)*. These sites minimise the leachate penetrating the groundwater by provision of an impermeable or semi-permeable liner which consists of either natural (clays) or synthetic materials. The actual choice of liners to effect this isolation of the landfill from the environment, as well as the problems associated with liner systems, have been described (Batstone, 1989; Workman and Keeble, 1989; Daniel and Shackelford, 1989; Anon, 1994a) and will not be further discussed here;

b. *Attenuation Sites (Class 2)* which constitute the majority of U.S.A. and U.K. landfills. In these sites the groundwater is protected from the slowly migrating leachate by physico-chemical and microbiological intervention (attenuation) (Senior and Balba, 1990). According to Robinson (1992), despite the lack of any serious groundwater pollution in the U.K., the gradually increasing understanding of the processes of attenuation based upon research and experience, and in spite of advances in the construction of engineered attenuation zones beneath landfill sites, which all demonstrate that the 'dilute and attenuate' philosophy still represents a technically valid concept, the strategy is unlikely to remain acceptable at new landfill sites largely for socio-political reasons; and

c.   *Rapid Migration Sites (Class 3)* afford little or no environmental protection since leachate rapidly migrates from the landfill with only limited attenuation (Senior and Balba, 1990).

Hazardous waste may, in South Africa, only be disposed of at a landfill designed specifically for its disposal and legally permitted by the Department of Water Affairs and Forestry, in terms of the Environmental Conservation Act of 1989. The Minimum Requirements for Waste Disposal by Landfill (1994) has advocated a different method for classifying landfills. The Landfill Classification System defines the disposal situation or need and identifies the type of landfill required to meet that need according to:

a.   Waste type;

b.   Size of waste stream or landfill operation; and

c.   Potential for significant leachate generation and need for leachate management (Anon, 1994b).

As a first step, wastes are categorised into two types: General and Hazardous. Landfills which can accept hazardous waste are classified as either H:h or H:H sites which, in turn, may accept waste with a hazard rating (see section 1.2) of 2,3 and 4 or 1 to 4, respectively. Further, because of the risks posed by these landfills to the environment and public health, they must be containment sites (Anon, 1994b).

Site Selection

Landfill siting criteria comprise both environmental (technical) and socio-economic (institutional) constraints. Environmental considerations relate to the potential threat to the receiving environment, specifically water resources. These include factors such as:

a.   Geographical considerations - site topography, drainage, soils and geohydrology;

b.   Geological considerations - proximity to known faults, potential for landslides, seismicity, etc;

c.   Waste characteristics - waste properties and volumes; and

d.   Management priorities - these generally involve the financing of the facility (Young 1982; Pearce, 1983; Ball and Bredenhann, 1992).

Geophysical considerations are becoming increasingly prominent in site investigations. For example, before planning approval of the second licensed Class 1 disposal site in South Africa, the waste site at Mossel Bay, was received, a detailed hydrogeological investigation, encompassing a geophysical survey involving electromagnetic and resistivity methods to identify geological features, had to be made (Dorman, McPhail, Geldenhuis and Hojem, 1992) Socio-economic considerations include aspects such as distance from waste generation areas, site size, land availability, access and adjacent land use as well as all legal considerations (Ball and Bredenhann, 1992), and have been fully detailed in Minimum Requirements for Waste Disposal by Landfill (Anon, 1994a).

Arguably, the most important factor of site selection and engineering is the protection of the water resources, particularly the groundwater. In "Attenuate and Disperse" sites the underlying geology is of critical importance. Ideally, geological formations for such sites are those with significantly high contents of clay minerals and where leachate movement will be through pores or micro-fissures (Robinson, 1989). An illustration of the effect of the underlying rock formation on leachate movement can be seen at the waste site at Ingham, East Anglia, U.K. This site is situated on Cretaceous Upper Chalk which is a micrite composed mostly of coccolith fragments 2μm in size. Even though the groundwater table is between 7 and 23m below ground surface, a site survey indicated high concentrations of mineral oils, phenolics and chlorinated solvent pollutants (Baxter, 1985).

To compound the problem, the deleterious effects of high strength organic leachate on groundwater may persist for protracted periods due to the limited concentration of dissolved oxygen available for microbial metabolism and the slow rates of dispersion. Once the groundwater has become polluted it may be unsuitable as a source of potable water supply for many years (U.K. Department of Environment, 1988).

Unfortunately, internationally, groundwater pollution has and still occurs at many landfill sites (Rudy and Caoile, 1984; Miller and Miller, 1991). For example, in a study made in Denmark in the early 1980's it was noted that out of 501 sites known to contain chemical waste, 380 required remedial measures to ensure that the aquatic environment was not damaged (Korkman, 1985). In an effort to nullify the hazardous effects of leachate, the U.S. EPA in 1991 officially adopted the "dry tomb" landfilling approach for municipal solid waste (MSW) management. This approach is the placement of untreated MSW in lined landfills that are eventually covered. The concept is based on the premise that if buried wastes can be kept dry, and thus not produce any leachate, groundwaters will not be polluted (Lee and Jones-Lee, 1993).

In an endeavour to protect the ground and surface water resources in South Africa, the Minimum Requirements prohibit the development of a landfill in an area that:

a.   Lies below the 1 in 50 year floodline, which eliminates wetlands, vleis, pans and floodplains;

b.   Is in close proximity to significant surface water bodies such as dams or water courses;

c.   Is intrinsically unstable, which would include fault zones, seismic zones, dolomitic or karst areas where sinkholes and subsidence are likely;

d.   Is a catchment area for important water resources;

e.   Is characterised by flat gradients, shallow or emergent groundwater;

f.   Consists of highly permeable soils or that are areas of groundwater recharge due to topography; and

g.   Is characterised by shallow bedrock with little soil cover (Anon, 1994a).

These situations may represent a fatal flaw but only in the sense that they prohibit the development of an environmentally or publicly acceptable waste disposal facility except at excessive cost (Anon, 1994a).

Liner Integrity

More and more reliance is being placed on natural and synthetic liners to contain leachate until it no longer poses any threat to the environment. For example, the pits at the Mossgas waste site (Mossel Bay) have been designed in accordance with U.S. EPA standards and comprise a double high density polyethylene (HDPE) lined system with a leakage detection layer between the two liners (Dorman *et al.*, 1992).

Emplacement

Although landfill practices vary from country to country one of the most common approaches is the use of the "cell" emplacement strategy. In this, the collected refuse is covered on all sides by soil at the end of each working day which results in an irregular stratification (Leach, Middelbeek and Mijnbeek, 1994). The size of the cells depends on the daily volume that is tipped and each cell is compressed and roughly levelled by mechanical bulldozers. Generally, the depth of each cell is limited to about 2 to 5 metres and the depth of covering soil used at the end of each day is usually about 20 cm. This practice is particularly effective in controlling vermin and insect infestation in addition to limiting the windborne spread of refuse (U.K. Department of Environment, 1988). Another method, which is widely used at sites within the U.K., is the pretreatment of household and commercial wastes by compaction into bales, with densities ranging from 0.75 t m^{-3} to > 1 t m^{-3} (Sinclair, 1994).

1.5.7 Co-Disposal

Co-disposal is often defined as the disposal of hazardous and non-hazardous wastes in the same refuse site. (Department of Environment, 1992b). Since the hazardous fraction consists mainly of industrial wastewaters and sludges the term co-disposal includes the joint disposal of liquid wastes with dry solid wastes. Co-disposal of hazardous waste can, therefore, be perceived as both a treatment process, which should, if correctly controlled, result in the mineralisation of the molecule(s), and a disposal process.

In the U.K., co-disposal has been officially endorsed by both the Department of the Environment (1987) and the Hazardous Waste Inspectorate (1986)(Watson-Craik, Sinclair and Senior, 1992a). These endorsements resulted from extensive investigations which started as early as the 1960's when the UK government appointed two specialist committees to undertake fundamental reviews. The results of both these reports motivated new legislation. In addition, the co-ordinated programmes of landfill research, which were initiated in 1973 (and still continue), have been instrumental in producing guidelines in the form of Waste Management Papers. Although the guidelines are still non-statutory, they do have considerable standing in a court of law (Robinson, 1992).

In its wide acceptance of the philosophy of co-disposal, the U.K. is at odds with most of its European neighbours and North America. Recently, at a meeting of the Council of Environment Ministers of the European Community, held in Luxembourg in June 1994, it was agreed that the Landfill Directive will allow the U.K. to continue to operate existing co-disposal sites for the remainder of their lifetimes, provided that the operations are closely supervised and that no adverse environmental affects are recorded. The development of new co-disposal sites is, however, forbidden (Anon, 1994f). At present, in South Africa, co-disposal is widely practised and, according to the Department of Environment Affairs and Tourism, where planned and properly controlled, can be very successful and cost effective (Department of Environment, 1992b)

Unlike many countries in Europe, South Africa has an abundance of relatively cheap land which can be made available for landfill facilities. Also, the apparent lack of efficient and cost effective alternatives together with the present belief that co-disposal is not necessarily detrimental to the environment ensures that landfilling, and especially co-disposal, is likely to remain the most widely used disposal method for the foreseeable future.

Requirements for Co-Disposal Operation

Correctly operated, co-disposal, as understood and accepted in the U.K. tries to

minimise the time of isolation but to maximise the rate of alteration and dilution. This implies that co-disposal must be practised in such a way as to enhance all degradative mechanisms but only to the extent that (Cossu, 1990):

a.   Inhibition of the normal refuse catabolism does not occur;

b.   There is no effect on leachate quality that would make its disposal more difficult or its environmental impact more severe;

c.   There are no unacceptable hazards to operators, visitors or site neighbours and public opinion must be satisfied (Watson-Craik, 1995); and

d.   Restoration, after-care and after-use are not significantly hampered (Cossu, 1990).

Despite the apparent advantages of co-disposal, the practice is discouraged, or even outlawed, in many countries, such as Canada, Australia, Germany and the U.S.A., where the landfilling of hazardous wastes is only permitted in dedicated and secure facilities. These regulations are due, principally, to previous co-disposal practices, their environmental impacts and resultant public opinion backlash (Watson-Craik et al., 1992a). For example, according to Batstone (1989), in 1969 a survey of the "Status of Solid Waste in California" found major deficiencies in the way in which wastes were managed. These deficiencies ranged from *inadequate planning, financial preparation and working standards to poorly developed technology and fragmented authority. Compounding the problem, it was found that very little was known about the volumes, types of wastes and ultimate disposal of hazardous wastes. Further, the wastes were being disposed of in private dumps on the premises where they were generated, placed in open dumps off site and dumped indiscriminately. In general, there was very little control of where and how these hazardous materials were disposed* (Batstone, 1989). Invariably, one or more of the above factors (a - d) is/are compromised and it is usually leachate quality.

Site Selection and Liner Integrity

The siting procedures, and in particular the role of the surrounding communities, for new landfills become even more important when designing a new co-disposal facility.

The empowerment of the general public and the effect that this can have on a disposal facility was also demonstrated in the new Waste-tech (Pty) Ltd hazardous waste site at Chloorkop, Kempton Park. The principles of integrated environmental management were not used to scope the project during the early planning stages since this was not required by existing legislation. This omission resulted in considerable difficulties with respect to community objections to the site. The commissioning of the landfill, after site preparation was completed in 1993, was thus delayed, and continues to be delayed, for more than two years while community objections were heard in a series of public hearings (Boswell, 1994). In this regard, it is interesting to note that by November 1995 the Institute of Waste Management had indicated that the Gauteng area was experiencing a waste crisis due to the protracted period for the commissioning of the Chloorkop site. This has led to what is viewed by many people to be an unacceptable situation, involving excess liquid wastes being disposed of at the only other available site (Holfontein landfill site) (Ball, 1995).

It is now widely recognised in the U.K. that landfilling and, more specifically, co-disposal should only take place within a "contained" environment. It is, therefore, essential that sites are adequately engineered to ensure the containment of generated leachate (Greedy, 1993). In contrast, until the introduction of the Minimum Requirements, there were no current standards available in the R.S.A. pertaining to the liners needed for hazardous waste sites. Therefore, the development of the Holfontein Class 1 site near Springs relied on experience gained overseas and opted for a double clay liner which incorporates a leachate leakage detection system (Jewaskiewicz, 1992).

The Minimum Requirements for Waste Disposal by Landfill now sets minimum standards for all landfill sites which generate significant volumes of leachate. These standards target leachate management and the construction of liners. Thus, the provision is made for a liner to be provided for every H site (sites receiving hazardous waste) regardless of whether it produces leachate or not (Anon, 1994a).

The confidence placed in natural (clay) and synthetic liners to contain leachate for indefinite periods has been questioned by many researchers (Pearce, 1983; Lechner, 1989;

Lee and Jones-Lee, 1993). For example, the hydraulic conductivity of a nearly flawless geomembrane is in the order of 1×10^{-12} cm s^{-1} as compared with 1×10^{-7} cm s^{-1} for a low permeability, compacted soil. Geomembranes are not perfect, however, since, even with careful construction, they may contain about two to five defects per hectare. These defects may result in an effective permeability of two to five orders of magnitude lower (Pacey, 1989a).

The swell properties of clay soils in contact with water and with organic solvents has been examined (Green, Lee, Jones and Palit, 1983). The octanol/water coefficient (log K_{ow}) was found to be an appropriate parameter for correlating the shrink - swell behaviour of the clay soils with the hydrophilic or hydrophobic nature of the solvent. It was found that hydrophilic solvents (solvents with a negative log K_{ow} value) caused the clay soils to swell (decreased permeability). Shrinkage and cracking of clay was observed in apolar, hydrophobic solvents. It is, therefore, clear that hydrophobic liquids can damage the integrity of landfill clay liners (Green *et al.*, 1983).

Emplacement

In practice, the co-disposal of liquids presents few problems since the waste is usually dispensed into trenches and shallow lagoons excavated in the domestic refuse. Likewise, sludges are often co-disposed in landfill sites either in trenches dug in MSW, where a methanogenic environment has established, or by spreading in thin layers prior to their incorporation (U.K. Department of Environment, 1988; Greedy, 1993). Trenches dug into the refuse mass should not be wider than one excavator bucket and the liquid level should not be allowed to reach the top of the trench at any time (Watson-Craik, 1990; Havinga, 1993). Hydraulic loading of the trenches should take into account the absorptive capacity of the surrounding refuse and at least three metres of refuse should be below the trench bottom. Trenches should also be backfilled with MSW and moved at regular intervals, depending on the quantity of liquid waste handled and the depth of the refuse in the site (U.K. Department of Environment, 1988; Havinga, 1993). The spraying of liquid waste, compared with subsurface introductions, facilitates greater evaporation and more

efficient dispersal over the site. This method is, however, only suitable for wastes which have a low toxicity and odour.

Where there are no safety problems, hazardous solids can be spread over the working face of the landfill and then covered with a layer of MSW. Compared with trenches, this has the added advantage that the waste is not concentrated in small pockets. One notable exception to this technique of spreading is the landfilling of hazardous solids such as asbestos. When this material is received on site it is buried in trenches and immediately covered with other waste (Parker and Williams, 1981). It is interesting to note that in the U.K. waste for co-disposal is only added to sites which handle a considerable quantity of MSW or similar waste. It is unlikely that a site accepting purely industrial waste would have sufficient substrate to provide the degree of activity required for effective co-disposal (Greedy, 1993).

1.6 Co-Disposal Operation and Potential Environmental Impacts

Co-disposal normally utilises properties inherent in MSW to attenuate the polluting and, potentially, hazardous components found in the wastewater or sludge. The key objective of co-disposal then is to take full advantage of all attenuation/containment mechanisms inherent in the landfill site to reduce the polluting potential to environmentally acceptable standards.

Co-disposal landfill sites are generally licensed to accept numerous different types of hazardous waste from diverse industries. An example of this is given in Table 1.5 which shows data obtained between 1978 and 1980 for the Stewarthy landfill site in Bedfordshire, U.K. (Knox, 1989).

The spectrum of liquid wastes and sludges has widened since the above analysis although the site licence prohibits the deposit of (Knox, 1989):

a.   Acids (pH < 4); and

b.   Soluble heavy metals, unless pre-treated to pH 8-11.

It also restricts;

a. Cr^{4+} (< 5000 mg l^{-1})

b. Cyanide   (< 100 mg l^{-1}); and

c. Phenols   (< 200 mg d^{-1} t^{-1} of solid degradable waste deposited that day).

Table 1.5   Different Types of Waste Disposed at the Stewartby Landfill Site During the Period 1978 to 1980 (Knox, 1989)

WASTE DESCRIPTION	% (w/w) of INPUTS
Tannery and Fellmongers Waste	23
Oil/Water Mixtures	17
Adhesive Waste	17
Miscellaneous Chemical Waste	11
Miscellaneous Waste	9
Tank Sludge/Interceptor Waste	7
Effluent Treatment Plant Sludge	5
Paint Waste	5
Other Waste	6

The operation of a co-disposal site as a multi-million m^3 anaerobic bioreactor could confer advantages similar to those offered by smaller-scale Downflow Stationary Fixed Film Reactors (DSFFR). These reactors have proved highly flexible, with successful operation between 10 and 55°C and a high tolerance of severe and repeated hydraulic and organic overloadings. Downflow stationary fixed film reactors can tolerate down-times of weeks or even months without a great loss in activity, particularly at temperatures of ≤25°C (Watson-Craik, 1987; Watson-Craik, Sinclair and Senior, 1992a). Further, the mean hydraulic retention time (HRT) of a landfill is often several years and, thus, very much longer than in conventional reactors. However, the mixing characteristics are poorly developed and may be highly variable with the possibility of short-circuiting or channelling in some circumstances (Knox and Gronow, 1989).

Various large, and well known, co-disposal sites in the U.K. were assessed to determine the effects of a balanced refuse fermentation (including physical, chemical and biological mechanisms) on the attenuation of many organic and inorganic components present in industrial wastes (Knox, 1989; Knox and Gronow, 1990). Analytical data from the Pitsea, Stewartby and Himlet Wood landfill sites, at which co-disposal practices have been in operation since the 1950's, 1978 and 1982, respectively, were examined. The results showed that no impairment of the leachate quality occurred thus indicating that the applied organic and heavy metal loadings had not disrupted the normal degradative processes. At all the sites, low total organic carbon (TOC) concentrations, together with BOD:COD ratios of <0.2, indicated fully methanogenic conditions. The concentrations of phenols, total cyanides and heavy metals were within the ranges which are typically found in domestic waste leachates (Knox, 1983; 1989).

Notwithstanding the encouraging results described above, it is clear that in many landfill sites, where co-disposal has been practised, leachate quality and refuse catabolism are adversely affected as indicated by, for example, the persistence of high concentrations (8 to 375 mg l^{-1}) of phenol in the leachates. At one site, the disposal of substantial volumes of acid wastes, particularly sulphuric acid, resulted in pH values of 1.3 to 1.8 and 2.0 to 2.2 in the aqueous phase of the lagoon and the saturated zone at the base of the site, respectively. Not surprisingly, no microbial activity was recorded in the saturated zone (Watson-Craik *et al.*, 1992a).

To accomplish the targets detailed in Section 1.4 clearly necessitates a thorough understanding of the attenuation mechanisms operative for specific wastes, the pertinent hydraulic and organic loading rates, and the interdependent effects of refuse metabolism and added xenobiotic. Further, as co-disposal is, fundamentally, a superimposition on to landfill catabolic processes, effective co-disposal must be assessed in terms of these targets (Watson-Craik, 1987). To date, however, development of effective co-disposal strategies has been constrained by several factors, such as:

a.   Paucity of research on microbially-mediated degradative processes in refuse.

Management of hazardous and, indeed, of all wastes, is very much the most recent sector of environmental control, with the U.K. fully investigating co-disposal options (Robinson, 1992). Senior (1990b) pointed out that, due to the complexity and heterogeneity of the landfill ecosystem, in addition to increasing amounts of wastes requiring disposal and to the decreasing number of suitable sites in the right places, research has focussed on the civil engineering aspects of landfill technology and fundamental microbiological and biochemical studies have largely been neglected. For example, although the aromatic molecule, phenol, has received the most attention so far, no reports of phenol co-disposal in full-scale landfills have been found (Knox, 1989).

b. The absence of information on the possible hazards of mixing wastes on site. When two or more wastes are to be deposited at the same location undesirable reactions can occur when mixing incompatible wastes. These include:

1. The generation of heat by chemical reactions which in extreme cases may result in fires or even explosions (e.g., alkali metals, metal powders);

2. The generation of toxic gases (e.g. arsine, hydrogen cyanide, hydrogen sulphide);

3. The generation of flammable gases (e.g. hydrogen, acetylene); and

4. The generation of gases such as nitrogen oxides, carbon dioxide and chlorine (Batstone, 1989).

Unfortunately, very little information is available on adverse reactions mediated by three or more wastes (Batstone, 1989). Further, Cook (1984) predicted that chemical reactions could take unexpected paths if the chemicals were in proximity for long periods in the presence of oxidising agents and if catalytic materials were present. Therefore, landfilling is not recommended for highly flammable materials, for very strong oxidising agents, for shock sensitive explosive compounds, for obnoxious smelling wastes, for very volatile substances of significant toxicity, for substances that easily react with water or dilute alkali and

acids to produce toxic gases, for concentrated acids or alkalis, or for solvents (Department of Environment, 1992b). A further major difficulty with disposing of radioactive, hazardous or mixed wastes in a given environment is predicting their behaviour several decades into the future on the basis of short-term tests (Godbee, Rivera, Kasten, Jolley and Anders, 1992). In order to overcome these potential problems, toxic wastes must be correctly identified and their disposal strictly controlled (Cossu and Serra, 1989).

c.   Apart from the numerous types of wastes co-disposed with MSW the problem is compounded by the fact that many co-disposal sites are licensed to take a wide range of wastes containing a large number of potentially polluting components (Batstone, 1989). In addition, the contribution of domestic refuse to the production of hazardous wastes is often overlooked. It has been estimated in the U.S.A., that between 0.0015 and 0.4 % (w/w) of MSW can be classified as hazardous waste (Pohland, 1989b).

        A further problem that microorganisms have to cope with is that many of the co-disposed xenobiotics are present in very low concentrations (Jacobsen and Pederson, 1992). Alexander (1973) proposed that the rate of microbial growth was at the expense of soluble chemicals present in low concentrations and may be proportional to their concentration, exactly as the rate of enzymatic reactions is governed by the substrate concentration (Alexander, 1973). Experimental data have indicated that these sub-maintenance concentrations generally fall below ng ml^{-1} (Lewis and Gattie, 1991). For example, the microorganisms of stream water have been shown to mineralize little 2,4-dichloro- phenoxyacetate (2,4-D) and 1-naphthyl-N-methylcarbamate at concentrations of 2 to 3 ng ml^{-1}, but they mineralized 60 % or more of the compounds in 6 days when they were provided with higher concentrations (Wang, Subba-Rao and Alexander, 1984); and

d.   Exploitation and enhancement of the refuse catabolic processes require an understanding of not only the inherent microbiology and biochemistry but also the

performance of the refuse fermentation as a whole. As discussed above, co-disposal of hazardous waste with refuse is a superimposition on the natural landfill catabolic processes. It is, therefore, necessary to appraise the efficacy of this practice, in relation to the refuse fermentation. In practice, however, it has proven difficult to select representative indicator parameters. Indices of refuse catabolism which have been used include:

1.   Leachate composition (Kasali, 1986; Senior, Watson-Craik and Kasali, 1990). This approach is, however, problematic as the type and age of refuse, as well as the stage of refuse fermentation, significantly affect the leachate constituents. Typically, anaerobically decomposing refuse initially displays a sharp decrease in the mean pH value as a result of the accumulation of reduced organic acids which inhibits the methanogenic population (Senior, Watson-Craik and Kasali, 1990). For this reason, most landfills have an acidic environment initially but, within the first few years, the pH rises towards neutrality (Emcon Associates, 1982). For example, at the Crompton Bassett landfill site in Southern England, U.K., the leachate exhibited a dramatic rise in pH from 5.7 to 7.9 as methanogenesis proceeded (Robinson, 1989).

A more meaningful parameter is the ratio between Biochemical Oxygen Demand (BOD) and chemical oxygen demand (COD). This ratio indicates the biological degradability of organic substances present in the leachate (Cossu, Stegmann, Andreottola and Cannas, 1989). The acidogenesis phase is characterised by high organic molecule concentrations with BOD:COD ratios > 0.4 and low pH, methane content and gas production. After the transition to the methanogenic phase the methane content and pH are high although the BOD, COD and the BOD:COD ratio are low (Ehrig, 1989). This reflects the higher proportion of semi-recalcitrant/recalcitrant compounds such as humic and fulvic acids in the leachate from "old" wastes (Watson-Craik, 1995);

2. Gas production and, more specifically, the methane content have been used as an indication of landfill/refuse stabilisation in many studies on control and optimization of refuse catabolism (Kasali, 1986; Barlaz, Ham and Schaefer, 1990; Senior, Watson-Craik and Kasali, 1990). Even though all the essential requirements are usually satisfied in an anoxic refuse mass, the internal landfill ecosystem is extremely dynamic and competitive as well as hostile to the methanogens (Farquhar and Rovers, 1973; Westlake, 1990). For the degradation of refuse to proceed at an optimum rate enhancement of the methanogenic process is vital. This is made extremely difficult by the close and complex interactions between the operating variables and microbial associations responsible for refuse decomposition (Senior and Balba, 1990); and

3. Temperature. The rate of heat production in a landfill is determined by the rate of decomposition of organic matter. Thus, the temperature attained by a landfill will be determined by the balance between the rates of heat production and addition and the rate of heat loss to the surrounding soil and atmosphere (Senior and Kasali, 1990). Rees (1980) summarized the major contributions to the thermal regime of an anaerobic refuse landfill as heats of reaction and neutralization, solar radiation, microbial metabolism and specific heat of water/refuse mixtures.

Clearly, the potential for a hazardous waste facility to contaminate the environment, via gaseous and leachate emissions, is significant and if not strictly controlled could leave a devastating legacy for future generations. In recent years it has become apparent that the air pathway can be an important route of exposure of the public and environment to toxic substances from hazardous waste disposal facilities (Navarro and Quan, 1992). For example, in Bayou Sorrel, Louisiana, U.S.A., millions of litres of toxic waste were dumped in huge open pits. The air currents then carried the volatilised pollutants to nearby areas (Miller and Miller, 1991). According to Seiber, Hsieh, Kado, Kuzmicky, Ning, Wong and Woodrow, 1992), vapour-phase mutagens are diverse and include both volatile

compounds (for example, methyl bromide, methylene chloride and formaldehyde) as well as semi-volatile compounds (for example, polycyclic aromatic hydrocarbons (PAHs) and some halogenated chemicals). Clearly, the method of refuse emplacement and leachate recycle as well as the compound co-disposed must be strictly controlled to prohibit this form of pollution

The very serious effects that landfilling, as such, has had in the U.S.A. can be appreciated by the 'cleanup' figures estimated by the U.S. Office of Technology Assessment which predicted that 100 billion dollars would be required to restore up to 10,000 sites which pose a serious threat to health (Chiras, 1994). In an experiment to determine the genotoxicity effects of hazardous waste sites, Houk, deMarini, Watts, and Lewtas (1992) collected feral rodents living on two Superfund sites in the U.S.A., and compared the frequency of chromosomal aberrations to rodents from an uncontaminated control site. The results demonstrated a clear association between hazardous waste exposure and the number of aberrations (Houk *et al.*, 1992). This serves to illustrate the varied way in which co-disposal operations can affect not only the air and water environment but also the surrounding fauna and further indicates that "on-site" workers should also take the necessary precautions against contamination. Although, in most cases, serious environmental contamination has resulted from leachate leakage, other factors such as dust, noise and soil erosion from slopes also pose a threat to the immediate environment (Anon, 1994a) .

1.7 Attenuation Mechanisms

Modern landfilling practices place great reliance on the long-term integrity of any site liner as large and high-density domestic landfill sites will continue to produce "strong" and polluting leachates for a very long time (Robinson and Gronow, 1992). Therefore, to limit any potential pollution due to liner failure or contaminant leaching all attenuating mechanisms available in a refuse mass must be optimised.

1.7.1 Immobilisation of Wastes in Landfill Sites

Often recalcitrant wastes such as heavy metals or semi-recalcitrant substances are co-disposed in landfill sites. These wastes rely on physico-chemical measures to dilute and/or immobilise them in the refuse mass or this could be effected by a site liner system (Watson-Craik, 1987). Despite the range of attenuating mechanisms operative within a refuse mass, very little quantitative data are available on the contribution of each mechanism. Further, hazardous compounds are usually attenuated by a combination of mechanisms. For example, in laboratory studies, co-disposal of barium containing salts with refuse was examined and the importance of physico-chemical processes, particularly adsorption, was identified. Microbial activity was, however, also implicated since microbially produced carbon dioxide and bicarbonate effected the precipitation of barium carbonate (Lagas, Loch, Bom and Gerringa, 1984).

1.7.2 Absorption into the Refuse Mass

The volume of liquid waste that can be immobilised by absorption in the refuse mass is site specific and is a function of the absorptive capacity of the refuse mass. This, in turn, depends on the site water balance and the field capacity of the refuse. Typically, MSW as placed contains between 10 and 30% (w/w) water which is the minimum moisture needed for anaerobic microorganism survival and methane production (Buivid, Wise, Blanchet, Remedios, Jenkins, Boyd and Pacey, 1981). With an increase in moisture content the field capacity is reached and with additional moisture the formation of leachate results. The volume of liquid which can be added is influenced by many complex factors which include:

a.   Particle size and refuse density which are functions of landfill operation procedures. Liquid absorption increases in the presence of high surface area to volume ratios (low refuse density) (Rees and Grainger, 1982). At many landfill sites densities of 0.7 to 0.8 t m^3 of wastes as received are achieved and at such densities it is likely that about 0.1 to 0.2 m^3 of added liquid per cubic metre of waste as received can be

absorbed before substantial leachate generation results. However, at higher compaction densities absorptive values will fall. For example, at emplacement densities $> 1.0 \, t \, m^3$ the absorptive capacity may fall to as low as 0.02 to $0.03 \, m^2$ liquid per cubic metre of received waste (U.K. Department of Environment, 1988). Further, the pulverisation or shredding of refuse will significantly decrease the mean particle size and increase its reactivity (Rees, 1980; Barlaz, Milke and Ham, 1987);

b. Water availability. All sources of moisture must be considered important. These include: initial moisture at emplacement, infiltration water due to rainfall or groundwater, and moisture generated during aerobic metabolic decomposition of refuse (Leckie, Pacey, Members of ASCE, and Halvadakis, 1979; Emcon Associates, 1982);

c. Nature of cover material. The type of soil used for intermediate as well as final covering and whether vegetation is planted or not also play significant roles in the volume of water entering/leaving the site (Canziani and Cossu, 1989); and

d. Liquid distribution. In landfills, *in situ* liquid is not uniformly distributed but is held in the refuse mass in three forms: gravitational, capillary and hygroscopic. Capillary and hygroscopic waters are held in micropores and void spaces whereas gravitational water is often present in macrovoids between the refuse components and, as such, may result in the formation of perched water tables (Senior, 1991). Further, liquids will always follow the path of least resistance and could form, so called, "channels" where liquids pass through a refuse mass without being absorbed (Senior, Watson-Craik and Kasali 1990).

Although a site specific maximum hydraulic loading rate can be defined in relation to a known weight of refuse, at which no inhibition of microbial catabolic processes results, and at which there is, therefore, no impairment of leachate quality, quantified rates are seldom available (Senior, 1991). Further, Senior, Watson-Craik, Sinclair and Jones, (1991) reported that site licence-directed rates are

often unrealistically low and err considerably on the side of safety (Senior *et al.*, 1991).

1.7.3 Refuse/Soil Adsorption

Adsorption may be defined as the accumulation of a chemical at an interface (Knox, Sabatini and Canter, 1993) resulting in, at least, a partial resistance to extraction by a salt solution (Fuller, 1983). Adsorption, is essentially a two phase process with the initial, and fast, phase usually being followed by a longer and slower phase which is controlled by the transfer of the solute to internal adsorption sites. There is also, quite often, an irreversibly sorbed, recalcitrant fraction, which has been shown to increase in size with time (Karickhoff, 1981).

The extent and significance of the adsorptive processes in refuse are not well understood. Further, adsorption and desorption are controlled by the chemical properties of the adsorbate and the surface properties of the adsorbing medium (the adsorbent). Together they regulate the solution concentration of adsorbed chemicals present in the leachate. The properties of the adsorbent which influence its behaviour in interactions with the adsorbate include magnitude, distribution and intensity of the electrical field at the surface (Kaufmann, 1983). The properties of the adsorbate which influence its adsorption/desorption include (Kaufmann, 1983; Blakey, 1984; Watson-Craik, Sinclair and Senior, 1992a):

a   Chemical character, shape and configuration;
b.   Acidity/basicity of the molecule;
c.   Water solubility;
d.   Charge distribution;
e.   Polarity;
f.   Molecular size; and
g.   Polarizability.

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Adsorption is further influenced by a number of environmental factors including:

a. Soil pH;
b. Surface acidity;
c. Temperature;
d. Moisture content; and
e. Redox (Eh) conditions.

The adsorption attachment may be as a result of one or a combination of electrostatic forces ie. van der Waals/London forces, hydrogen bonding, ion exchange and chemisorption (Knox et al., 1993).

The solid components of MSW present a wide variety of physical and chemical surfaces which may interact with fluids passing over them (Senior, 1990b). This is quite a different situation to that found in most anaerobic digesters where the bacterial sludge presents a more uniform and less diverse surface. This direct interaction may be an important factor both for labile organic compounds and for inorganic constituents such as heavy metals. Organic compounds may be adsorbed to an extent which is sufficient to reduce solution concentrations to non-inhibiting levels thus allowing them to be degraded. Further, adsorption could lead to the compounds being retained within the reactor for longer periods than would be predicted purely from hydraulic considerations and so allow longer time periods for degradation of recalcitrant compounds (Knox, 1989).

A common misinterpretation is that once a compound is adsorbed it no longer poses a threat to the system, as it is irreversibly bound. However, adsorption studies conducted with pesticides in soils have shown the reversible nature of adsorption (Knox et al., 1993). Desorption studies have illustrated that when the pesticide concentration in the soil pore water decreases, desorption of the pesticide from the adsorbent phase to the solution phase occurs.

Desorption, therefore, plays a significant role in determining the environmental fate

of organic compounds as the movements of chemicals have been shown to be governed by desorption (Overcash, 1986). This process is dependent on a number of factors such as the nature of the refuse, with regards to the availability of surfaces and the surface area, and anaerobic conditions etc (Watson-Craik, 1987). As for adsorption, quantitative data on desorption in refuse systems are largely lacking. In addition, the absence of standardisation of experimental procedures makes comparison of results very difficult.

Initially, adsorption models assumed that desorption was completely reversible and, hence, that the desorption curve was symmetrical to the adsorption curve. However, subsequent studies have shown that desorption is not symmetrical to adsorption. Desorption is not 100% since a portion of the original adsorbate remains adsorbed and accounts for the lack of symmetry between the two curves. This phenomenon is termed hysteresis and results in an asymmetrical desorption curve (Swanson and Dutt, 1973; Miller and Chang, 1989). Studies involving soil and phenol showed a considerable hysteresis and it was concluded that a portion of the phenol adsorbed was irreversibly held (Sawhney, 1989).

Adsorption on Refuse

The landfill environment is characterised by both spatial and temporal heterogeneity the result of which makes the study of the numerous attenuation mechanisms very difficult, and accounts for the general lack of studies with regards to these mechanisms (Sawhney, 1989). Consequently, studies made in other environments, particularly soils (Artiola-Fortuny and Fuller, 1982, Scott, Wolf and Lavy, 1982), have been applied to the landfill ecosystem, with the assumption that these mechanisms have a general form of function which does not alter significantly with a change in the supporting environment.

Attenuation mechanisms may be divided into three broad categories, namely: hydrodynamic, abiotic and biotic processes. Interactions between the abiotic and biotic components in landfills are extremely complex and contribute to the non-uniform distribution of chemicals within the refuse mass (Weber and Miller, 1989; Knox, Sabatini and Canter, 1993). Abiotic processes rarely bring about significant changes in the chemical

structure whereas biotransformations are much more important in the removal of toxic xenobiotics (Tibbles and Baecker, 1989a).

One of the few studies to assess the adsorption of organic molecules to the surfaces found in a refuse mass was made by Knox and Newton (1976)) who challenged batches (1kg) of both 'fresh' (8 weeks) and 'aged' (4 years) refuse with phenol, p-cresol and 2,6-xylenol. The initial concentrations of phenols ranged from 200 to 2000 mg l^{-1} (phenol) and 100 to 1000 mg l^{-1} (p-cresol and 2,6-xylenol). All three phenols were adsorbed to a significant extent and the adsorption appeared to be complete within one hour. The extent of adsorption varied with the age of the waste, with fresh refuse adsorbing more than "aged" refuse. However, the pattern also varied with the individual phenols depending upon whether the overall concentrations were high or low. The adsorption isotherms for "aged" refuse were found to give a better fit to the Freundlich equation than the Langmuir equation. The actual values obtained are given below:

phenol $Y = 0.035C^{1.42}$

p-cresol $Y = 1.107C^{1.00}$

2,6-xylenol $Y = 2.913C^{0.98}$

where: C = concentration in the leachate (mg l^{-1}); and

                Y = concentration adsorbed on the solid

                phase (mg kg^{-1})

A further experiment made by Blakey and Barber (1980) with 200 litre containers and partly decomposed refuse indicated that although elevated phenol concentrations occurred in the leachate, they accounted for < 1 % of the added phenol. However, some phenol appeared to have been biodegraded although no measurements of catabolic intermediates were undertaken. In a further experiment to determine the affinity for sorption of three compounds, 1,4-dichlorobenzene, 1,2,4-trichlorobenzene and naphthalene, by various refuse components, Reinhart, Pohland, Gould and Cross (1991) concluded that an increasing affinity was associated with decreasing solid phase surface energy and increasing surface wettability. Therefore, the majority of refuse sorption would

be expected to occur on low energy organic surfaces such as fats, oils, waxes, microbial cell walls, hydrocarbon side chains of humic-like substances, lignin, plastic and leather (Reinhart *et al.* 1991).

Knox (1989) concluded that the role of refuse adsorption in phenol degradation was not clearly demonstrated and that in column lysimeter studies there was no delay in the breakthrough of phenol.

Another factor in the early breakthrough of phenol which must be borne in mind is that many columns and lysimeters exhibited some degree of short-circuiting within the refuse thus reducing the opportunity for adsorption (Knox, 1989). Further, Willetts (1983b) cautioned that the adsorptive capacity of refuse is often difficult to establish and impossible to predict since it depends on a wide variety of factors many of which are uncontrollable (Willetts, 1983b).

Perhaps more importantly, the experiments made by Knox and Newton (1976) demonstrated that the adsorption of the three phenols was at least partly reversible. Unfortunately, the data were not considered adequate by the author for the calculation of desorption isotherms. However, the desorption was sufficiently extensive to suggest that complete reversibility was a reasonable possibility.

The fate of heavy metals is one of the most important aspects of co-disposal. Unlike organic compounds, heavy metals are not catabolized by microorganisms and cannot, therefore, be removed from the environment in which they occur. Further, reducing (anoxic) conditions favour accelerated migration of heavy metals compared with oxidative (oxic) conditions. For example, the contaminants As, Be, Cr, Fe, Ni, Se and Zn are much more mobile in soil under anaerobic than aerobic conditions, all other factors being equal (Fuller, 1983). Similarly, Francis and Dodge (1988) reported that under anaerobic conditions reductive dissolution of metal oxides (Mn^{3+}, Fe^{3+}, Co^{3+} and Ni^{3+}) from a higher to a lower oxidation state (divalent ions) can increase the solubility by several orders of magnitude. An exception to this should be noted, however, since H_2S production can

greatly reduce the migration of many heavy metals by precipitation of insoluble metal sulphides (Fuller, 1983). For example, from lysimeter experiments made by Pohland and Gould (1986) it was concluded that sulphide precipitation may be the chief control of Cd and Zn immobilisation.

A further influence on heavy metal mobility is the pH of the environment. According to Fuller (1983), even a slight lowering of the pH value from 8 to 6 will markedly influence the attenuation of most heavy metals whereas lowering the pH even further to 3 will greatly increase the solubility of heavy metals. Blakey (1984) studied the adsorption of arsenic (III) and (V) with both "aged" and "fresh" refuse and noted that with "aged" refuse the concentrations of As (III) were reduced in the leachate by approximately 86 % at pH 9 and 43 % at pH 5. For "fresh" refuse the reduction was from 86 % at pH 9 to 51 % at pH 5. In the presence of very low sulphide concentrations, carbonates and hydroxy-carbonates play fundamental roles in the precipitation of metals (Cossu and Serra, 1989). In a study made by Pohland and Gould (1986) it was suggested that chromium precipitation, to below detection limits, was effected mainly by hydroxides.

Despite these influencing factors, the five co-disposal landfill sites studied in the U.K. (Section 1.7) by Knox (1989) indicated that the heavy metal concentrations were within the ranges which are typically found in domestic refuse leachates. These results showed that the applied organic and heavy metal loadings had not disrupted the normal degradation processes and that the loadings were within the capacity of the refuse mass to degrade or attenuate them (Knox and Gronow, 1989). Therefore, the U.K. Department of the Environment suggested that an initial loading of ±100 g of soluble Cr, Cu, Pb and Zn per tonne of mature refuse was unlikely to produce a significant change in leachate concentrations three metres distant from the added heavy metal (U.K. Department of Environment, 1988).

The question of toxicity and a reduction in the pollution potential afforded by heavy metal co-disposal have been widely studied (Parkin, Speece, Yang and Kocher, 1983; Pohland, Gould and Ghosh, 1985; Pohland and Gould, 1986). Jarrell and Saulnier (1987)

investigated the effects of the heavy metals Ni, Cu and Zn on methanogenesis by monocultures of *Methanosarcina barkeri*, *Methanospirillum hungatei*, *Methanobacterium formicicum* and *Methanobacterium thermoautotrophicum*. The workers found that the inhibitory concentration of Cu was between 5 and 10 mg l^{-1}, while 1-10 mg l^{-1} caused 50% inhibition of all four methanogens. Conversely, nickel was less toxic towards the methanogens studied. *Methanospirillum hungatei*, *Methanospirillum barkeri* and *Methanobacterium thermoautotrophicum* were sensitive to concentrations of between 0.25 and 1.2 g Ni l^{-1} but *Methanobacterium formicicum* was resistant to Ni concentrations > 15 g l^{-1} (Jarrell and Saulnier, 1987).

It is interesting to note that in the control of heavy metals, microorganisms can accumulate them. Microorganisms can accumulate (biosorption) heavy metals by a variety of physical, chemical and biological methods. Living and dead cells, as well as products excreted by, or derived from, microbial cells, e.g. cell wall constituents, pigments and polysaccharides, are capable of removing heavy metals (Gadd, 1992). This microbial resistance to toxic metals has arisen in two distinct ways:

a.   Some microorganisms have inherited the ability to resist high concentrations of toxic elements through their evolution under extreme selection pressures; and

b.   Other species, particularly bacteria, have acquired a transferred resistance to the polluted environment by, for example, the acquisition of plasmid DNA (Senior, 1990a) .

Adsorption on Soil

Unlike refuse, the effect of adsorption of organic molecules has been extensively studied in soil (Scott *et al.*, 1982; Fuller, 1983; Weissenfels, Klewer and Langhoff, 1992). Although soil coverings (intermediate and final) constitute only a small fraction of the landfill volume, the total surface area per unit volume of soil greatly exceeds that of refuse. It is, therefore, probable that soil has a significant influence on both microorganisms and

pollutants migrating through the refuse/soil mass (du Plessis, 1995). Also, in Class II, or "dilute and disperse" sites, leachate is allowed to migrate through the underlying strata (soil or gravel). The unsaturated attenuation zone below the site is, thus, of critical importance in protecting groundwater aquifers. In these soils, adsorption of organic compounds is affected by organic matter content, type and amount of clay, oxide content and characteristics, ion exchange capacity and surface activity (Scott, Wolf and Lavy, 1982; Ferguson, 1994).

Due to the different characteristics afforded by soils, the adsorption of phenol, on soil and sediments, unlike refuse, has been shown to be considerable. Sawhney (1989), for example, proposed that the higher sorption, was a result of the greater electron-donating ability of the substituents, and suggested that phenols form H-bonds with soil surfaces. The author indicated that as much as 90% of the 2-chlorophenol sorbed by a sediment sample was irreversibly held. The strong affinity of phenols is exemplified by high values (2900-4900) of partition coefficients calculated on an organic matter basis. Partition coefficients predicted from octanol/water partitioning studies are almost two orders of magnitude lower. These differences clearly show that sorption of phenols by soils and sediments occurs through a more specific interaction, such as H-bonds, rather than by general hydrophobic mechanisms (Sawhney, 1989). On the other hand, Saltzman, Kliger and Yaron (1986) indicated that adsorption of the pesticide parathion was affected by the amount of organic matter in the soil and that the slope of the desorption isotherm was rather steep thus demonstrating that adsorption was easily reversible (Saltzman et al., 1986).

Probably the single most important, laboratory determined, parameter for predicting the movement and adhesion of organic compounds in soils is the octanol/water partitioning coefficient (K_{ow})(Chiou, 1989). Green, Lee and Jones (1981) found that the hydrophobic or hydrophilic nature of the organic compounds, as measured by the octanol/water partitioning coefficient (or, approximately, by the dielectric constant), were important for predicting the solvent's rate of flow through soils. The octanol/water partitioning coefficient measures the tendency of molecules to escape from the aqueous phase. Thus,

hydrophobic substances would be expected to adhere more strongly to the soil solid phase than water. This fact was clearly demonstrated in experiments made by Weissenfels, Klewer and Langhoff (1992). In these experiments the bioavailability and, thus, the biodegradation of polycyclic aromatic hydrocarbons (PAHs) was related to the amount of sorption. It was found that there was a reduction in the PAHs degradation rates with increasing sorption capacity of the sorptive substrates used. Further, as the PAHs are characterised by high partitioning coefficients, naturally occurring organic matter was an excellent sorbent for these compounds (Weissenfels *et al.*, 1992).

The organic content (humic and fulvic acids) in soil was also shown to exert an influence on the amount of phenol adsorbed due to the processes of complexation/chelation (Reinhardt, Gould, Cross and Pohland, 1990). Scott, Wolf and Lavy (1982) observed that phenol adsorption was twice as great on Palouse soil, which had a greater organic content, than Captina soil. Karickhoff (1981) concluded that for neutral hydrophobic solutes, sorption isotherms in the low loading limits are linear, reversible and can be characterised by a partition coefficient (K_p). These partition coefficients are closely correlated with the organic carbon content of sediments/soils. Further, relating sorption to organic carbon gives a partition coefficient to organic carbon (K_{oc}) which is highly sediment/soil independent (Karickhoff, 1981).

The oxide content (hydrous) can also play a role in contaminant adsorption. Unlike silicate clays, where the inherent charge is mostly permanent because of isomorphous substitution, the exchange capacity of the oxide minerals is pH dependent (Foth, 1984). Oxides undergo protonation (producing a positive charge) in acidic conditions and, thus, create an anion exchange capacity, while deprotonation (producing a negative charge) results at higher pH values thus creating a cation exchange capacity (Foth, 1984; Ferguson, 1994). The soil oxide minerals often form coatings on the surfaces of other soil minerals such as silicate clays and sand-sized particles. These coatings alter the surface characteristics of the minerals and may influence microbial and organic compound interactions with these minerals (du Plessis, 1995).

It is interesting here to note that enhanced mobility of hydrophobic pollutants can result from co-transport with bacteria in saturated soil (Lindqvist and Enfield, 1992). For example, in a column study, 1.2×10^9 cells ml^{-1} of Bacillus sp. enhanced dichlorodiphenyl-trichloroethane (DDT) transport about 8-fold. It was concluded from literature data that 10^8 cells ml^{-1} would increase the mobility of very hydrophobic compounds ($K \geq 6$) whereas higher counts of bacteria (10^8 cells ml^{-1}) would have a significant impact on compounds with a log K of ≥ 4 (Lindqvist and Enfield, 1992).

Most contaminants are transported through soil in an aqueous medium, either in solution or suspension. The exceptions are volatile organic compounds which are transported in the gaseous phase. For this reason the moisture content of the soil is of utmost importance (Ferguson, 1994). Also, mobility of contaminants may be influenced by differential solubilities.

It has also been shown, by Watson-Craik (1995) who reviewed previous work, that many chemicals are more soluble in organic solvents than in water. Phenol, for example, although soluble in water (82 g l^{-1} at 25°C) is more soluble in a range of organic solvents such as benzene (distribution coefficient (D_e) at 20°C = 2.2), diethylether (D_e = 17.0) and isopropylether (D_e = 17.0) (Watson-Craik, 1995).

The pH of the solution/soil also plays a significant role in contaminant (solute) mobility. For example, Daniel and Shackelford (1989) reported that strong acids and bases can dissolve solid material in soil, form channels and dramatically increase the permeability. Some acids, such as hydrofluoric and phosphoric, are particularly aggressive and dissolve soil readily (Daniel and Shackelford, 1989). Fortunately, concentrated acids are not directly landfilled since they are extremely corrosive and also have the potential to cause fires and produce toxic gases by chemical reactions (Batstone, 1989).

1.7.4 Biodegradation.

The key to the assessment of the fate of organic chemicals in the environment is a realistic evaluation of their susceptibility to mineralization to the end products CO_2, H_2O or CH_4 as well as nitrate, sulphate and biomass (Ghisalba, 1983). While photooxidation (Cerniglia, 1993) and other abiotic mechanisms play a role in the transformation of many chemicals, few such mechanisms are able to totally convert the molecules to inorganic products *in situ*. Therefore, assessments of the environmental fates of organic chemicals are highly dependent on evaluations of their susceptibility to biological attack (Grady, 1985).

By strict definition, xenobiotics are compounds considered to be unnatural (van der Meer *et al.*, 1992). A wider definition in current usage, and one which will be used here, emphasises that the term xenobiotic should not be restricted to compounds with structural features foreign to life but should be used for all compounds that are released in any compartment of the environment by the action of man and thereby occur in a concentration that is higher than natural (Müller, 1992; Leach *et al.*, 1994). A typical example is mineral oil. As long as it remains in the geological strata it poses no threat but as soon as it is released by human activities into the oceans, it poses a serious threat to all higher organisms living in these waters, and it may be called a xenobiotic (Müller, 1992).

During the last few decades the degradation pathways of many compounds, which are xenobiotics in the sense described above, have been elucidated. However, the importance of anaerobic transformations has been underestimated, perhaps, as a result of the seemingly slow reaction rates achieved in the absence of oxygen. The versatility of oxygenase enzymes, especially the monooxygenases of the cytochrome P450 type, allows a broad range of reactions to be catalysed in the presence of oxygen (Schink, 1988). In contrast, the persistence of some compounds in sediments and waterlogged soils, notably lignin and polycyclic aromatic hydrocarbons, shows that they are not metabolized by anaerobes (Atlas and Bartha, 1987). One reason for this is that of the many catabolic reaction types observed in natural environments, only a few are operative in the absence of oxygen, namely hydrogenations, dehydrogenations, hydrations, dehydrations, hydrolyses,

condensations, as well as carboxylations and decarboxylations (Schink, 1988) and dehalogenations (Hakulinen, Woods, Ferguson and Benjamin, 1985).

Notwithstanding this delayed appreciation of the anaerobic degradation capabilities, numerous xenobiotics have been successfully catabolized in laboratory and full-scale studies. These include organic molecules, such as the substituted aliphatics dibromomethane and trichloroethane (Reinhart *et al.*, 1991), aromatic (El-Mansi, 1986; Senior and Watson Craik, 1991) and halogenated aromatic molecules, such as 2,4-dichlorophenol (Zhang and Wiegel, 1990; Motosugi and Soda, 1983), chlorinated heterocyclic compounds, such as hexachlorocyclohexane isomers (Jagnow, Haider and Ellwardt, 1977) as well as complex insecticides and herbicides (Ghisalba, 1983; Cork and Kreuger, 1991).

Degradation of Aromatic Compounds

Cork and Kreuger (1991) reported that anaerobic degradation of aromatic compounds can be accomplished by: a) photosynthetic anaerobic metabolism; b) metabolism by nitrate-reducing bacteria; c) anaerobic dissimilation through sulphate respiration; d) anaerobic fermentation; and e) anaerobic fermentation by an undefined methanogenic consortium.

Although, individually, organic compounds can serve as the sole carbon source for the purple phototrophic non-sulphur bacteria (Evans, 1977), they will not be further discussed here as their contribution is thought to be minimal in a landfill situation.

The first report of aromatic ring cleavage during nitrate respiration resulted in the postulation of a degradation process not unlike that found in aerobic organisms. Based on mixed culture studies, it was at one stage proposed that during ring cleavage of both hydroxybenzoate and protocatechuate, the oxygen atoms of nitrate behaved as if they were O_2 (Colberg, 1988). However, later studies of Evans (1977) indicated that the anaerobic metabolism of benzoate by a *Moraxella* sp. in the obligatory presence of nitrate was different as no oxygenase enzyme(s) could be detected. Flyvbjerg, Jorgensen, Arvin,

Jensen and Olsen (1993), indicated that the degradation of o-cresol by a mixed culture was dependent on toluene as a primary substrate under nitrate-reducing conditions. This dependency on toluene metabolism indicates that o-cresol was transformed by co-metabolism. Thus, nitrate-reducing bacteria couple the oxidation of organic compounds with water to the exergonic reduction of nitrate via nitrite to N_2 or NH_3. Energy is derived mainly from electron transport phosphorylation during nitrate respiration while cell carbon is derived from breakdown products of the organic compound (Cork *et al.*, 1991). Microorganisms which carry out nitrate respiratory metabolism (e.g. the denitrifiers) are facultative in character and appear to prefer oxygen as their electron acceptor. Under anoxic conditions, however, this group of microorganisms uses a wide range of organic compounds as carbon and energy sources. Aftring and Taylor (1981) demonstrated that a *Bacillus* sp. using only phthalic acid as the carbon source and nitrate as the sole electron acceptor grew either aerobically or anaerobically on phthalate. Further, experiments with fluorobenzoate compounds provided evidence that benzoate was a common intermediate in the degradation of phthalate under both aerobic and anoxic conditions.

Wallnöffer and Engelhardt (1981) showed that under nitrate-reducing conditions a reductive pathway is operative for the decomposition of benzoic acid to adipic acid by a *Moraxella* sp. and for the anaerobic degradation of phenol and other aromatic compounds. Concomitant with the reduction of the aromatic substrate, nitrate is reduced mainly to nitrogen gas. Cyclohexanecarboxylic acid, cyclohex-1-enecarboxylic acid, 2-hydroxycyclohexanecarboxylic acid and adipic acid were also identified as intermediates (Wallnöffer and Engelhardt, 1981).

Likewise, the sulphate-reducing bacteria couple the oxidation of organic matter with water to the exergonic reduction of sulphate to sulphide. The obligate anaerobic sulphate-reducing bacteria are often responsible for degradation of organic matter, such as aromatics, in marine environments which contain approximately 27 mM sulphate (Berry, Francis and Bolag, 1987; Cork *et al.*, 1991).

In the absence of oxygen, microorganisms adopt a pathway that is different from

aerobic microorganisms for the catabolism of aromatic rings. Evans (1977) summarised probable pathways for the anaerobic degradation of aromatic compounds. It was concluded that the conversion of aromatic compounds to methane and carbon dioxide is accomplished, first, by the reduction of the benzene nucleus via hydrogenation resulting in the formation of some alicyclic compounds as intermediate products. These intermediates are then further cleaved by hydrolysis to form aliphatic acids and, subsequently, volatile organic acids. Finally, the organic acids are converted to suitable substrates (acetate, hydrogen, formate) for methanogens to complete the process. Dwyer, Krumme, Boyd and Tiedje (1986) examined an association of three types of bacteria which were responsible for the conversion of phenol to methane and carbon dioxide. The results showed that an unidentified cocco-bacillus degraded phenol to acetate and hydrogen while there were two types of methanogens: a *Methanothrix*-like organism, which converted acetate to methane and carbon dioxide; and *Methanobacterium formicicum*, grew on hydrogen and carbon dioxide to produce methane. Godsy, Goerlitz and Grbic-Galic' (1992) identified several distinct populations of bacteria in the biodegradation of phenolic compounds in aquifer-derived microcosms. *Methanobacterium bryantii* was isolated by H_2/CO_2 enrichments while *Methanothrix soehngenii* was isolated from acetate enrichments with a sulphate-reducing bacterium, similar to *Desulfobacterium*. Further, numerous facultative aerobic and denitrifying rods capable of heterotrophic growth on various media were also recorded. These studies serve to demonstrate that the cooperation of several bacteria utilising different substrates and electron acceptors are required for phenol catabolism to methane and carbon dioxide. For example, Ferry and Wolfe (1976) showed that the methanogenic fermentation of benzoate required the cooperation of several groups of bacteria and that the methanogens served only as the terminal organisms of the catabolism.

In an experiment made by Haggblom, Rivera, Bossert, Rogers and Young (1990) anaerobic degradation of *p*-cresol was studied with one sediment source under three reducing conditions - denitrifying, sulphate- reducing and methanogenic. The results demonstrated that the same initial pathway for *p*-cresol degradation, through *p*-hydroxybenzaldehyde and *p*-hydroxebenzoate, was operative under both sulphate-reducing and methanogenic conditions. Unique to the methanogenic enrichments, however, was the

transient appearance of substantial concentrations of benzoate as an intermediate after *p*-hydroxybenzoate. Benzoate was not observed in the denitrifying or sulphidogenic cultures, suggesting that the pathway might diverge after *p*-hydroxybenzoate, depending on which electron acceptor was available (Haggblom *et al.*, 1990).

Anaerobic dechlorination of 2,4-dichlorophenol in freshwater sediments in the presence of sulphate reduction was demonstrated by Kohring, Zhang and Wiedel (1989). In their experiment 2,4-dichlorophenol and 4-chlorophenol were reductively dehalogenated while sulphate reduction occurred. It was also shown that in the presence of added sulphate , the adaption periods for 2,4-dichlorophenol transformation to 4-chlorophenol were longer than those found under methanogenic conditions and the dechlorination rates were notably lower (Kohring *et al.*, 1989). Mohn and Kennedy (1992) suggested that reductive dehalogenation is the only reported means of biodegradation of highly chlorinated ethenes, benzenes, phenols and biphenyls. It was further shown that *Desulfomonile tiedjei* DCB-1 is the only anaerobe in monoculture capable of aromatic reductive dehalogenation (Mohn and Kennedy, 1992).

Methanogenic bacteria are thought to form methane from very few substrates ie: a) methanol; b) formate; c) methylamines; d) by decarboxylating acetate; and e) by utilizing hydrogen as an electron donor during CO_2 reduction (Farquhar and Rovers, 1973; Senior and Balba, 1990). The production of methane from more complex substrates, thus, depends on the activity of non-methanogenic bacteria in association with the methanogens. Therefore, studies of the degradation of aromatic compounds under methanogenic conditions have by necessity relied on mixed cultures the, so-called, microbial interspecies associations (Large, 1983; Kasali, 1986). The necessity of this is illustrated by benzoate catabolism (Senior and Balba, 1987):

$$C_6H_6COO^- + 6H_2O \rightarrow 3\ CH_3COO^- + 2H^+ + CO_2 + 3H_2 \quad \Delta G_o = +53\ kJ\ mol^{-1} \quad (1.2)$$
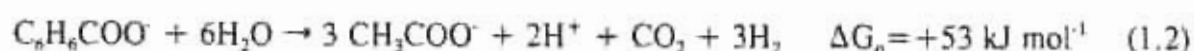
Thus, for this reaction to proceed, either sulphate-reducing bacteria or methanogens

are required to maintain low cultural conditions of both hydrogen and acetate. Therefore, in the presence of methanogens:

$$C_6H_5COO^- + H^+ + 4.5\ H_2O \rightarrow 3.25\ CO_2 + 3.75\ CH_4 \qquad \Delta G_o = -152kJ\ mol^{-1} \qquad (1.3)$$

The ability of these closely interacting bacterial species to effectively cooperate in the metabolism of various substrates accounts for the stability and efficacy of the overall degradative process (Senior and Balba, 1990). This implies that ring fission is tightly coupled to product removal and this hypothesis is supported by the work of Mountfort and Bryant (1982) who were able to isolate a benzoate-utilising bacterium only in co-culture with a *Desulfovibrio* sp. which "scavenged" the hydrogen. However, Grbić-Galić and Young (1985) showed that it is possible to separate benzoate degradation and methanogenesis. The authors used 2-bromoethanesulphonic acid (BESA), which acts as a metabolic inhibitor of methane formation and showed that although methane production decreased to 5 % of the normal concentration, ring cleavage of benzoate continued.

Using benzoate enrichment cultures from sheep rumen fluids and sewage sludge, Wallnöffer and Engelhardt (1981) detected several potential pathway intermediates including 1-cyclohexene-1-carboxylic acid, cyclohexane carboxylic acid, adipic acid, caproic acid, propionic acid and acetate. Healy and Young (1979) investigated the anaerobic degradation of lignin-derived aromatic compounds by a methanogenic culture obtained from sewage sludge. They were able to demonstrate the degradation of the following compounds: vanillin; vanillate; ferulate; cinnamate; benzoate; catechol; protocatechuate; phenol; p-hydroxybenzoate; syringate and syringaldehyde. Considering the number of different sources used to obtain benzoate degrading cultures, such as sewage sludge (Grbić-Galić, 1985), lake sediments (Horowitz, Suflita and Tiedje, 1983), it is not surprising that a wide range of intermediates have been proposed.

The sequence of reactions in the anaerobic dissimilation of benzoate under methanogenic conditions is, however, common: reduction of the aromatic ring with the formation of a cyclohexane derivative; and cleavage of the cyclohexane ring by hydrolysis

yielding aliphatic acids (Colberg, 1988; Evans, 1977). These in turn can be further degraded via the normal β-oxidation pathway until, eventually, methane and carbon dioxide result (Berry et al., 1987). Knoll and Winter (1987) demonstrated a second pathway where phenol is first carboxylated to benzoate before reduction and cleavage of the ring. This carboxylation as an initial step in degradation of aromatic compounds has so far only been reported for phenols. In a further study, Béchard, Bisaillon and Beaudet, (1990) also demonstrated the formation of benzoate during phenol degradation. Although the formation of benzoate is energetically favourable from phenol, CO_2 and H_2 the authors postulated that the carboxylation of phenol is accomplished by co-metabolism. Further, the inhibition of the methanogens did not influence the carboxylation of phenol which suggested that the carboxylating microorganisms are non-syntrophic (Béchard et al. 1990). Bisaillon, Lepine and Beaudet (1991) used the same bacterial association as Béchard et al. (1990) and identified the metabolic intermediates and the microorganisms responsible for phenol carboxylation. It was suggested that benzoate is transformed to 1-cyclohexene carboxylate and heptanoate. However, a part of the 1-cyclohexene carboxylate was transformed to an apparent dead-end product which was identified as cyclohexane carboxylate (Bisaillon et al., 1991).

Another important class of aromatic compounds degraded by methanogenic associations is the halogenated benzoates and phenolics. Horowitz et al. (1983) demonstrated that complete removal of the halogens to yield benzoate was required before mineralisation to carbon dioxide and methane could take place. The sequence of events effecting anaerobic degradation of halogenated aromatic compounds (ie. dehalogenation followed by ring cleavage) is quite different from the aerobic degradation pathway of the same compounds. Under aerobic conditions, the metabolism of halogenated benzenoids generally proceeds through one of two pathways: 1) replacement of a halide by a hydroxyl group after ring cleavage; or (2) ring cleavage followed by dehalogenation (Berry et al., 1987). In a study made by Cozza and Wood (1992) the electronic properties of the parent compound were correlated to the anaerobic degradation pathways. The authors used a semi-empirical computation method to determine the sum of the charges of the carbon-chlorine bonds which were then correlated with the reductive dechlorination pathways observed for

unacclimated microbial associations. For chlorobenzoic acids it was demonstrated that they are preferentially dechlorinated at the *meta* position to the carboxyl group. The carbon-chlorine bond charges were evaluated for chlorobenzoic acids and the charge of the *meta* chlorine clearly had a larger negative value than either the *ortho* or *para* chlorines. Likewise, comparison of the net carbon-chlorine charges for tetrachlorocatechol indicated that the most negative charges were adjacent to the hydroxyl groups.

For the chlorophenols it was reported that unacclimated associations preferentially dechlorinated chlorophenols at the *ortho* (adjacent to the hydroxyl group) position (Cozza and Woods, 1992). However, acclimated associations may affect different biotransformation pathways than unacclimated associations. For example, Boyd and Shelton (1984) observed that sludges acclimated to 2-chlorophenol degraded 4-chlorophenol and 2,4-dichlorophenol but not 3-chlorophenol, and those acclimated to 3-chlorophenol were not capable of degrading 2-chlorophenol. Further, organisms acclimated to different chlorophenols produce different initial pentachlorophenol degradation products. In a study by Bryant, Hale and Rogers (1991), chlorophenol reductive dechlorination pathways were determined for organisms acclimated to either 2,4-dichlorophenol or 3,4-dichlorophenol. The 2,4-dichlorophenol-acclimated association produced *ortho* dechlorination products, while the 3,4-dichlorophenol acclimated association produced *para* dechlorination products.

Pesticide Degradation

Compounds that probably evoke the most concern are the highly toxic pesticides. These synthetic compounds are often highly complex organochlorine aromatics that can, in many instances, be accumulated in microorganisms. Some microorganism have been reported to accumulate very high concentrations of insecticides. For example, after 4 hours of incubation with 1,1,1-trichloro-2,2-bis[*p*-chlorophenyl]ethane (DDT) and dieldrin, in concentrations ranging from 0.1 to 1 mg l^{-1} in distilled water, three species of fungi had accumulated 83% of the DDT and 75% of the dieldrin. Likewise, *Agrobacterium tumefaciens* had accumulated 100% of the DDT and 90% of the dieldrin (Lal and Saxena, 1982). These results indicate that microorganisms have the ability to bioconcentrate and

biomagnify xenobiotics from the environment, making the process of biodegradation even more important.

For four decades 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) and its derivatives have been widely used as herbicides, especially for the control of brush, jungle and aquatic weeds (Motosugi and Soda, 1983) In reality, however, 2,4,5-T contains traces of the highly toxic compound 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) as an impurity. In addition, about 0.0002% (w/w) of 2,4,5-T is converted to TCDD when wood or brush containing 2,4,5-T is burned. The spraying of the infamous herbicide agent orange (butyl esters of 2,4-dichlorophenoxyacetic acid (2,4-D) and 2,4-T in equal amounts) over the jungles of Southeast Asia has resulted in the accumulation of TCDD in the soil at a mean level of 1.9 mg l^{-1} with maximum concentrations of 47 mg l^{-1} (Motosugi and Soda, 1983).

Fortunately, the vast majority of the 500 or so active pesticidal chemicals can be biologically degraded by fungal or bacterial cultures (Munnecke, 1981). The ability of particular bacteria and fungi to transform the parent pesticide into less complex metabolites which may then be further metabolised by the same organism or by secondary microorganisms in soil or water, plays an important role in pesticide degradation. For example, in the cases of parathion (o-o-diethyl o-p-nitrophenyl phosphorothioate) and paraoxin, the hydrolysis products are 60 to 200 times less toxic than the parent pesticides (Munnecke, 1981).

In a study made by Wahid, Ramakrishna and Sethunathan (1986) it was found that parathion degradation was faster in flooded soils in comparison with dry soils and, therefore, more rapid under anaerobic conditions. Of the three types of soil used, the fastest degradation was obtained in soil with the highest organic content. In this soil the fall in redox potential after flooding was much faster than in the other soils.

Several studies made with DDT and other chlorinated hydrocarbon pesticides demonstrated that both aerobic and anaerobic degradation could contribute to the transformation of these compounds in soil and that generally the anaerobic processes were

faster. Similarly, pesticides possessing nitro groups are more rapidly transformed under anaerobic than aerobic conditions (Saltzman, Kliger and Yaron, 1986). For example, Golovleva and Skryabin (1981) demonstrated that for *Pseudomonas aeruginosa* degradation of DDT proceeded more rapidly under anaerobic conditions and that the first step in the dechlorination of DDT to 1,1-dichloro-2,2-di(p-chlorophenyl)-ethane took place without an additional substrate. All other degradative reactions proceeded exclusively under co-metabolic conditions. Lal and Saxena (1982) showed that DDT degradation, by strains of Hydrogenomonas, proceeded via reductive dechlorination to bis(p-chlorophenyl) methane.

Hydrocarbon Mineralisation under Anaerobic Conditions

Oxidation of methane or higher saturated hydrocarbons by anaerobic bacteria is the subject of controversial discussions. Generally, degradation can only be initiated under aerobic conditions since oxygenase reactions appear to be necessary for the initial metabolic activation of alkane molecules (Watkinson and Morgan, 1990). Thermodynamically, however, an oxidation of these hydrocarbons would be possible (Widdel, 1988), as the following calculation for ethane demonstrates, with the reaction becoming even more exergonic with increasing chain length (Schink, 1988):

$$8C_2H_6 + 6H_2O \rightarrow 14CH_4 + 2HCO_3 + 2H^+ \qquad \Delta G^{0'} = -34 \text{ kJ mol}^{-1} \qquad (1.4)$$
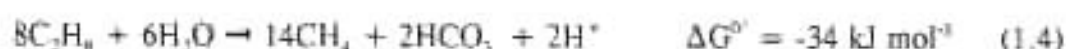
Notwithstanding this controversy, saline enrichments with hexadecane as the carbon source and oil field water as the inoculum resulted in the development of a sulphate-reducing bacterial culture after four months. Further study showed that pentadecane, heptadecane and octadecane only could serve as alternative organic substrates since with hydrocarbons of lower molecular weight the sulphide production decreased significantly (Widdel, 1988).

The situation becomes very different if there is at least one double bond in the hydrocarbon molecule. Hydration of this double bond would form an alcohol which, depending on its position, could be oxidised to a ketone or, via an aldehyde, to the

corresponding fatty acid. Thermodynamically, methanogenic degradation becomes even more exergonic as, for example, with ethene (Schink, 1988).

$$2C_2H_4 + 3H_2O \rightarrow 3CH_4 + HCO_3^- + H^+ \qquad \Delta G^\circ = -102 \text{ kJ mol}^{-1} \qquad (1.5)$$

Anaerobic, sulphate dependent, oxidation of acetylene was observed in estuarine sediment samples (Culbertson, Zehnder and Oremland, 1981). Furthermore, the authors were able to maintain an enrichment culture with acetylene as the sole source of carbon and energy.

The degradation of halogenated hydrocarbons has been well documented. Braus-Stromeyer, Hermann, Cook and Leisinger (1993), for example, identified a strictly anaerobic mixed culture capable of utilizing dichloromethane (DCM) as the sole carbon source. In this study it was recorded that complete and speedy degradation of DCM necessitated syntrophic metabolism of a fermentative DCM-dehalogenating bacterium with an acetogenic bacterium (Braus-Stromeyer et al., 1993). Reductive dehalogenation was also observed with tetrachloroethylene and 1,1,2,2-tetrachloroethane, and resulted in trichloroethylene and 1,1,2-trichloroethane, respectively (Bouwer and McCarty, 1983).

Often, under conditions of anaerobiosis and with mixed cultures, the intermediates or products formed during degradation are as or even more toxic than the original molecule. Evidence of this was found for the anaerobic degradation of trichloroethylene (TCE) which resulted in the formation of dichloroethylenes and vinyl chloride (Nelson, Montgomery, Mahaffery and Pritchard, 1987). A further example was given by Senior and Balba (1984) who demonstrated how a toxic chemical (aniline) may be generated from an innocuous plant component (anthranilic acid) under anoxic conditions. However, Schnell and Schink (1991) indicated that in the presence of Desulfobacterium anilini aniline was degraded via a carboxylation to 4-aminobenzoate which was followed by an activation to 4-aminobenzoyl-CoA. This in turn was reductively deaminated to benzoyl-CoA which entered the normal benzoate pathway. This carboxylation as an initial step in the degradation of aromatic compounds has so far only been reported for phenols. Benzoate has

been shown to form from phenol and CO_2 in phenol-degrading sewage sludge in the presence of hydrogen (Knoll and Winter, 1987).

Notwithstanding the fact that many of the studies mentioned above were not conducted in landfill sites *per se* it is clear that the ability of microorganisms to degrade numerous xenobiotics is significant. Further, it is expected that this ability of the microorganisms should be enhanced by the specific landfill conditions, which include:

a.   Landfill sites are not usually characterised by nutrient limitations and usually contain a variety of mixed substrates;

b.   A significant inoculum size and interactions among microbial associations;

c.   Long hydraulic retention times which allow for extended periods for microbial acclimation; and

d.   Enhanced physiological conditions such as high temperatures.

1.8   Sewage sludge treatment and disposal options

It is generally recognised that sewage sludge disposal is one of the most pressing environmental problems currently facing developed nations (Meyer, 1995). Sewage sludges are not generally considered industrial wastes although they share many characteristics since they are produced in large volumes from a, relatively, few distinct point sources (Watson-Craik, 1995). Furthermore, it has been shown that the total pollution load resulting from domestic wastewaters is usually as great as that from industrial wastewaters (Corbitt, 1990). The disposal of sewage sludge is a controversial subject as it is considered, by some, to be a beneficial substance and, by others, to be a hazardous waste (Crawford, Bredenhann and van der Westhuizen, 1994).

1.8.1 Sewage Sludge Types and Composition

According to Vesilind (1991) sludge characteristics are specific to the country, city, part of town and even the time of year. Generally, raw sewage is more than 99.9% (w/w)

water with the solids consisting of dissolved and suspended organic and inorganic materials (Steel, 1953; Anon, 1974). The organic solids consist mainly of proteins, carbohydrates and fats which are degraded by saprophytic microorganisms.

Of the various nutrients present in sewage sludge, nitrogen and phosphorus are undoubtedly of primary concern and can occur in both organic and inorganic forms (Byrom and Bradshaw, 1989). Soil experiments have demonstrated that sewage sludge serves as a low grade fertilizer, typically with a 4:12.1 % N,P,K ratio which can improve soil fertility.

The inorganic solids are usually dissolved and consist of calcium, sodium and sulphate with numerous toxic heavy metals (Imhoff and Fair, 1956; Corbitt, 1990). Of particular concern are concentrations of the 'standard six', namely Cd, Zn, Cu, Ni, Pb, and Cr, and also Hg (Meyer, 1995). Finally, there can be a large number of pathogenic organisms capable of causing various diseases which are also borne along in the sewage. Many bacteria, such as *Vibrio cholerae*, *Salmonella typhi* and *Mycobacterium tuberculosis* as well as viral agents, such as infectious hepatitis and poliomyelitis, are routinely isolated from sewage sludge.

According to the final draft of the South African Department of National Health and Population Development (DNH&PD) guide 'Utilisation and Disposal of Sewage Sludge' there are, basically, four types of sewage sludge. The four types are categorised according to the degree of sludge stabilisation and disinfection as well as the heavy metal content (Ekama, 1992; van der Merwe and Vivier, 1994):

a.   *Type A*: Unstable with high odour and fly nuisance potential as well as a high content of pathogenic organisms. These are usually raw or cold digested sludges;

b.   *Type B*: Stable with low odour and fly nuisance potential, with a reduced pathogenic content. These are anaerobically digested sludge as well as surplus activated sludge;

c.   *Type C*: Stable with negligible odour and fly nuisance potential and insignificant numbers of pathogenic organisms; and

d.   *Type D*: Sewage sludge included in this classification is of similar hygienic quality as *Type C* but since it is designated for unrestricted use on land at a maximum application rate of 8 dry tonnes $Ha^{-1} y^{-1}$, the metal and inorganic contents are limited to acceptably low concentrations. Further, all *Type D* sludges must be registered in accordance with the regulations of Act 36 of 1947.

As it is difficult, and costly, to analyse for all the pathogenic organisms, only the numbers of *Ascaris* ova, *Salmonella* spp. and faecal coli are included as indicators of hygienic quality requirements of *Types* C and D (van der Merwe and Vivier, 1994).

According to Bruce and Davis (1989), technology is capable of producing at least nine different types of sludge end products which are suitable for final disposal. These types of sludge differ from the DNH&PD classification in that they depend on the processes used in the sludge stabilisation phase.

1.8.2 Legal Provisions - The South African Situation

The greatest part of the guidelines and recommendations and other regulations enacted in most of the European countries deals mainly with spreading sewage sludge, manures and slurries on arable and grassland (Strauch, 1990). Until 1994, in South Africa, there were no regulations for the agricultural use of sewage sludge - only guidelines (Ross, 1990). Where sewage sludge is co-disposed with refuse Section 20 of the Environmental Act, Act 73 of 1989 applies (Ekama, 1992; Crawford *et al.*, 1994).

According to Crawford *et al.*, (1994) the guidelines, drafted by the DNH&PD, focus attention on using different types of sludge for agriculture and concentrates specifically on human health and protection of the soil with little attention given to the protection of water quality. Further, the guidelines list the safe uses for various types of

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sewage sludge but do not prescribe that sludge must be disposed of at all. In effect this means that sludge could merely be stockpiled and still meet all necessary requirements. However, in 1989 the Department of Water Affairs & Forestry (DWAF) was made responsible for administering legislation formulated by the Minister of Environmental Affairs to properly manage the disposal of wastes (Crawford *et al.*, 1994). With regards to control of waste disposal, Sections 20 and 24 of the Environment Conservation Act, 1989 apply. As a result of this, the DWAF regards sewage sludge as a hazardous material which should be controlled in the same manner as other hazardous wastes. To the DWAF this means `cradle to grave' control for which adequate legislation does not yet exist. However, the formulation of the Minimum Requirements standards (refer to section described) are relevant as the Department does not intend to have an *ad hoc* policy for sewage sludge but to deal with it within a broader waste disposal policy.

1.8.3 Sewage Sludge Treatment/Disposal

The objectives of sludge treatment processes are to lower the concentrations of organic matter, suspended matter and potential inorganic nutrients thus reducing the pollution potential. At municipal wastewater treatment plants there are usually two stages of treatment of: 1) wastewater treatment; and 2) sludge treatment and disposal. In the wastewater treatment stage a number of physical and biological unit operations follow sequentially whereby low concentrations of particulate and dissolved organic and inorganic pollutants are transformed and removed from the wastewater stream as a low volume high concentration sludge to leave a clear water stream as final effluent. After treating the wastewater to an environmentally acceptable standard, the sludge must be treated/disposed of in such a way to prevent pollution of the environment (Ekama, 1992).

There are five key stages in the treatment of wastewater at municipal wastewater treatment plants (Solt and Shirley, 1991; Montgomery and Montgomery, 1994). These are:

a.   Preliminary treatment, mainly to remove grit, heavy solids and floating debris;
b.   Primary treatment, to remove a substantial portion of the suspended matter;

c.   Secondary treatment, an aerobic or anaerobic step in which oxidisable organic material is removed by microorganisms;

d.   Tertiary treatment, to remove specific materials (for example, ammonia, phosphates); and

e.   Sludge treatment, which is designed to render safe and dispose of organic materials and organisms sedimented in other stages.

Preliminary Treatment

This phase of the treatment process removes grit and heavy solids as well as all floatable materials, and thus protects pumps, valves and pipelines from damage or clogging (Bruce and Davis, 1989). According to Ross, Novella, Pitt, Lund, Thomson, King and Fawcett, (1992) the majority of digester failures are caused by an accumulation of grit or scum, as a result of sub-standard performing screening and grit removal units.

Primary Treatment

Primary settlement of sewage removes up to 70% of the suspended matter and 40% of the BOD by flocculation, adsorption and sedimentation (Steel, 1953). The hydraulic loading rate of the wastewater is reduced and the heavier solids settle by gravity to the bottom of the tank where they are collected and removed, whilst the buoyant material floats (Montgomery and Montgomery, 1994).

Secondary Treatment

The purpose of secondary treatment or sludge stabilization is twofold; to substantially reduce the number and prevent regrowth of pathogenic organisms and, thereby, minimise the associated health hazard; and to reduce the odours and putrescibility (Ekama, 1992; Bruce, 1991).

Processes which may be used to stabilise sludges include: biological stabilization;

chemical stabilization; composting; heat treatment; irradiation; and lagooning.

1.8.4 Biological Stabilization

Aerobic Treatment

Biological treatment effects the removal biodegradable organic materials (expressed as Biochemical Oxygen Demand (BOD)) from water/sewage and also often includes biological oxidation of ammonia to nitrate and, occasionally, the removal of phosphate (Solt and Shirley, 1991).

Two principal aerobic treatments of wastewater exist. Firstly, the fixed film process which is the oldest form of wastewater treatment and includes trickling filters (biological filters) and rotating biological contactors (RBC) (Montgomery and Montgomery, 1994). In these processes the microorganisms grow as a film or a slime on the surface of coarse supporting media or on the discs of the RBC. Therefore, since the microorganisms are attached, no sludge return is necessary (Solt and Shirley, 1991). The second, the dispersed growth or activated sludge process, is a popular method for large-scale treatment works. It comprises a large vessel, with provision for aeration, which is constantly provided with organic matter and a mass of microorganisms which grow in flocs (Section 1.4.4).

Nutrients, trace elements and other growth factors required for balanced biological growth are usually present in non-limiting concentrations in normal domestic wastewaters, while toxic materials are not often present at bactericidal/bacteriostatic concentrations (Barnes, Bliss, Gould and Vallentine, 1981). Oxygen requirements, on the other hand, have to be satisfied by aeration of the sludge and this normally accounts for >75% of the power consumption on the treatment plant. Further, the requirements for a sewage works to produce a fully nitrified effluent more than doubles the power needed for aeration and, as a consequence, nearly doubles the power consumption of the whole works (Mosey, 1980).

Anaerobic Digestion

Anaerobic digestion is a process whereby microorganisms convert organic compounds (consisting mainly of proteins, lipids and carbohydrates) to methane, carbon dioxide and cellular material in the absence of oxygen (air). Anaerobic digestion is an expensive but effective treatment method rendering the sludge suitable for utilisation on farm land. Other desirable features, which help to offset the costs, are the production of methane and the reduction in solids content facilitating further dewatering by simple settlement in the secondary digesters, and thus further reducing sludge disposal costs (Mosey, 1980). Other advantages are the removal of fats and grease as well as the obnoxious odour of pretreated sludge, and a reduction in the numbers of pathogenic organisms (Ross *et al.*, 1992). The disadvantages of the process are the slow bacterial growth rates which result in long start-up periods and limit the flexibility of the process to adjust to changing feed loads, temperatures and other environmental conditions (Ross *et al.*, 1992).

Anaerobic treatment invariably involves a wide variety of organisms which exhibit a great complexity of interactions although, in general, three trophic groups of microorganisms can be identified (Leach, 1994b, Corbitt, 1990). These are graphically illustrated in Figure 1.1 and may be categorised as:

a.　Hydrolytic organisms which catabolize the complex molecules in the wastewater and produce smaller molecules, especially short chained organic acids, as end products:

b.　Hydrogen-producing organisms (heteroacetogens) which mainly utilise the products from hydrolytic catabolism and, in turn, produce, primarily, acetic acid together with propionic acid and butyric acid: and

c.　Methanogens which produce methane and carbon dioxide. These may be divided into hydrogen utilisers and acetic acid users. Generally, most of the problems occurring in the anaerobic digestion of sludges are associated with the methanogenic

stage of the reaction as the methanogens are highly sensitive to environmental stresses (Barnes et al., 1981).

Figure 1.1 Simplified Relationships Between the Groups of Organisms Involved and Products Formed in Anaerobic Digestion
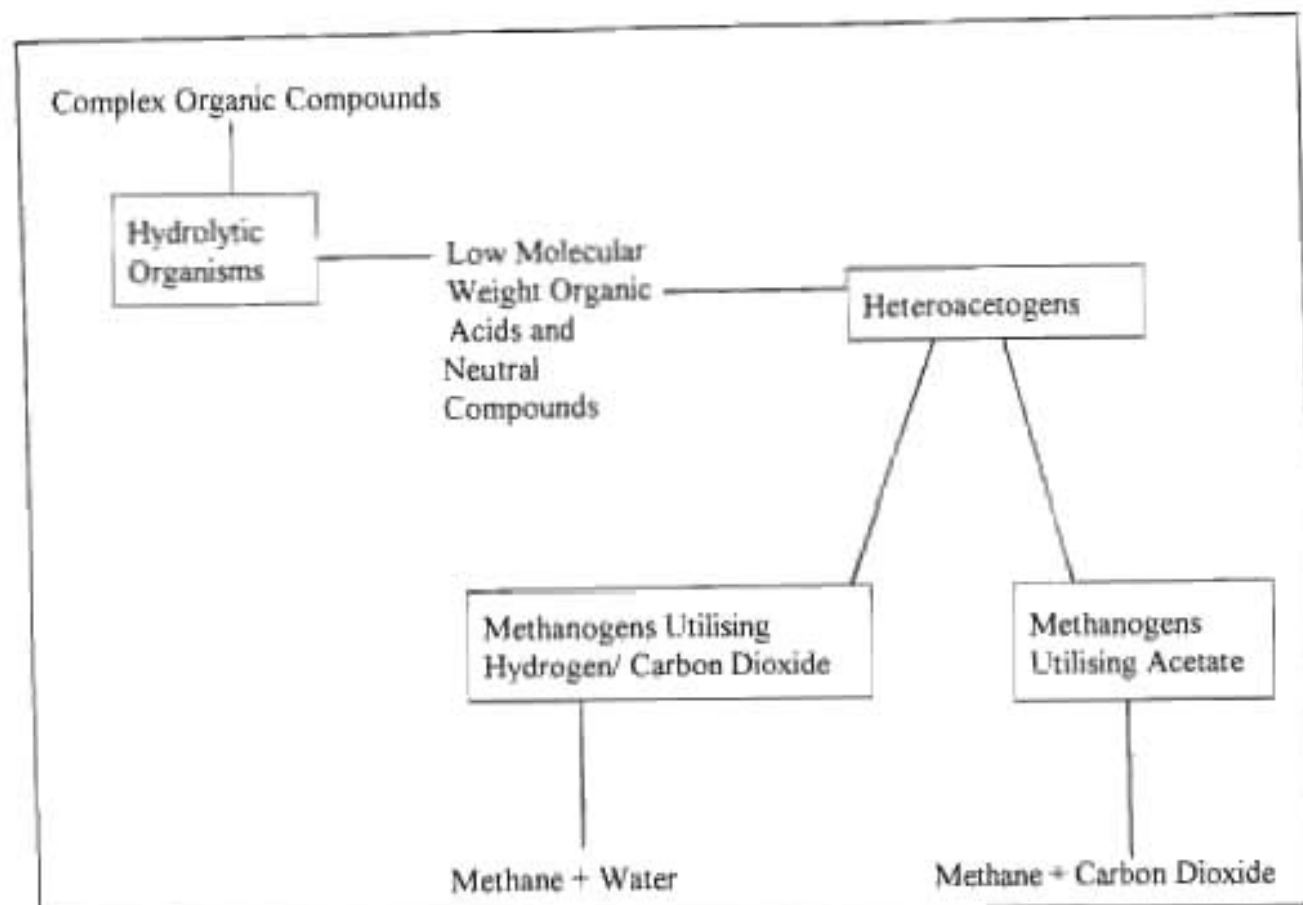
The above classification is, however, an over simplification as bacteria utilising SO_4^{2-} or NO_3 as electron acceptors can also oxidise organic material. Sulphate-reducing bacteria, such as *Desulfovibrio* spp., reduce sulphate to hydrogen sulphide while denitrifying bacteria reduce nitrate to nitrogen gas and other nitrous oxides (Senior, 1986; Senior and Balba, 1990). However, the end result of the digestion process is a well stabilised sludge in which 40 to 60% (w/w) of the volatile solids have been destroyed and biogas has been generated which consists of between 60 and 75% (v/v) methane, with the remainder accounted for by carbon dioxide (Ross et al., 1992).

Anaerobic microorganisms can exist either as suspended growth or fixed films. An example of a suspended growth system is the anaerobic upflow sludge blanket (AUSB) reactor which was discussed in Section 1.4.4. Currently, two-stage anaerobic digesters are the favoured process. These consist of a primary digester, which is mixed to ensure effective distribution of the feed sludge and active organisms in the bioreactor, and which is, preferably, heated to control the temperature, and a secondary digester, which is neither mixed nor heated but is used mainly to facilitate the separation of the digested sludge from the supernatant. Digested sludge is removed from the secondary digester for further treatment, such as dewatering and disposal, while the supernatant, which has a high BOD, is returned to the aerobic treatment plant for further treatment (Barnes *et al.*, 1981; Ross *et al.*, 1992). All fixed growth systems provide some form of solid support for microbial attachment and, hence, biomass retention, and has been described in Section 1.4.4.

It has been shown that combining aerobic thermophilic and anaerobic digestion of sewage sludge has numerous advantages such as reduced stabilization times, the generation of methane and the possibility of heat transfer from the stabilized sludge to the raw sludge (Loll, 1989).

1.8.5 Chemical Stabilization

Chlorine oxidation involves the application of high doses (> -200 mg l^{-1}) of chlorine gas directly to the sludge in an enclosed reactor. A stabilized sludge results which is suitable for dewatering (Forster, 1985). Lime treatment is, however, the preferred treatment and involves the addition of lime to raise the pH to above 11. This stabilizes the sludge and kills the pathogens (Haug, Kuchenrither, Oerke, Prakasam, Soszynski and Zenz, 1992).

1.8.6 Other Methods

Sludge composting is an alternative method. This is an aerobic process and may involve the use of the in-vessel, static pile or Windrow methods (Hill 1989). The sludge is

maintained at a minimum operating temperature of 40°C for five days, during which, for four hours, the temperature must exceed 55°C (Corbitt, 1990). Existing methods of composting sewage normally use dewatered sludge cake as a starting material although a novel process is the composting of liquid and untreated sewage sludge utilising cereal straw to absorb the liquid component (Matthews and Border, 1991).

Irradiation of sludge, although presently not cost effective, is expected to increase in the future. In beta ray irradiation the sludge is irradiated with beta rays from an accelerator at dosages >-1.0 megarad at room temperature (20°C). Gamma radiation, on the other hand, utilises gamma rays from specific isotopes, such as ^{60}Co and ^{135}Cs at dosages >-1.0 megarad at room temperature (Haug et al., 1992).

Thermophilic aerobic digestion entails agitating the liquid sludge with air or oxygen to maintain aerobic conditions during the 10 days residence time at 55°C to 60°C. This method can result in a volatile solids reduction of $>38\%$ (Hamer, 1989). Pasteurization of sludge involves heating to temperatures above 70°C for at least two hours. This effectively stabilizes the sludge prior to disposal (Ekama, 1992).

Aerated lagoon treatment is a biological process whereby attempts are made to increase the oxygenation of the sludge by use of surface aerators or diffusion systems (Solt and Shirley, 1991). These systems are usually employed in small-scale plants where land is not too expensive and where seepage pollution of groundwater is not a problem (Barnes et al., 1981).

1.8.7 Sludge Conditioning

Sludge conditioning *per se* is a treatment process specifically designed to improve the dewatering characteristics of the sludge (Bruce and Davis, 1989). Chemical conditioning and heat treatment are the most common methods used and are designed to enhance the aggregation of suspended sludge particles and, thus, optimise floc formation.

Chemical conditioning involves the use of chemical flocculants to coagulate sludge solids and release sorbed water. This can be effected by either inorganic chemicals, such as activated sodium silicate, where the process is predominantly associated with charge neutralisation, or organic polymers, to bridge the sludge particles. Since most suspended sludge particles are negatively charged, cationic conditioning agents are used to effect charge neutralisation. In some instance, however, anionic polymers are also used (Geldenhuys, 1992). Lime stabilisation can also be part of the sludge conditioning by enhancing the aggregation of suspended sludge particles to optimise floc growth.

Heat conditioning is effected at temperatures of between $150^{\circ}C$ and $260^{\circ}C$ and pressures of 1035 to 2760 kPa. The cellular composition of biological sludges breaks down and the solid fraction is composed of mineral matter and cellular residues, which facilitates sludge dewatering (Haug *et al.*, 1992).

1.8.8   Sludge Concentration

Sludge is usually low in solids, therefore, one of the most crucial processes is to reduce the liquid content. Sludge concentration usually precedes dewatering (and in some cases stabilization) processes for the purpose of thickening can achieve this simply with the only basic requirement, being tanks where the sludge can be stored for several days (Forster, 1985).

1.8.9   Sludge Dewatering

Dewatering is a sludge handling process which removes sufficient water so that the sludge transforms from a fluid state to that of a damp solid or sludge cake. Dewatering of sewage sludges is, generally, a prerequisite for all major disposal routes although it is an expensive and difficult step (Smollen, 1990).

Following thickening, some form of dewatering to $> 15\%$ (m/v) dry solids is usually attained which equates to 77% of the water being removed (Geldenhuys, 1992).

Drying beds are probably the simplest method for dewatering sludges although filter presses are the most widely used in the UK (Forster, 1985; Bruce and Davis, 1989). Belt presses and centrifugation offer further alternatives (Hodgkinson and Rencken, 1992).

1.8.10 Ocean Dumping

This disposal method has been discussed in Section 1.5.1.

1.8.11 Incineration

The cost-effectiveness of sludge incineration is significantly affected by the auxiliary fuel requirement which is a function of the sludge composition (moisture and inert and combustible materials). Therefore, the treatment of sewage sludge by incineration is a two-step process involving, firstly, some form of dewatering of the sludge and, secondly, the combustion of the solids (Corbitt, 1990).

Incineration of industrial and sewage sludges is also coming under severe pressure due to inimical atmospheric emissions. For example, the construction of a 600,000 tonnes capacity waste incinerator in the most densely populated province of the Netherlands, which includes the Hague, Leiden and Zoetermeer, has been cancelled due to growing public concerns and the wastes are now to be disposed of by landfilling (Anon, 1992).

These moves away from ocean dumping and incineration will surely increase the amount of sewage sludge to be co-disposed in sanitary landfills. Evidence presented by de Beker and van den Berg (1989) suggested that increased landfilling of sludge was already imminent in the Netherlands. For this reason, it is vital that our understanding of sludge co-disposal with domestic refuse should increase to ensure that disposal operations effect minimal environmental impacts.

1.8.12 Residue Disposal

The design of a sludge-handling system must include a method for final disposal of sludge residue. Due to the abovementioned restrictions on ocean dumping, final disposal generally involves some type of application to land and usually involves landfilling or application to agricultural land as well as utilization for land reclamation. Even though incineration might be an effective treatment process there always remain a final residue which must be permanently disposed of usually in landfill.

Land Application

Applying sludge to the land is a popular method because of the relatively simple operating requirements and low operating costs if suitable land is nearby. Also, since sewage sludge contains considerable quantities of organic matter and essential plant nutrients, it is an excellent soil conditioner for agricultural land (Korentajer, 1991) and for reclaiming disturbed land (Byrom and Bradshaw 1989).

Depending on the soil and sludge characteristics, the soil will filter, buffer, sorb and/or chemically and biologically react with the sludge (Corbitt, 1990). On the other hand sewage sludge improves the soil characteristics, such as texture, to permit easier root penetration, enhanced water retention capacity decreased soil bulk density and increased hydraulic permeability (Van Niekerk, Richards and Duvenhage, 1988)

However, opposition to this disposal method has grown mainly due to fears of groundwater pollution, odour production and contamination of the food chain (Feachem, Bradley, Garelick and Mara, 1981). According to Ekama (1992), the problem of potential odours is easily dealt with if the applied sludge is immediately ploughed into the soil. Further, an application rate of 8 t dry weight ha⁻¹ y⁻¹, with a solids concentration of 5 % (w/v), would introduce the same volume of liquid as 20 mm of rain, which could clearly be absorbed by the soil and, therefore, no groundwater pollution would result

However, the contamination of the food chain is still a controversial subject. Due to the possible health hazards, it is advisable that the application of sewage sludge to agricultural land should be strictly controlled in order to minimise the risks associated with this practice. Contamination of the food chain, with trace elements and metals, pathogens and toxic organic chemicals can result from using sewage sludge as a soil conditioner.

The concentration of heavy metals in sewage sludge is a matter of great concern at present and many countries have set limits for the total annual amount of sludge used for land application and the concentrations of heavy metals in these sludges (Strauch, 1990). However, numerous researchers and experts in the field believe that trends in legislation are unnecessarily stringent (Wu, 1987). For example, Byrom and Bradshaw (1991) indicated that, if the recommended limits are adhered to, heavy metal concentrations will not increase above recommended limits, even despite repeated sludge loadings.

Until 1984, there were no guidelines in existence in South Africa for the application of sewage sludge to agricultural land, specifically with respect to potential inorganic chemical contaminants. Therefore, a report to the Water Research Commission made by the CSIR Division of Water Technology, using experience gained by various overseas countries, suggested guidelines for the application of disinfected sewage sludge to agricultural land in South Africa. The maximum permissible concentrations of various inorganic compounds were suggested and these have been incorporated in the DNH&PD guidelines and are given in Table 1.6.

Further, the DNH&PD guidelines also specify the maximum application rate per annum of some metals as well as inorganic contaminants, and the total maximum permitted accumulation thereof in soil (van der Merwe and Vivier, 1994).

Pathogenic organisms are inherently contained in sewage sludges worldwide and can survive the treatment processes in a sewage works, and can, therefore, be found in the sludge (Feachem et al., 1981). Thus, to prevent infections of animals and man via the food chain resulting from agricultural utilisation of sewage sludge, some countries have

enacted legal regulations. For example, in Europe, Switzerland allows only 'hygienized' sludge to be applied on crops for animal feed.

Table 1.6   South African Guidelines for Maximum Permissible Concentrations of Inorganic Chemical Contaminant in soil and Permissible Application Thereof (van der Merwe and Vivier, 1994)

Contaminant	Maximum Permissible Content in Soil (mg kg^{-1} dry weight)	Maximum Amount of Metal and Inorganic Contaminants that can be Applied to Soil g ha^{-1} yr^{-1}
Cadmium	2	160
Chromium	80	14000
Cobalt	20	800
Copper	100	6000
Lead	56	3200
Nickel	15	1600
Zinc	185	22000
Mercury	0.5	80
Arsenic	2	120
Selenium	2	120
Molybdenum	2.3	200
Boron	10	640
Fluoride	50	3200
pH	5.8-7.8	

A sludge is considered to be 'hygienized' if at the time of delivery it does not contain more than 100 *Enterobacteriaceae* per gram of sludge nor any viable worm eggs (Strauch, 1991). In South Africa, the DNH&PD guidelines indicate that *Type A* and *B* sludges will contain pathogenic organisms as little or no stabilisation of these sludges has occurred. For *Type C* and *D* sludges it is envisaged that stabilisation will result in no viable *Ascaris* ova, and a maximum of O *Salmonella* organisms and 1000 faecal coliform organisms per 10 grams dry sludge immediately after treatment (van der Merwe and Vivier, 1994).

The occurrence of organic priority pollutants in raw sewage has not been as completely characterised as the heavy metals although it is known that raw sludge often contains volatile compounds, such as 1,1,1, trichloroethane, chloroform, ethyl benzene, methylene chloride and toluene, and semi-volatiles such as phenol together with low concentrations of pesticides and polychlorinated biphenyls (Gschwind, Harper, Kelada, Lordi, Richardson, Soszynski and Sustich, 1992). The DNH&PD guidelines do not deal with organic toxins in any detail in the sense that none of these are specified. However, according to Ekama (1992), at this time there is not much knowledge on toxic organic compounds in South African sludges and, therefore, before this issue can be addressed in the guidelines, a survey of the toxic organic compounds commonly found would need to be made.

Landfilling

According to Brunner and Lichtensteiger (1989), a literature search through files of Water Resource Abstracts, Environmental Bibliography and the Pollution Abstracts yielded 1026 citations on the agricultural utilisation of sewage sludge, and only 142 citations (12%) on landfilling of sludge. This is surprising considering the fact that about half of the sewage sludge generated in Europe is landfilled (Brunner and Lichtensteiger, 1989). This paucity of studies on the fundamental microbiology and biochemistry of refuse catabolism particularly in respect to co-disposed sewage sludge has also been noted by Senior and Balba (1987).

The disposal of sewage sludge to sanitary landfills is by no means a new concept. In the Netherlands, for example, this disposal option has been practised for centuries (Beker and van den Berg, 1989). In general, and worldwide, landfilling accounts for a major proportion of sludge disposal. For example, in the United States, where landfilling of wastewater sludge is viewed as, primarily, a disposal method, about 25% of the generated sludge is landfilled (Novella, Ross, Lord, Stow, Fawcett and Greenhalgh, 1994). Further, in 15 selected European countries, an average of 43% of all sewage sludge is

landfilled, with the common practice comprising dewatering, lime stabilisation and co-disposal with MSW (Brunner et al., 1989)

According to Novella et al., (1994) the practice of sludge co-disposal in sanitary landfills in South Africa has not been as widely implemented due to various reasons including the relatively dry climate which improves the operation of drying beds. In the Western Cape, sludge from most sources has mostly been accepted as a good, cheap soil conditioner and is currently widely used by vegetable farmers as a fertiliser. These practices are expected to decrease with the implementation of the DNH&PD guidelines relating to sludge disposal and utilisation.

The co-disposal of sewage sludge with refuse is regulated in South Africa by Section 20 of the Environmental Conservation Act, Act 73 of 1989. The permit issued stipulates the hazard rating of the sludge and the co-disposal ratios to be employed, which usually do not exceed 1:20 (liquid to solid) (Crawford et al., 1994). Since co-disposal of sludge and domestic refuse is practised at various landfill sites in South Africa (Ross, 1990; Novella et al., 1994) the pressures on this biotechnology is likely to increase.

The relative prominence of this disposal method may be attributed to several factors, of which the principal one is, undoubtedly, cost (Watson-Craik, Sinclair and Senior, 1992b). Further, all ocean dumping of sewage sludge is coming under international pressure (Wu, 1987) and, in general, there is much less national regulation on landfilling than on any other sludge disposal option (Brunner et al., 1989).

1.8.13 Co-Disposal of Sewage Sludges with Refuse

In general, co-disposal with municipal solid waste (MSW) is the favoured landfilling method for treated sludge wastes although monofills are now the chosen option in Switzerland (Lichtensteiger, Brunner and Langmeier, 1989).

Operation

The method followed by West Yorkshire Waste Management, UK, is in accordance with the guidelines of Waste Management Paper NO.26 (Hill, 1989). Briefly, this involves depositing the sludge at the foot of the working face before spreading evenly in a thin layer, approximately 0.25 m thick, over the underlying refuse. Loose refuse is then pushed down the inclined working face onto this layer, always ensuring that there is sufficient thickness to prevent the sludge forcing its way to the surface. Using this technique, it is possible to co-dispose crude controlled waste and sewage sludge in the ratio of 3:1 (w/w) (Hill, 1989). Sludge may also be disposed in a pre-excavated trench in advance of the working face, or in one formed within the body of the refuse mass in a manner similar to that used for the disposal of liquid wastes (Sinclair, 1994).

Site Water Balance Considerations

According to Hill (1989), liquid sludges (4-8% dry solids), by their very nature, are unsuitable for co-disposal. The high water content makes the landfill working conditions extremely difficult and increases the likelihood of unacceptable quantities of leachate being produced. Conversely, dewatered sludges (20-30% dry solids), lend themselves more favourably to co-disposal operation. Van den Berg, Geuzens and Otte-Witte (1991) indicated that co-disposal of sludge, with 35% dry solids, was feasible and that the sludge/waste ratio was not very critical. This dry solids content can easily be achieved for filter press sludge, conditioned with lime and ferric chloride, and for thermally-conditioned sewage sludge. For sewage sludge conditioned with polymers and dewatered with a sieve belt press or a centrifuge, a weight ratio of refuse and sludge of approximately 7:1 is recommended (van den Berg *et al.*, 1991).

1.8.14 Effects of Sewage Sludge Co-Disposal on Refuse Stabilization

According to Senior (1991), co-disposal practices are, fundamentally, a superimposition on the normal refuse catabolic processes. Therefore, a comprehensive

understanding of the interdependent effects of refuse catabolism with added compounds is essential.

Co-Disposal of Stabilised and Anaerobically-Digested Sewage Sludge

Blakey (1991) co-disposed three different types of sewage sludge (raw dewatered, primary/mixed sludge and liquid digested sludge) with refuse in a laboratory-scale experiment. The results indicated that constituents (such as total organic carbon (TOC), biochemical oxygen demand (BOD), chemical oxygen demand (COD), sulphate, iron, manganese, zinc, nickel and lead) leached from the co-disposal reactors were less than for those recorded from the domestic waste only controls. However, the masses of ammoniacal nitrogen and total phosphorus released were found to be higher in the co-disposal reactors.

Further, methane generation and refuse stabilization were both enhanced as a result of the co-disposal of the sewage sludges. Watson-Craik, Sinclair and Senior (1992b) also reported increases in methane release rates in co-disposal columns compared to refuse only controls. However, in their study, the columns operated with leachate recycle and with activated sludge: refuse loading ratios of 5.9:1 and 9.7:1 the leachates had lower residual nitrogen concentrations.

Studies which have recorded enhanced refuse stabilization and gas production also, showed lower COD concentrations in the leachate compared with refuse controls (Chapman and Ekama, 1991). For example, Blakey (1991) reported that during the methanogenic phase the chemical oxygen demand of leachate from refuse co-disposed with de-watered sludge was 50% lower than the equivalent refuse concentration, while the leachate COD concentration from refuse receiving un-dewatered or liquid digested sludge was only 7% lower. In contrast, the experiments of Barlaz et al., (1987) revealed increases in COD concentrations for leachate samples in response to sludge additions.

Sewage sludge addition to landfill should be appraised in three ways: firstly, on a

hydraulic loading basis; secondly, by applying current permissible standards for heavy metal application to landfill; and thirdly, on the organic loading rate. In terms of sludge metal loading, the current maximum limits are 100 g per tonne of refuse. The two exceptions are cadmium and mercury where loadings of 10 g and 2 g per tonne of refuse, respectively are considered more appropriate (U.K. Department of Environment, 1988). It is, therefore, encouraging to note that numerous researchers have shown that co-disposal of sewage sludge, which contains relatively high concentrations of heavy metals, significantly reduces the quantity and concentrations of metals leached (Blakey, 1991).

Co-Disposal of Anaerobically-Digested Sewage Sludge

Co-disposal of anaerobically digested sewage sludge with domestic refuse in a landfill has been shown to benefit landfill stabilization (Wise, Leuschner, Levy and Sharaf, 1986; Fletcher, 1989; Leuschner, 1989; Stegmann and Spendlin, 1989). However, other studies have not revealed any positive effects (Chapman and Ekama, 1991; 1992; Pacey, 1989b; Barlaz, Milke and Ham, 1987; Novella, 1992). Unfortunately, the studies cannot be compared directly since different amounts (Novella, 1992) and different types of sewage sludge (Leuschner, 1989) were added and at different times. There is also a disparity in the type of refuse used. For example, Stegmann et al., (1989) used fresh shredded and hand-sorted refuse whereas Novella (1992) used municipal solid waste as placed in a landfill site.

The co-disposal of anaerobic sewage sludge with refuse holds promise for dealing simultaneously with two problems of waste management: finding an environmentally-friendly method for sewage sludge disposal since factors such as high concentrations of heavy metals, pathogens and other toxicants often militate against the use of sewage sludge in agriculture as a fertiliser (Ross, 1990); and accelerating refuse stabilization and, thus the onset of methanogenesis (Chapman and Ekama, 1992). Therefore, apart from the environmental benefits of co-disposal, significant financial savings can also be realized since about half of the operating costs of a sewage treatment plant are associated with the treatment and final disposal of the sludge (Chapman and Ekama, 1991).

Addition of anaerobically-stabilized sewage sludge to a refuse mass is possibly beneficial for the accelerated onset of methanogenesis in the following ways (Buivid et al, 1981; Watson-Craik, Sinclair and Senior, 1992b):

a   The liquid sludge significantly increase the moisture content of the refuse. The moisture content of refuse during active degradation is perhaps one of the most important governing abiotic factors (Senior et al., 1991). Typically, municipal solid waste as delivered contains between 10 and 30% (w/w) water which is the minimum moisture needed for anaerobic microorganism survival and methane production (Buivid et al., 1981). Sewage sludge, with a solids content of 2% to 3%, is considered an ideal medium which can be added to increase the moisture content of the refuse during landfilling operations (Novella, 1992). Increasing the moisture content above the field capacity of the refuse could, however, result in increased leachate volumes;

b.   The sludge provides an inoculum of methanogenic bacteria to the refuse. As a result of refuse placement strategies the refuse will accumulate significant concentrations of air which inhibit the methanogens. In contrast, acetogenic bacteria flourish in the presence of oxygen and will continue to produce acids which could inhibit methanogenesis. The addition, therefore, of methanogenic bacteria is thought to promote a balanced bacterial ecosystem (Pacey, 1989b);

c.   An increase in the concentrations of essential nutrients especially phosphorus and nitrogen. Not surprisingly, most of the interest regarding nutrient availability from sewage sludge has been in connection with agricultural uses and land reclamation schemes, with little such focus being given to landfilled sludge wastes (Sinclair, 1994);

d.   Alkalinity, provided by ammonia fixation of organic nitrogen in the sludge, buffers the liquid fraction against pH changes. Chapman and Ekama (1991) found that the anaerobic sludge added between 1800 and 2300 mg l^{-1} H_2CO_3 alkalinity (as $CaCO_3$) to the liquid fraction of the refuse; and

e. Refuse often has a carbon:nitrogen ratio that is not suitable for optimum catabolic conversion. For optimum anaerobic fermentation the C:N ratio should be in the range of 20-30:1. Typically, MSW has a ratio much greater than this (Senior, 1991) and, thus, the addition of sludge tends to correct the imbalance.

Possible disadvantages of co-disposal of anaerobic sludge with refuse include elevated leachate ammonium concentrations, increased volumes of leachate and, hence, leachate treatment costs, lowered refuse temperatures and the necessity to de-water the sludge first to 25-30% (w/w) solids (Watson-Craik, Sinclair and Senior, 1992b). A further consideration is the workability and manoeuvrability of the landfill compactor which is greatly inhibited at higher sludge:refuse ratios. It was found at the Coastal Park landfill site in Cape Town, for example that the lowest practical ratio which could be employed was 4,5:1 (refuse:anaerobically-digested sludge) before the conditions made the heavy vehicles inoperable (Novella, 1992).

A few researchers concluded that anaerobically-digested sludge additions inhibited the methanogenic stage of refuse degradation (Chapman and Ekama, 1991; Barlaz et al., 1987). Studies made by Chapman et al., (1991) indicated that anaerobically-digested sewage sludge additions to "fresh" refuse were a distinct disadvantage as they could severely inhibit the onset of methanogenesis. In their study, six lysimeters were poised at a moisture content equal to field capacity (65% w/w) by the addition of water in the case of the control, and sewage sludge for the test lysimeters. The short-chain fatty acid (SCFA) concentrations of the leachate from the test lysimeters were consistently higher than the control. This was also evident in the lower pH values of the same test reactors. It was calculated that the sludge added between 1800 and 2300 mg l^{-1} H_2CO_3 alkalinity (as $CaCO_3$) to the liquid fraction of the refuse. However, this gain in alkalinity was lost after only eight weeks of lysimeter operation as a result of the high concentrations of short chain fatty acids produced, which neutralized the alkalinity gain. Subsequently, these high acid concentrations decreased and the leachate pH stabilised.

Barlaz et al., (1987) investigated the effects of both partially-degraded refuse and anaerobically-digested sewage sludge inoculations on refuse degradation. Methane production began almost immediately with the former but only very low rates were

apparent with the latter. It was, therefore, concluded that sludge additions to enhance refuse degradation were not successful. The same results were also obtained by Stegmann and Spendlin (1986) who recorded enhanced methane production with additions of partly-composted MSW but not with sewage sludge. The researchers concluded that the sewage sludge concentrations added were probably insufficient to act as an effective inoculum. The positive effect of the addition of partly-composted refuse might have been due to organic acid dilution rather than to microbial inoculum addition *per se*.

The objective of this research programme was to make a fundamental definitive study of co-disposal relative to the needs of South Africa and so underpin a detailed Code of Practice. Further, key questions to be addressed in this study are:

1. Which are the hydraulic and organic loading rates required to obviate inimical environmental impacts?
2. Can a potential environmental impact be negated by dual co-disposal?

To satisfactorily answer these questions, and since the use of ecologically-realistic models was central to the programme, it was necessary to determine the relative effects of the co-disposal practices on both the free-living (growth rate-dependent) and fixed (growth rate-independent) microbial population types. For the former, it has been shown that multi-stage chemostat models can effect separation of the species habitat domains, with retention of overlapping activity domains, and so facilitate examination of species in isolation without violating the integrity of each association. To examine co-disposal in the presence of, especially, growth rate-independent microbial associations, 42 single-stage, glass column bioreactor microcosms were commisioned and subjected to phenol and/or anaerobically digested sewage sludge co-disposal.

The results and findings culminated in the ultimate goal of this study, which was, to provide a detailed and fundamental Code of Practice in terms of co-disposal relative to the needs of South Africa that can be used as guidelines for managerial purposes and effective policy making.

CHAPTER 2

MATERIALS AND METHODS

2.1   Refuse

Approximately one month old refuse was collected from the Pietermaritzburg municipal landfill site and sorted to remove all visible non- degradable fractions such as metal, glass and plastic. 'Aged' refuse was obtained from the Umlazi Landfill Site, Durban. The specific age of the refuse was not known as the refuse was taken from a depth of 2.5 metres out of co-disposal pits which had been dug by a backactor.

In the laboratory the refuse was homogenized with a Haecksel Max 1500 (Steinmax and Co. Model: D-8800 Ansbach) blender. Following this, dry weight determinations were made by drying to constant weight at 105°C for 48 hours.

For the adsorption/desorption experiments fresh, hand sorted, refuse was obtained from the Pietermaritzburg Landfill Site and, subsequently, homogenised with a Haecksel Max 1500 (Steinmax and Co., Model: D-8800 Ansbach) garden blender and stored in a sealed Sterilin autoclavable disposal bag. The refuse was sterilised by gamma radiation (25.5 kGy for 6 hours), and then stored at 4°C until use.

2.2   Sewage Sludge

The anaerobically digested sewage sludge was collected from the Sea Cow Lake Sewage Purification Works, Durban. The sludge was collected in 10 litre plastic drums and stored at 4°C until needed.

2.3   Basic Mineral Salts Medium

The basic mineral salts medium used in this study was adapted from Coutts, Senior

and Balba (1987) and contained the following (g l^{-1} glass-distilled water): K_2HPO_4, 1.5; $NaH_2PO_4.2H_2O$, 0.85; NH_4Cl, 0.9; $MgCl_2.6H_2O$, 0.2; $NaHCO_3$, 0.5; Na_2CO_3, 0.2; trace minerals, 1.0 ml; trace elements, 1.0 ml; nickel solution (1mM), 1.0 ml; vitamins, 1.0 ml; resazurin (0.01% w/v), 1.0 ml.

The trace elements contained (mg l^{-1} glass-distilled water): $FeCl_2.4H_2O$, 1500; $NaCl$, 9000; $MnCl_2.4H_2O$, 197; $CaCl_2$, 900; $CoCl_2.6H_2O$, 238; $CuCl_2.2H_2O$, 17; $ZnSO_4$, 287; $AlCl_3$, 50; H_3BO_3, 62; $NiCl_2.6H_2O$, 24.

The trace minerals contained (mg l^{-1} glass-distilled water): $NaMoO_4.2H_2O$, 48.4; $NaSeO_3.xH_2O$ (31% Se), 2.55; $Na_2WO_4.H_2O$, 3.3.

The vitamins contained (mg l^{-1} glass-distilled water): biotin, 10; p-amino benzoic acid, 19; α-lipoic acid, 20; folic acid, 10; pyridoxine HCl, 20; thiamine HCl, 20; riboflavin, 30; nicotinic acid, 50; D (+) Ca-pantothenate, 30; cyanocobalamine, 20.

For every litre of medium, a mineral salts solution deficient in $NaHCO_3$, Na_2CO_3, trace elements, vitamins and the limiting carbon and energy source was diluted to 900 ml with distilled water. Before the addition of ammonium chloride, the pH of the medium was adjusted to 6.5 with HCl (1N) to prevent any precipitation. The medium was dispensed into flasks, which were then closed with cotton wool plugs and aluminium foil and autoclaved at 121°C (100 kPa) for 15 min.

Appropriate weights of $NaHCO_3$ and Na_2CO_3 were added to 97 ml of distilled water and sterilized by autoclaving, as above, and added following the pH adjustments.

Prior to use of the medium, 1 ml each of trace elements, trace minerals and vitamins together with the appropriate amounts of electron acceptors and carbon source were filter sterilized by passage through 0.2 μm Millipore membrane filters, and added to the medium.

2.4   Inoculum

Hexanoic acid-degrading microbial associations were enriched at 30°C and pH 7.2 in the presence of three different hexanoic acid concentrations (5, 7.5 and 10 mM) with three different inoculum source materials. The latter were obtained from the Umlazi landfill site (Durban) with the first sample taken from soil adjoining an actively gassing waterhole, at the base of the landfill site. The second was taken from soil in the area where the landfill cover was stored with the final sample taken from the municipal refuse. The samples were placed in plastic bags which were sealed and transported to the laboratory where they were stored at 4°C until required.
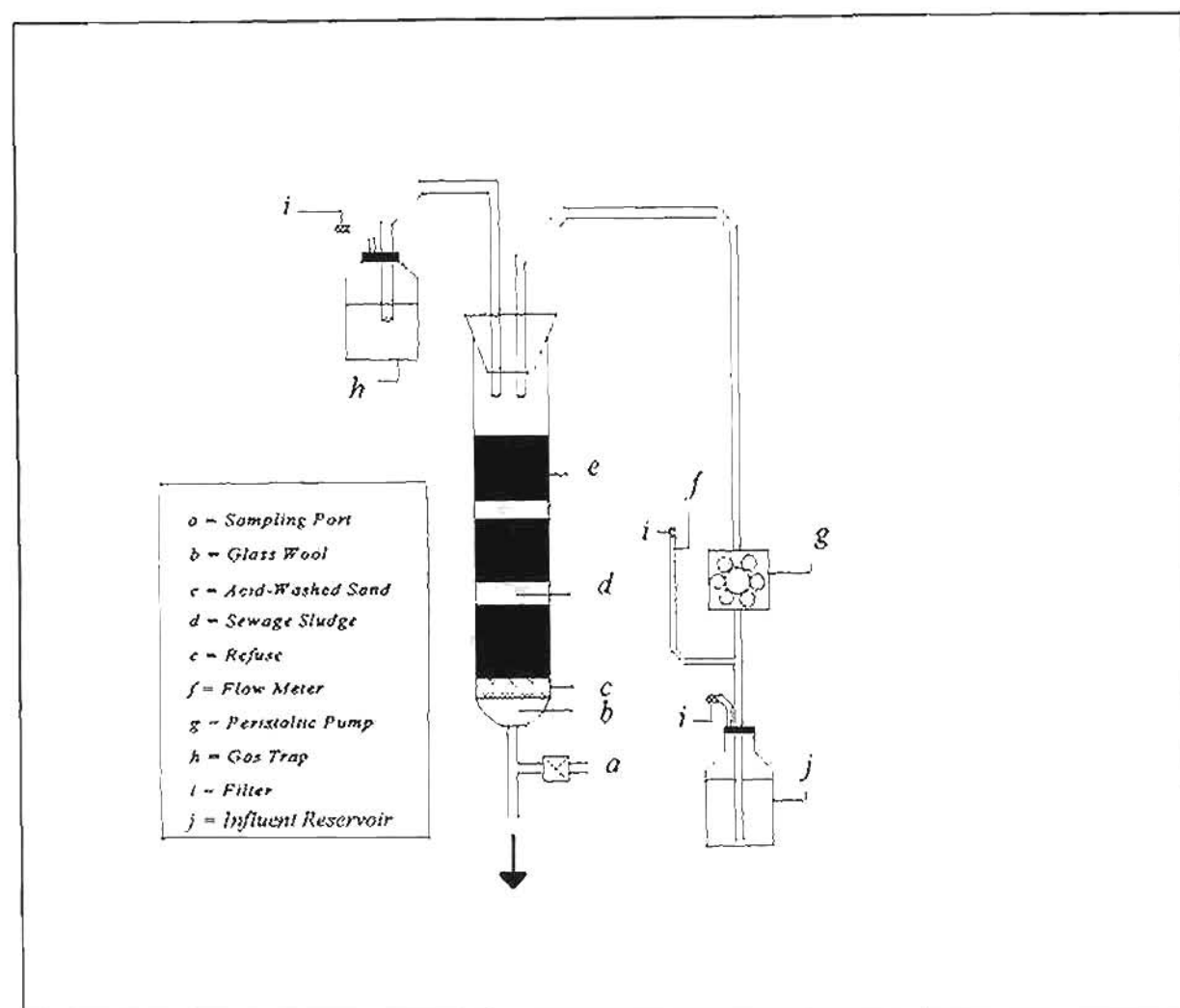
Both aerobic and anaerobic enrichments were made. A 1:10 (w/v) ratio of inoculum source material and medium was used in the aerobic enrichments while a 1:3 ratio was used in the anaerobic enrichments. The anaerobic cultures were made in medical flats (230 ml) filled to a 1:10 (v/v) ratio of culture:gas headspace. These culture were overgassed with oxygen-free nitrogen (OFN) and incubated static at 30°C in the dark. In contrast, the aerobic cultures were made in conical flasks (100 ml) which were incubated at 30°C with shaking (150 rpm).

2.5   Model/Microcosm Configurations

2.5.1 Refuse Microcosms

Thirty-six experimental refuse columns (individual working volume 970 ml) were adapted from the laboratory microcosm system developed by Watson-Craik and Senior (1989a) and mounted in a constant temperature box. The temperature (30°C $\pm$3°C) was maintained by means of a heating element and a regulating thermostat. The individual column design varied according to the operating procedure: single elution (Figure 2.1); or leachate recycle (Figure 2.2).

Figure 2.1   Diagram of a Refuse Microcosm Operated with Single Elution

Legend:
- a - Sampling Port
- b - Glass Wool
- c - Acid-Washed Sand
- d - Sewage Sludge
- e - Refuse
- f = Flow Meter
- g - Peristaltic Pump
- h - Gas Trap
- i - Filter
- j = Influent Reservoir

The base of each column was packed with glass wool (b) and acid-washed sand (c) to facilitate easy outflow of effluent. Polyvinyl chloride tubing (o.d. 6mm) was used throughout and all connections were made with glass.

Gaseous emissions from each column were allowed to vent to the atmosphere via a zinc acetate (1 % w/v) gas trap (h) which was employed to trap hydrogen sulphide as zinc sulphide. The influent medium was introduced into the single elution columns by means of a Watson-Marlow (503 U) peristaltic pump (g) at a constant flow rate of 29.1 ml h^{-1} to give an empty bed dilution rate (D) of 0.03 h^{-1}. The flow rate was frequently checked with a flow meter (f) and any variations, due to the stretching and deformation of the peristaltic tubing, corrected.

Dilution rate is given by:

$$\frac{F}{V} = D$$

<div align="right">2.1</div>

Where   F = flow rate (l h^{-1});

                      V = culture volume (l); and

                      D = dilution rate (h^{-1})

The leachate recycle columns were operated by drawing off the leachate 2-3 times per week by inserting 25 ml syringes (l) at the column bases and allowing them to fill, usually at a slight negative pressure (Figure 2.2). The leachates were then reintroduced to the top of the columns while maintaining anaerobiosis.

Figure 2.2   Diagram of a Refuse Microcosm Operated with Leachate Recycle
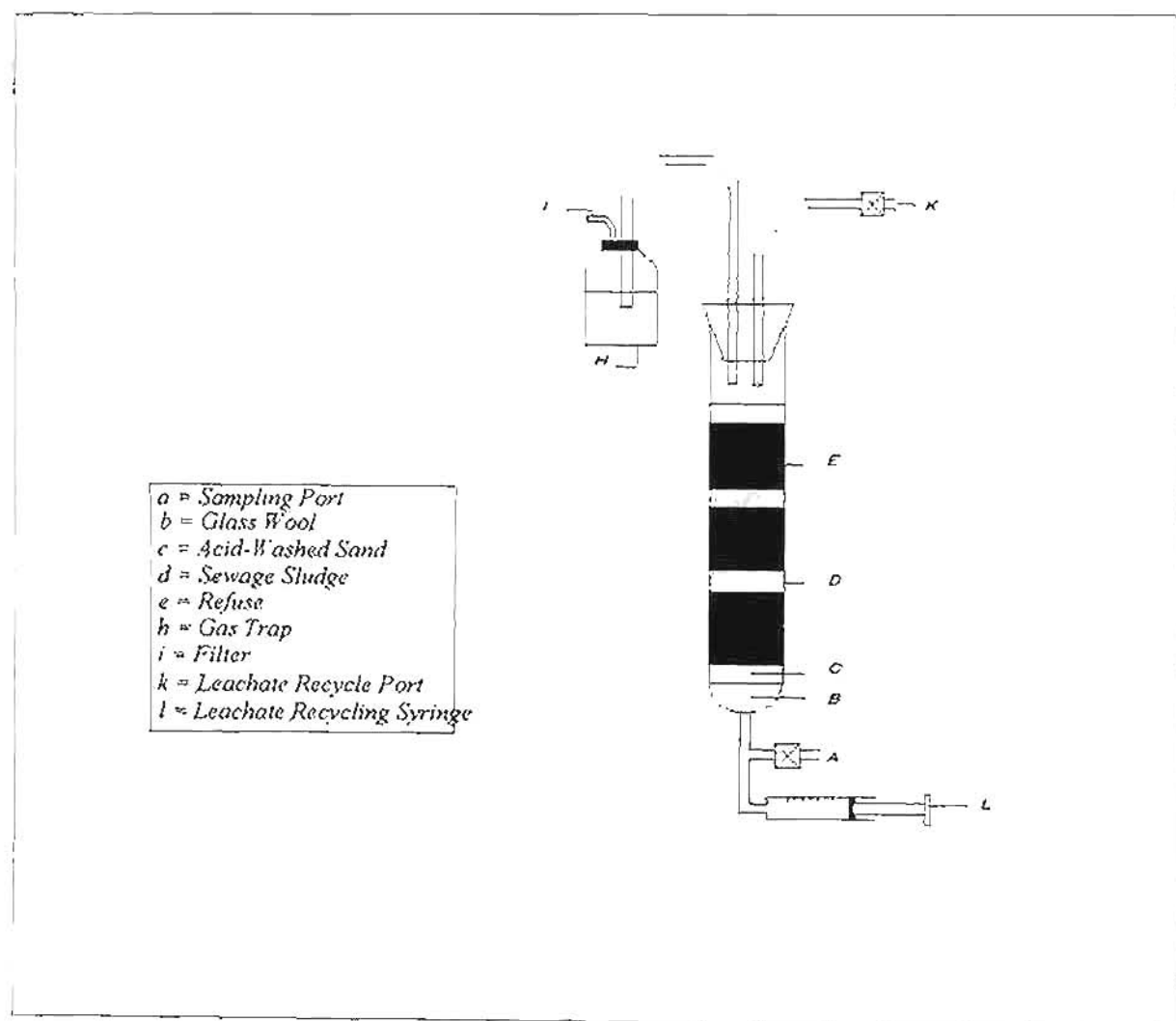
2.5.2 Multi-Stage Chemostat Model

Six 4-stage chemostats were constructed as shown in Figure 2.3, which shows a single-stage of the multi stage chemostat model. The influent medium was introduced (Watson Marlow (Model 202) peristaltic pump) into the first vessel of each array via a glass tube equipped with a line break (a) to minimise microbial growth in the influent tube.

Figure 2.3 Configuration of a Single-Stage of the Multi-Stage Chemostat Model

a = Medium or Culture Inflow and Line B

b = Water Jacket

c = Culture Overflow

d = Influent Medium or Culture

e = Syringe

f = Oxygen-Free Nitrogen Influent

g = Gas Trap

h = U-Tube Assembly and Suba-Seal

i = Aluminium Foil
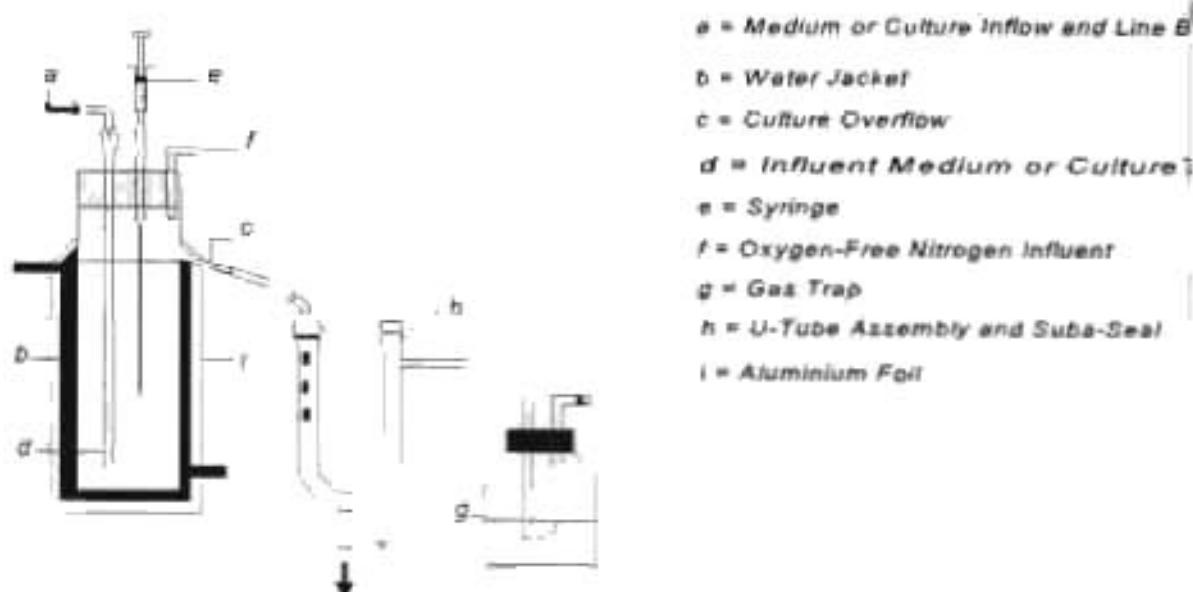
The working volumes of the individual vessels were 155 m/ (1), 331 m/ (2), 736 m/ (3) and 1732 m/ (4) to facilitate a regime of non-constant dilution rates. The dilution rate for Vessel 1 was 0.05 h^{-1} and for the entire system 0.003 h^{-1}.

The culture vessels were maintained at a constant temperature of 30°C by means of water jackets (b) in conjunction with a thermocirculator (Fisons, Haake G). Further, the vessels were arranged so that all of the effluent of the first vessel became the influent of the

second vessel and so on. This was accomplished via an angled effluent overflow tube (c) for which the height was calculated to give the required volume. The culture liquid for each individual vessel was introduced via a medium input glass tube (d) which allowed the influent to be emitted at the base of the vessel. This design ensured that the bulk flow characteristics of the system approximated to a continuous but segmented plug flow (Coutts *et al.*, 1987). A second glass tube was provided which was connected to a sterile syringe (e) for withdrawing culture samples. An overpressure of oxygen-free nitrogen (OFN) was employed to maintain anaerobiosis (f). This was further ensured by a pressure head (g) of zinc acetate (1% w/v) which also served to trap gaseous H_2S. The U-tube arrangement (h) permitted gas sampling by inserting a hypodermic needle through the rubber Suba-seal and facilitated degasification of the effluent after each stage so that no gas entered the succeeding vessel. Oxygen-impermeable butyl rubber tubing was used throughout. Each vessel was covered with aluminium foil (i) to exclude light. The effluent from the final vessel was collected in a 5 litre reservoir and autoclaved (121°C and 100 kPa for 15 minutes) before disposal.

2.5.3   Phenol Adsorption/Desorption Models

Adsorption

Fifteen, sterile (autoclaved at 121°C for 15 min) 250 m*l* flasks, each supplied with a 10 g of sterile refuse and aseptically supplemented with 100 m*l* aliquots of the relevant phenol concentration were prepared in triplicate (Table 2.1). The flasks were then placed in an incubator and shaken (100 rpm) for 12 hours at 26°C. For the adsorption isotherm determination a stock solution of 2500 mg l^{-1} phenol, in K_2HPO_4/KH_2PO_4 buffer, was used following dilution and filter sterilisation (0.45 μm).

Desorption

Duplicate columns (length 17 cm, internal diameter 50mm) were sterilised by soaking in alcohol and then lightly (524 kg m^{-3}) packed with refuse. To facilitate the flow of the influents, glass wool was placed above and below the refuse.

The columns were perfused with sterile (autoclaved at 121°C for 15 minutes), distilled water with a Watson-Marlow flow inducer (Model number 2055), at a flow rate of $1 ml\ min^{-1}$. Simultaneously, the pore volumes were determined. Each pore volume was estimated by the volume of water required to fill the void spaces.

Table 2.1   Phenol Concentrations Used in the Adsorption Isotherm Study

Reactor	Phenol Concentration $(mg\ l^{-1})$
Control	0
Flask A	50
Flask B	100
Flask C	200
Flask D	400
Flask E	600
Flask F	800
Flask G	1000
Flask H	1200
Flask I	1400
Flask J	1600
Flask K	1800
Flask L	2000
Flask M	2200
Flask N	2400

Following displacement of two pore volumes with water the influent was replaced by 200 or 2000 mg l^{-1} phenol. The effluents were coupled to a Gilson (Model 203) microfraction collector from which 10 ml samples from each column were collected every

30 minutes. Sampling was continued until parity was obtained in the influent and effluent phenol concentrations. Following this, the phenol influents (200 or 2000mg l^{-1}) were replaced with water. Collected samples (1.5ml) were centrifuged in a Eppendorf centrifuge (Model 5410) at 14000 rpm x g for eight minutes.

2.6   Leachate Sample Preparation

Leachate samples were abstracted and stored in a deepfreeze ($< -10^0$C) in polypropylene bottles until required. Prior to analytical determinations the samples were first centrifuged (Eppendorf, Model 5413) at 12000 rpm x g for 15 minutes followed by filtration through a Whatman cellulose filter (30 μm).

2.7   Analyses

2.7.1 Phenol

Phenol concentrations were determined with a Varian 3600 gas chromatograph (GC) equipped with a flame ionization detector (FID) in which the temperatures of the injector, oven and detector were 150, 120 and 180°C, respectively. The flow rate of the OFN carrier gas was set at 30 ml min^{-1}. Duplicate samples (1 μl) were injected directly into the glass column (length 2 m, i.d. 2 mm) packed with 5% polyphenyl ether on Chromosorb W-HAPG (80-100 mesh). During the latter part of the study phenol concentrations were determined by GC with a glass column packed with 3% OV101 on Chromosorb W-HAPG (80-100 mesh). The concentrations were determined, after standard curve construction, by mean peak area comparison with phenol standards (100 - 2000mg l^{-1}).

2.7.2 Volatile Fatty Acids

Short-chain volatile fatty acids (VFA) ($C_2 - C_6$) were analysed with the same GC and detector. The samples were prepared by adding 0.1μl of pure formic acid to 0.9μl of culture supernatant and then injecting directly into the stainless steel column (length 2 m,

i.d. 2 mm) packed with 10%/1% FFAP/H_3PO_4 on Chromosorb W-AW (80 -100 mesh). This column, however, proved unsatisfactory as a result of the short working life of the packing which led to baseline drift and irregular peak shape. Therefore, subsequently, a glass column was packed with 5% neopentyl glycol sebacate and 1% H_3PO_4 on Chromosorb (80-100 mesh). For this column the injector and detector (FID) were both maintained at 180°C while the column temperature was increased from 100°C to 160°C at a ramp rate of 8°C min^{-1}. The flow rate of the carrier gas (OFN) was set at 30 m/ min^{-1}. The concentrations were determined, after standard curve construction, by mean peak area comparison with volatile fatty acid standards (500 - 3000mg l^{-1}).

2.7.3 Methane

For headspace methane concentration determinations the Varian 3600 GC was used with the gas samples (100 μl) injected directly into the glass column (length 2 m, i.d. 2 mm) packed with Poropak T (80-100 mesh). The injector, column and detector (FID) were maintained at 120, 35 and 150°C, respectively. The concentrations were determined, after standard curve construction, by mean peak area comparison with methane standards (5 to 50% (v/v).

2.7.4 Ammonia

Dissolved ammonia was determined with an ammonia electrode (Orion 95-12) in conjunction with an Orion 701A digital ion analyser. Concentrations of aqueous samples (0.5 m/) were determined by comparison with an ammonium chloride calibration curve (10-1000 mg l^{-1}).

2.7.5 Sulphate, Nitrate and Nitrite

The concentrations of the electron acceptors sulphate, nitrate and nitrite were determined with a Waters HPLC utilising a Waters (486) conductivity detector in conjunction with a Waters IC-Pac anion column (4.6 x 50 mm) with a particle size of 10

μm. The separation was effected by a boro/gluconate eluent at a flow rate of 1 ml min^{-1}. The concentrations were determined, after standard curve construction, by mean peak area comparison with the relevant standards (5 to 50mg l^{-1}).

Alternatively, sulphate was also assayed according to the standard barium chloride turbidimetric method (Standard Methods, 1985). After the addition of 0.5 ml of conditioning reagent and a standard spoonful ($\pm$0.6 g) of BaCl$_2$ crystals (dry, 20-30 mesh), the sample (10 ml) was mixed with the help of a Whirlimixer for 60 seconds. After standing for 4 minutes, the absorbance of the solution was measured at 420 nm with a Milton Roy (Spectronic 301) Spectrophotometer. Distilled water was used as the blank. The conditioning reagent contained 50 ml glycerol, 30 ml concentrated HCl, 100 ml 95% (v/v) iso-propylalcohol, 75g NaCl and 300 ml glass-distilled water. The sulphate concentrations were calculated by reference to a standard curve (5-35 mg l^{-1} sodium sulphate).

CHAPTER 3

ASSESSMENT OF THE IMPACTS OF PHENOL CO-DISPOSAL WITH REFUSE ON REFUSE CATABOLISM

3.1 Introduction

It is now well known that *in vitro* anaerobic mineralisation of phenol to CO_2 and CH_4 occurs with inocula from sewage sludge (Healy and Young, 1978; Knoll and Winter, 1987), anoxic aquifer-derived microcosms (Godsy, Goerlitz and Grbić-Galić, 1992), anaerobic digesters (Wang, Gu and Chonghua, 1993) and soil (Scott, Wolf and Lavy, 1982). Healy and Young (1979) found that in most cases, of eleven aromatic compounds tested, more than 80% of the carbon was converted to gas, clearly indicating that ring cleavage occurred under strictly anaerobic conditions. Further evidence of phenol mineralisation was demonstrated by Knoll and Winter (1987) in experiments with ^{14}C-phenol. From 11 μmol of radio-labelled phenol 39μmol of methane, containing 50% of the radioactivity, and 28 μmol of carbon dioxide, containing 38% of the radioactivity, were produced. The residual acetate (4.3 μmol) contained 4% of the ^{14}C-label and 6.7% of the label was found in the sludge pellet. It, therefore, seems likely that the capability exists in the landfill microbial gene pool for the mineralisation of phenol to CO_2 and CH_4. However, the effect of phenol co-disposal on refuse fermentation balances is less certain and was, therefore, one of the major objectives of this study.

An assessment of the impacts of the co-disposal of 1000, 2000 and 4000 mg l^{-1}/L phenol and/or anaerobically digested sewage sludge on landfill catabolic processes, with leachate components and methane generation the analytical criteria, was made. For the first criterion it must be stressed that discrete VFA concentrations represent balances between genesis and trophy (Watson-Craik, 1990). Although discrete methane concentrations (v/v) have been used to monitor the effects of xenobiotic molecules on anaerobic microbial associations (Fedorak and Hrudey, 1984) this criterion is less valid than either the methane

generation rate or the total volume of methane evolved. Due to the difficulty of measuring extremely low generation rates in open cultures this parameter was not measured in this study.

3.2   Experimental Procedure

Homogenised wet refuse was packed into the columns to an approximate density of 650 kg m^{-3}. The refuse and anaerobically digested sewage sludge were loaded in layers in the columns as shown in Figures 2.1 and 2.2. Initially, phenol was used in the perfusion strategies in two different concentrations, 1000 and 2000 mg l^{-1}/L.

The first 12 columns (C1-C12) contained refuse only whilst the second twelve (A1-A12) were packed with refuse and anaerobically digested sewage sludge in a ratio of 4.5:1 (w/w). The final 12 (B1-B12) were packed to a co-disposal ratio of 9:1. All the experimental columns were perfused at a very slow dilution rate (0.02 h^{-1}) initially and left at ambient temperature for 30 days before the temperature was increased and maintained at 30°C (+/-1.0°C). Specific details of the microcosm perfusion and operating conditions are given in Table 3.1.

For the recycle columns (A2,6,10; B2,6,10; C2,6,10), the leachates were collected 2-3 times per week by inserting 25 ml syringes (a) at the column base and allowing them to fill, usually at a slight negative pressure (Figure 2.2). The leachates were then reintroduced to the top of the columns while maintaining anaerobiosis.

Each column operated in batch mode (A3,7,11; B3,7,11: C3,7,11) was perfused with its respective concentration of phenol for approximately 36 hours and was then, with the exception of the gas exhaust port, closed and left to incubate for the duration of the experiment.

Table 3.1   Perfusion Strategies and Operating Conditions for "High Load" Dual
Co-Disposal Columns

Column Number	Packing Material	Eluent (mg l^{-1}/L)	Mode of Operation
A1	Refuse/Sludge(4.5:1)	Distilled Water	Single Elution
A2	Refuse/Sludge(4.5:1)	Distilled Water	Leachate Recycle
A3	Refuse/Sludge(4.5:1)	Distilled Water	Batch
A4	Refuse/Sludge(4.5:1)	Distilled Water	Simulated Rain
A5	Refuse/Sludge(4.5:1)	1000 Phenol	Single Elution
A6	Refuse/Sludge(4.5:1)	1000 Phenol	Leachate Recycle
A7	Refuse/Sludge(4.5:1)	1000 Phenol	Batch
A8	Refuse/Sludge(4.5:1)	1000 Phenol	Simulated Rain
A9	Refuse/Sludge(4.5:1)	2000 Phenol	Single Elution
A10	Refuse/Sludge(4.5:1)	2000 Phenol	Leachate Recycle
A11	Refuse/Sludge(4.5:1)	2000 Phenol	Batch
A12	Refuse/Sludge(4.5:1)	2000 Phenol	Simulated Rain

Table 3.1 (Cont.)   Perfusion Strategies and Operating Conditions for "Low Load" Dual
Co-Disposal Columns

Column Number	Packing Material	Eluent (mg l^{-1}/L)	Mode of Operation
B1	Refuse/Sludge(9:1)	Distilled Water	Single Elution
B2	Refuse/Sludge(9:1)	Distilled Water	Leachate Recycle
B3	Refuse/Sludge(9:1)	Distilled Water	Batch
B4	Refuse/Sludge(9:1)	Distilled Water	Simulated Rain
B5	Refuse/Sludge(9:1)	1000 Phenol	Single Elution
B6	Refuse/Sludge(9:1)	1000 Phenol	Leachate Recycle
B7	Refuse/Sludge(9:1)	1000 Phenol	Batch
B8	Refuse/Sludge(9:1)	1000 Phenol	Simulated Rain
B9	Refuse/Sludge(9:1)	2000 Phenol	Single Elution
B10	Refuse/Sludge(9:1)	2000 Phenol	Leachate Recycle
B11	Refuse/Sludge(9:1)	2000 Phenol	Batch
B12	Refuse/Sludge(9:1)	2000 Phenol	Simulated Rain

Table 3.1 (Cont.) Perfusion Strategies and Operating Conditions for Phenol Co-Disposal Columns

Column Number	Packing Material	Eluent (mg l^{-1}/L)	Mode of Operation
C1	Refuse only	Distilled Water	Single Elution
C2	Refuse only	Distilled Water	Leachate Recycle
C3	Refuse only	Distilled Water	Batch
C4	Refuse only	Distilled Water	Simulated Rain
C5	Refuse only	1000 Phenol	Single Elution
C6	Refuse only	1000 Phenol	Leachate Recycle
C7	Refuse only	1000 Phenol	Batch
C8	Refuse only	1000 Phenol	Simulated Rain
C9	Refuse only	2000 Phenol	Single Elution
C10	Refuse only	2000 Phenol	Leachate Recycle
C11	Refuse only	2000 Phenol	Batch
C12	Refuse only	2000 Phenol	Simulated Rain

The intermittent (rain) operated microcosms were first perfused with the appropriate concentration of phenol for 36 hours. Subsequently, the columns were operated in the same mode as the batch reactors with the exception that twice weekly they were perfused with distilled water (30 ml h^{-1}) for three hours.

Six further microcosms (D1-6) were constructed which were identical to the previous columns although these columns were packed with 'aged' refuse (Table 3.2). The refuse was packed into the columns after which they were allowed to stabilise at 30"C for about one month. To determine the state of the refuse fermentation, headspace methane concentrations were regularly monitored and perfusion was initiated after all the columns had attained a headspace methane concentration > 20% (v/v).

Table 3.2 Perfusion Strategies and Operating Conditions for Phenol Co-Disposal with Mature Refuse Columns

Column Number	Packing Material	Eluent (mg l^{-1}/L)	Mode of Operation
D1	Mature Refuse	1000 Phenol	Single Elution
D2	Mature Refuse	1000 Phenol	Leachate Recycle
D3	Mature Refuse	1000 Phenol	Batch
D4	Mature Refuse	2000 Phenol	Single Elution
D5	Mature Refuse	2000 Phenol	Leachate Recycle
D6	Mature Refuse	2000 Phenol	Batch

3.2.1 Mineral Salts Supplementation

For all the single elution columns (A1,5,9; B1,5,9; C1,5,9) mineral salts were added at one third of the concentration detailed in Section 2.3. With the batch (A3,7,11; B3,7,11; C3,7,11) and recycle (A2,6,10; B2,6,10; C2,6,10) columns, the mineral salts were added in quadruple strength 20ml aliquots to minimise dilution phenomena. The first addition was made after 357 days of operation with the second supplementation after 369 days of operation. Finally, supplementations of one sixth and double concentrations were made to the single elution, and batch and recycle microcosms, respectively after 376 days of operation. Thus, in total, between days 357-376 the following amounts (g) of the individual mineral salts were provided to the single elution, batch and recycle columns:

	Single Elution	Batch and Recycle
K_2HPO_4	22	6
NaH_2PO_4	10.2	3.15
NH_4Cl	13	3.5
$MgCl.6H_2O$	2.9	0.75
$NaHCO_3$	3.5	2
Na_2CO_3	2.9	0.75

3.3 Results and Discussion

3.3.1 Microcosm Operation

The choice of the refuse (Section 2.1) used in this study was motivated by the results of Watson-Craik and Senior (1989a) who determined that when phenol (188 mg l^{-1}/L) was co-disposed with municipal refuse the buffering capacity afforded by the latter affected the sensitivities of the microorganisms present to the bacteriostatic action of the phenol. It was found that one month old ("fresh") refuse was characterised by a greater buffering capacity than the "older" (four months old) refuse and phenol catabolism was higher in the former. The moisture content (wet weight) of the refuse as received was 62 $\pm 3\%$ (w/w). Moisture contents of 69.2 % (w/w) had been determined with previous refuse samples taken from the same site (Havinga, 1993).

The phenol concentrations used in these experiments were selected after consultation with a leading S.A. waste disposal contractor (Waste-tech (Pty) Ltd). Further, the relatively high phenol concentrations were specifically selected as they are commonly disposed of in landfill sites in South Africa after chemical treatment of phenolic wastes (Havinga, 1993).

The combination of an initial slow perfusion regime and ambient temperature was maintained as these "start up" procedures were found by Watson-Craik and Senior (1990) to enhance microbial activity, particularly methanogenesis, within the refuse columns.

Due to the number of variables examined in this study, 42 microcosms had to be used. To consider each of the variables the results and discussion are structured as follows: a. Firstly, phenol co-disposal with refuse is considered in relation to the microcosm mode of operation (single elution, leachate recycle, batch and simulated rain); b. Subsequently, the effects of the perfusion strategy (water, 1000 and 2000 mg l^{-1}/L phenol); and c. Finally, refuse age are considered.

3.3.2   Refuse and Phenol Co-Disposal

Single Elution Columns

Phenols naturally occur in soils, including intermediate and covering soils of landfill due to, for example, enterobacterial degradation of proteins and amino acids, particularly tyrosine. Phenol concentrations are also often increased in the groundwater in oil-bearing areas (Watson-Craik, 1987). The majority of phenolic compounds in soils are, however, undoubtedly derived from the degradation of lignins, mainly through the intervention of filamentous fungi, particularly the Basidiomycetes which are responsible for the white-rot type of wood decay (Senior and Balba, 1990). However, lignin-derived carbon is recalcitrant in anaerobic environments (Colberg, 1988) such as water-logged refuse columns. This, therefore, provides an explanation for why the concentrations of phenol recorded in the control (Column C1) were at all times very low (< 5 mg l^{-1}) (Figure 3.1 A).

Initially, the residual phenol concentrations detected in Column C5 (Figure 3.1A), which was perfused with 1000 mg l^{-1} phenol, were <200 mg l^{-1}, possibly due to adsorption on the refuse surfaces (Chapter 6). After day 60, however, a general increase was noted to a maximum of 1000 mg l^{-1}. Subsequently, the concentration decreased and stabilised (day 74).

This decrease in residual phenol could have resulted from adaption of the microbial community to the xenobiotic. Little is known about the molecular events that lead to adaptions of microbial communities although they have been shown to depend on the ecosystem, the type and concentration of the xenobiotic and the presence of other compounds (Wiggins and Alexander, 1988; van der Meer *et al.*, 1992; Wang *et al.*, 1993). The mechanisms of adaption to a new substrate may be briefly described as follows:

a.   Selection and enrichment of the organism/association that is able to metabolise the new compound;

Figure 3.1 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of the Phenol Co-Disposal Columns C1 (Control), C5 (1000ppm) and C9 (2000ppm) Operated with Single Elution During an Incubation Period of 695 Days.
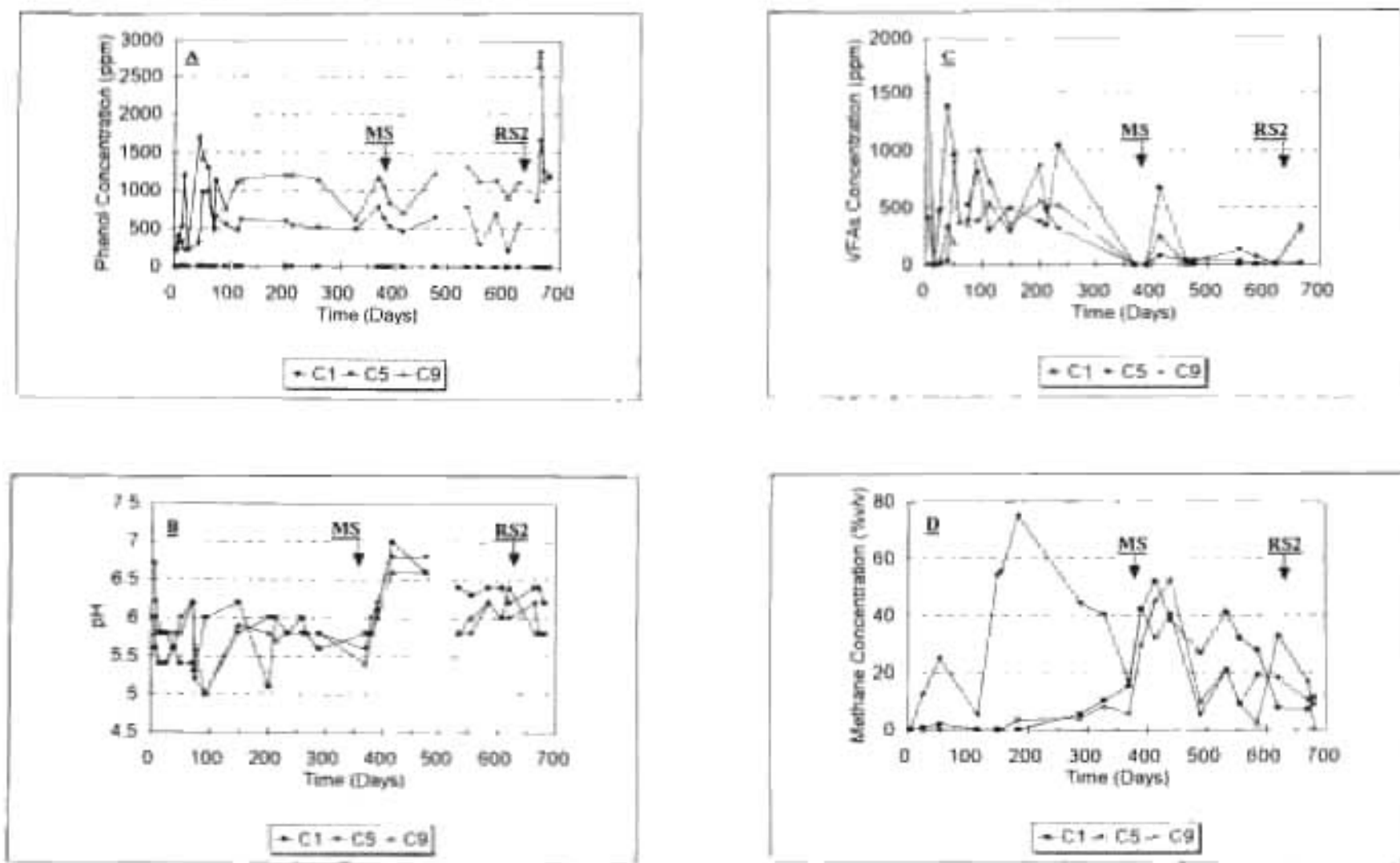
b. Induction of specific enzymes in members of the community;

c. Genetic alteration which includes: gene transfer, mutational drift and genetic recombination and transposition; and

d. Adaption to toxins, inhibitors or predators (Spain and van Veld, 1983; Young and Rivera, 1985; van der Meer *et al.*, 1992).

During the experimental period of 500 days 59.3% of the influent phenol was removed by percolation through Column C5. This was equivalent to an average removal rate of 0.27 g kg^{-1} d^{-1}. This, in turn, was equivalent to 270 g m^{-2} d^{-1} for refuse compacted to 1000 kg m^{-3}. On average, only 39.6% of the influent phenol was removed during the first 74 days which equates to a removal rate of 0.19 g kg^{-1} d^{-1}. This removal rate did, however, compare favourably with other recorded rates. For example, Watson-Craik (1987) demonstrated a removal efficiency of 31.5% for 188 mg l^{-1} phenol over a period of 530 days in single elution columns packed with "fresh" refuse (density approximately 525.6 kg m^{-3}), with a maximum phenol removal rate of 41 g m^{-3} d^{-1}. Following the phenol resupplementation (2000 mg l^{-1}) the residual phenol concentrations sharply increased with subsequent rapid decreases which were coincident with an increased average phenol removal rate of 0.59 g kg^{-1} d^{-1}.

Not surprisingly, the residual phenol concentrations of Column C9 (Figure 3.1 A) were higher than those of Column C5. With one exception, decreased concentrations were again detected initially. These low concentrations during the first 50 days can be attributed to physico-chemical phenomena such as absorption/adsorption of phenol to the various refuse surfaces (see Chapter 6). On average, over the first 500 days, Column C9 effected 55.9% removal of the added phenol. This is equivalent to a removal rate of 0.61g kg^{-1} d^{-1}. As with Column C5 sharp residual phenol concentration increases were recorded following the phenol resupplementation (RS2) although, subsequently, the concentration fell from 2642 mg l^{-1} on day 659 to 1209 mg l^{-1} on day 680 and averaged a phenol removal rate of 0.66g kg^{-1} d^{-1}. The mean phenol removal rates recorded for the various columns during the course of the study are illustrated in Table 3.3. These, apparent, enhanced phenol removal rates, following the phenol resupplementation, could have been due to either the adaption of the microbial community to the added xenobiotic or increased influent phenol

concentrations. For example, Watson-Craik (1987) showed that phenol removal increased as the influent concentration increased up to $^-$1500 mg l^{-1} with subsequent removal independent of the influent concentration. This suggested that first order kinetics applied ₂1500 mg l^{-1}, after which zero-order kinetics applied.

Comparisons of this nature must, however, be treated with care as experiments made by Rees and King (1981), in which a synthetic leachate supplemented with phenol was applied to Lower Greensand ($^-$80% sand and $^-$20% clay), indicated that, in general, the removal rates increased in response to higher flow rates. Watson-Craik, (1987); Watson-Craik and Senior, (1989b) also found that phenol removal was enhanced by increasing the irrigation rate. Thus, both the organic loading and hydraulic loading are key variables. The pH values of the single elution, water perfused Column C1 (Figure 3.1B) decreased initially from 6 to 5.4 and, with one exception, remained ₂pH 6 until day 390. Similar decreases have been noted by numerous researchers, including Barlaz, Schaefer and Ham, 1989b, and are accepted as indicative of fermentative microbial activity and the resulting accumulation of volatile fatty acids (Senior, Watson-Craik and Kasali, 1990) in the presence of low acid consuming activity of the methanogenic bacteria (Barlaz, Schaefer and Ham, 1989b), and the low alkalinity of fresh refuse (Barlaz, Ham and Schaefer, 1990).

Table 3.3   Mean Phenol Removal Rates for the Single Elution, Leachate Recycle and Batch Operated Columns During an Incubation Period of 695 Days

Column No.	Mode of Operation	Mean Phenol Removal Rate Prior to Phenol Resupplementation ($g\ kg^{-1}\ d^{-1}$)	Mean Phenol Removal Rate Following Phenol Resupplementation ($g\ kg^{-1}\ d^{-1}$)
C5	Single Elution	0.27	0.59
C6	Leachate Recycle	0.005	0.06
C7	Batch	0.0012	0.05
C9	Single Elution	0.61	0.66
C10	Leachate Recycle	0.0023	0.043
C11	Batch	0.0029	0.039

Following the mineral salts addition, however, the pH values increased although these elevated pH values were not maintained and, subsequently, decreased to values around a pH of 6.4.

For the two experimental columns (C5 and C9) similar initial decreases in pH were recorded. Both columns displayed higher pH values than the control column for the first 74 days. Throughout the study comparable gross pH values were recorded for the two experimental columns and brief pH increases were recorded following the mineral salts addition.

The higher pH values recorded in response to phenol elution, compared with the water control, were unexpected since although phenols are generally several orders of magnitude less acidic than carboxylic acids they are far more acidic than alcohols (Watson-Craik, 1987). Lallai and Mura (1989) showed that in experiments with two microbial associations, cultured aerobically in a batch reactor with phenol (60 - 1000 mg l^{-1}) as the limiting carbon source, the pH dropped until it reached a minimum in relation to the exhaustion of the phenol. This decrease in pH was ascribed to the production of organic acids from the intermediates of phenol catabolism. Watson-Craik (1987) examined phenol (188 mg l^{-1}) catabolism in a perfused multi-stage refuse column array and detected no residual phenol in the effluent during the first 45 days. Subsequently, the influent concentration was increased to 376 mg l^{-1} and this resulted in elevated residual phenol concentrations and reduced pH values from 6.55 to 5.7. Elevated VFA concentrations were not, however, detected in the leachate. There was, thus, some evidence of temporary shock loading with the 188 mg l^{-1} incremental increase in the influent phenol concentration (Watson-Craik and Senior, 1989c). In the present study both Columns C5 and C9 exhibited slight decreases in pH following the phenol resupplementation. Studies made by Sulisti, Watson-Craik and Senior (1996a) demonstrated the importance of pH on the catabolism of the substituted phenol, o-cresol, and reported maximum rates of o-cresol degradation in cultures poised at an initial pH of between 7 and 8. For example, > 72.6% of the added o-cresol was removed by day 70 in these cultures, whereas at pH 6.5, concentrations of o-cresol were reduced by only 37.5% (Sulisti et al., 1996a).

'Total' VFAs concentration was used as an analytical criterion for fermentation balance change and no distinction was made between the straight and branched-chain acids such as *iso*-valeric and *iso*-butyrate since these acids form as a result of deamination of amino acids and, as refuse is inherently low in protein, it was expected that these acids only constituted a minor component of the 'total' VFA's. It must be stressed that the 'total' VFAs concentrations recorded were balances between genesis and trophy and were not indicative of acid turnover *per se*.

In general, higher concentrations of 'total' VFAs (average 594 mg l^{-1}) were recorded in the leachate of Column C1 than Column C5 (average 348 mg l^{-1}) during the first 234 days (Figure 3.1C). Thus, it seemed likely that the lower pH values of Column C1 effluent during the same period were due to the presence of these acids. Subsequently, the reduced VFAs concentrations of both columns indicated either lower acid production, possibly due to labile carbon source limitation, or higher acid utilisation, due to the development of active catabolic groups. An increase in the 'total' VFAs concentrations, and a decline in the pH values, following the phenol resupplementation of Column C5, were possibly indicative of a phenol shock loading.

The co-disposal of 2000 mg l^{-1} phenol by single elution (Column C9) did not appear to effect increased 'total' VFAs generation in comparison with the water control since an average concentration of 515 mg l^{-1} was recorded during the first 234 days of operation. As with Column C5, after the phenol resupplementation, increases in the 'total' VFA concentrations were recorded.

As can be seen in Figure 3.1D, very low methane concentrations were recorded for the water-eluted column (C1) until day 280 after which a steady increase was noted to 15.2 % (v/v) by day 369. Subsequently, a major increase to 52 % (v/v) was recorded which coincided with the addition of the mineral salts. The lower methane evolutions in C1, prior to day 369 were, therefore, unlikely to have been a result of substrate limitation. A more likely reason was elemental limitations. The higher concentrations were not, however, maintained and a progressive decline in the methane concentration was recorded.

The phenol eluted column C5 showed rapid methane concentration increases to reach 74 % (v/v) on day 183. These increases coincided with decreasing residual phenol concentrations. The rapid increases in the presence of 1000 mg l^{-1} phenol (C5) were unexpected as Watson-Craik (1987) and Watson-Craik and Senior (1989a) showed that the addition of 188 mg l^{-1} phenol to refuse hindered the onset of methanogenesis. In contrast, the stimulation of methanogenesis by relatively low phenol concentrations has been demonstrated by other researchers. Fedorak and Hrudey (1984), for example, reported that addition of phenol (500 mg l^{-1}) significantly enhanced methane production while higher concentrations of 800, 1000 and 1200 mg l^{-1} did not effect productions which were significantly different (P < 0.05) from that of the control. In the presence of further concentration increases to 2000 and 3000 mg l^{-1}, the concentrations of methane produced decreased. It is doubtful that these results could be applied directly to South African conditions, refuse characteristics and landfilling practices since refuse composition can vary dramatically from country to country (Senior, 1990).

Following the above concentration increases, the methane concentrations declined to 17 % (v/v) on day 369 before again increasing following the addition of mineral salts. These increases in methane concentrations were, however, not sustained and the concentrations again rapidly decreased to 8.7 % (v/v) following the phenol resupplementation. It is interesting to note that, at the termination of the study, the control column (C1) exhibited a similar decreased methane concentration of 11.2 % (v/v)

A progressive decrease, from an attained maximum, in methane concentration recorded for Column C5 has also been observed by other researchers in similar experiments. For example, Watson-Craik and Senior (1989a) demonstrated that progressive decreases in methane release rates were apparent in single elution refuse columns. They speculated that nutrient/element limitation was the most likely cause although phosphate limitation was thought unlikely as a leachate PO_4-P concentration of 0.15 mg l^{-1} was coincident with low methanogenesis. Thus, exhaustion of the pool of labile carbon substrates was considered more likely. Although the methane generation rates and methane concentrations are not directly comparable the trend of a decrease in methane

concentration, from an attained maximum, was a recurring observation in this study (see Chapter 4 and 5) and it is, therefore, considered relevant to compare methane concentrations with methane release rates.

Column C9 (2000 phenol) followed a similar pattern to C1. Notwithstanding the low headspace methane concentrations recorded in Column C9 before day 369 significant phenol removal was evident (Figure 3.1A). This phenol removal together with the low methane concentrations was unexpected as Fedorak and Hrudey (1984) demonstrated that although phenol degradation was inhibited when the concentration exceeded ˜500 mg l^{-1}, acetate and propionate fermentation continued until the phenol concentration exceeded 2000 mg l^{-1}.

In this present study mineral salts additions to all three columns effected marked increases in methane concentrations. These could, however, have been due, in part, to the bicarbonate/carbonate additions as 3.5g of $NaHCO_3$ and 2.9 g of Na_2CO_3 were added, which should have increased the pH values as was observed (Figure 3.1B) and provided precursors for methanogenesis (Buvet, 1980):

$$4H_2 + HCO_3 + H^+ \rightarrow CH_4 + 3H_2O \qquad \Delta G^o = -135 \text{ kJ mol}^{-1} \qquad 3.1$$

Further, phenol as a disinfectant is more effective at pH values below its pK_a (pH10), that is the pH at which it is 50% dissociated (Watson-Craik, 1987) Therefore, the bacteriostatic/ bactericidal effects of phenol are greater at lower pH values and could thus inhibit methanogenesis.

Fedorak and Hrudey (1986) examined the role of five nutrients on anaerobic phenol mineralisation and showed that, initially, phenol was catabolized to methane and carbon dioxide. After various time intervals (Table 3.4) methanogenesis began to decline, and the residual phenol and p-cresol concentrations built up, due to the limitations of bicarbonate, major minerals and phosphate.

Table 3.4   Nutrients Omitted from the Reactors and the Time Required for Inhibition to Become Apparent (Fedorak and Hrudey, 1986)

Nutrient Omitted	Inhibition Started
Bicarbonate	¯25d (1.5 HRT)
Major Minerals	45d (¯3 HRT)
Phosphate	¯116d (7 HRT)

The authors concluded that bicarbonate was needed for phenol degradation rather than methane formation. The use of pH control in their experiments suggested, however, that the role of bicarbonate was not a pH effect. In a study made by Watson-Craik and Senior (1989a), addition of nitrogen (NH_4Cl, 0.9 g l^{-1}) and phosphorus (K_2HPO_4, 1.5 g l^{-1}) did not reverse the progressive decline in methanogenesis. The mineral salts addition, in the present study, did effect a temporary increase in phenol catabolism and coincided with the increased methane release.

Leachate Recycle Columns

In studies by Watson-Craik (1987) and Sulisti, Watson-Craik and Senior (1996b) leachates from co-disposal columns were collected in reservoirs before reintroduction into the columns on a continuous basis. However, to ensure constant flows, reservoirs (10 *l*) were used. These, therefore, constituted second continuous cultures in each experimental system and, thus, facilitated microbial catabolism external to the microcosms. According to Watson-Craik (1987), however, it was apparent that most of the biotransformation of phenol occurred in the microcosms rather than the reservoirs since 120 days were required by the reservoirs for the dissimilation of 2mM (188 mg l^{-1}) phenol.

As expected, the leachate recycle control column C2, at all times generated low (< 5 mg l^{-1}) phenol concentrations with a maximum concentration of 13 mg l^{-1} recorded on day 74 (Figure 3.2 A).

The recycle column subjected to 1000 mg l^{-1} phenol (C6) was characterised by variable, but initially low, phenol concentrations which reached 485 mg l^{-1} on day 45 before decreasing to below detection limits on day 414 (Figure 3.2A). The initially relatively low residual phenol concentrations were probably due to adsorption phenomena whereas the continued low concentrations could possibly be attributed to either the enrichment of a phenol catabolic population or the acclimation of key landfill microorganisms to the added compound. The first resupplementation (RS1), with a phenol concentration equal to the initial influent concentration, was made when the leachate recycle and batch columns recorded residual phenol concentrations approaching zero. Likewise, the second resupplementation (RS2), with double the initial phenol concentration, was made when the columns approached zero leachate residual phenol concentrations. Following both phenol resupplementations rapid breakthroughs of the phenol together with, subsequent, rapid concentration declines were recorded. Possible reasons for these rapid declines could have been the added nutrients which could have negated any elemental limitations and/or microbial adaption. The initial perfusion regime was described in Section 3.2 from which it can be calculated that a total of 0.7g phenol would have been added to Column C6. Therefore, a mean removal rate, prior to the first phenol resupplementation (RS1), of this column was $0.005g \ kg^{-1} \ d^{-1}$ while the rates following the resupplementations (RS1 and RS2) increased to $0.06g \ kg^{-1} \ d^{-1}$.

The residual phenol concentrations of Column C10 were, initially (<25days), relatively low, also possibly as a result of adsorption but subsequently increased to a maximum of 1004 mg l^{-1} on day 45. The relatively protracted completion of phenol catabolism by Column C10, compared to Column C6, was expected as increased concentrations of phenol affect microbial activity negatively. Blakey and Knox (1978), for example, indicated a significant reduction in the rate of phenol catabolism, under anaerobic conditions, when the phenol concentrations were >500 mg l^{-1}. Despite constant conditions operating in the microcosm (C10) the residual phenol concentrations did decline after day 250. Notwithstanding the prolonged period required to remove the initial added phenol at a mean removal rate of $0.0023g \ kg^{-1} \ d^{-1}$ this rate increased to $0.043g \ kg^{-1} \ d^{-1}$ following each resupplementation (RS1 and RS2).

Figure 3.2 Changes in Residual Phenol Concentrations (A), pH (B), Total Volatile Fatty Acids Concentration (C) and Methane Concentrations (D) of Phenol Co-Disposal Columns C2 (Control), C6 (1000ppm) and C10 (2000ppm) Operated with Leachate Recycle During an Incubation Period of 695 Days
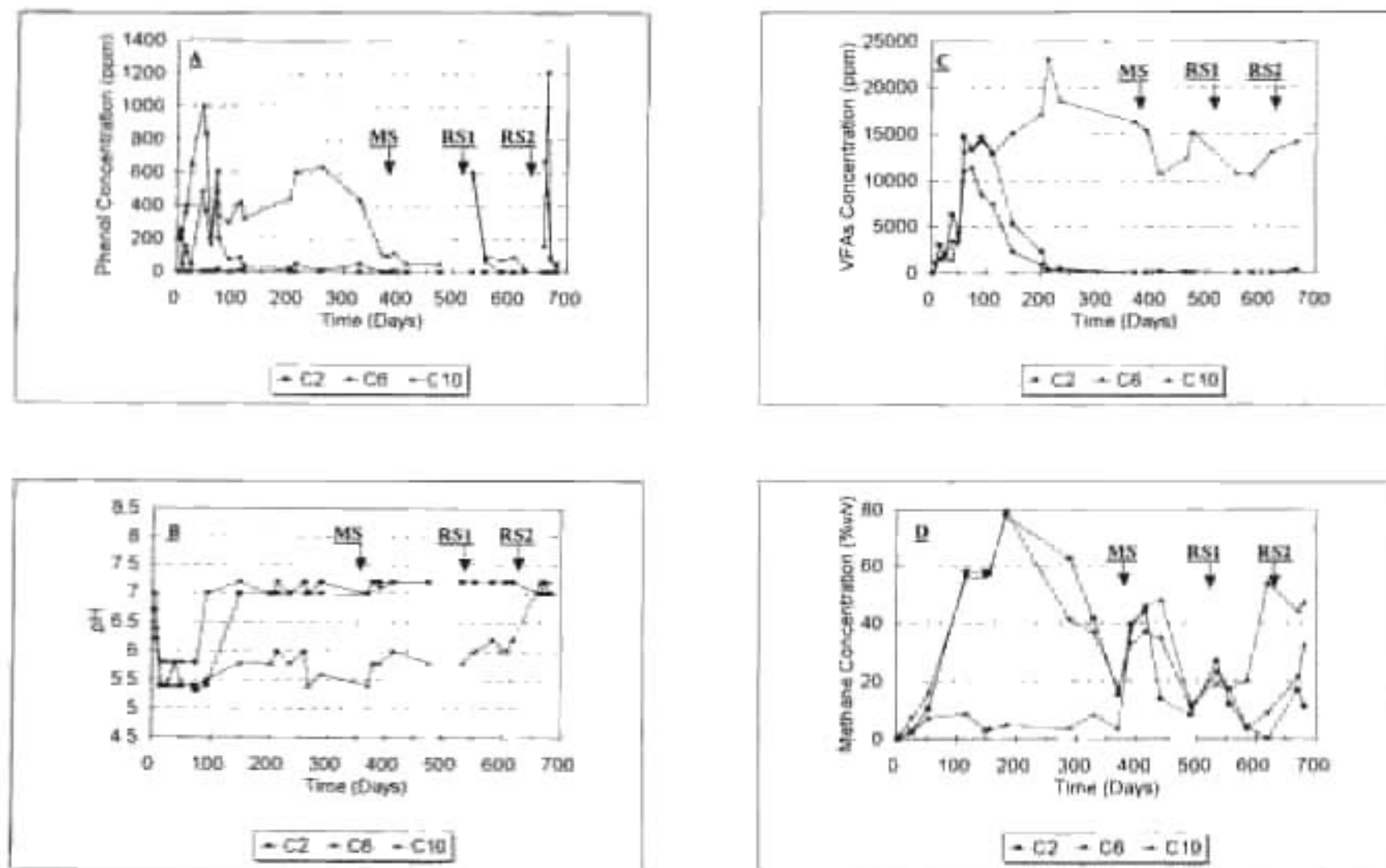
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (1000ppm)

RS2 Designates Phenol Resupplementation (2000ppm)

Unlike Column C2, which attained a neutral pH on day 148, Column C6, particularly in the early stages, was characterised by higher pH values (Figure 3.2B) and attained a neutral pH on day 92, after which it stabilised around this value. Column C10 on the other hand attained a neutral pH after 659 days. It is possible that the relatively protracted onset of phenol degradation could also have resulted from the low pH values. For example, Tibbles and Baecker (1989b) reported that the optimal pH growth range for four mesophilic phenol catabolising bacteria, which were isolated from refuse, was between 6.8 and 7.6, with growth and phenol catabolism inhibition evident below pH 6. Despite the high initial phenol loading and the resulting low pH regime which established in Column C10, the residual phenol concentration did decline after day 250. This could have been due to the enrichment of a phenol degrading population or the acclimation of key landfill microorganisms to the xenobiotic. It is also worth noting that the pH values for the co-disposal columns did not markedly decrease following the phenol resupplementations.

From Figure 3.2 C it can be seen that for Column C6 increasing pH values were accompanied by low concentrations of VFAs. Subsequently, Column C2 followed a similar trend. A comparable inhibitory effect on VFAs generation was noted by Watson-Craik (1987) following refuse column perfusion with either 750 or 1128 mg l^{-1} phenol. For Columns C2 and C6 there were no notable accumulations of 'total' VFAs following the resupplementations (RS1 and RS2).

Notwithstanding the similar trends in pH and 'total' VFAs concentrations between Columns C2 and C10, until day 92, Column C10 was never characterised by a decrease in the VFAs concentration, which could explain the consistently lower pH values recorded.

The toxicity of VFAs at low pH values has usually been explained by their action as "uncouplers". According to Russell, J.B. (1992), however, the analogy between fermentation acids and uncouplers is flawed. Uncoupler anions cycle through the cell membrane at rapid rates but there is little evidence that fermentation acid anions can operate in a similar fashion. Further, according to Russell, J.B. (1992), it has been suggested that anion accumulation is responsible for the toxic effects of VFAs at low pH.

When undissociated acids pass into the cell and dissociate in the more alkaline interior, some protons are released although the effect of this influx has often been overemphasised. If it is assumed that there is a 1M increase in the intracellular acetate concentration in a bacterium, the H^+/ATP stoichiometry of the membrane bound ATPase is 3 and the bacterium has an intracellular volume of 3 μl mg^{-1} protein. Based on these assumptions, 1μmol ATP mg^{-1} protein would be required to expel the protons of the acetate. Even anaerobic bacteria with moderate growth rates (0.5 h^{-1}) can produce more than 50μmol ATP mg^{-1} protein h^{-1} (Russell, J.B., 1992).

From a maximum of 78% (v/v) on day 183 the headspace methane concentrations of Column C2 declined until day 369. Subsequent increases and then decreases were recorded as a result of the mineral salts addition. The increase following the mineral salts supplementation could have resulted from the addition of carbonate which is one of the methanogenic precursors. It is interesting to note that a methane concentration of 58% (v/v) was recorded for this column while the pH was 5.3. This could have been due to the heterogeneous nature of refuse effecting a gross pH value of the leachate which was different from the pH values in the microniches of the refuse. Rees (1980), for example, speculated that an active and well established methanogenic population could tolerate and function at much lower pH values in the landfill ecosystem than it could in either liquid media or in full-scale anaerobic sewage sludge digesters.

The methane concentrations of Column C6 followed a similar trend although the decline from a maximum value of 79% (v/v) was more rapid. The increase in methanogenic activity in response to mineral salts supplementation was also evident in Column C6 despite the presence of phenol (100 to 483 mg l^{-1}) in the leachate during the same time period. The interrelated reductions of the exogenous electron acceptors, nitrate and sulphate, were apparently not inhibited in this column as low (<0.12 mg l^{-1}) concentrations were recorded prior to the mineral salts addition.

From the above results it can be seen that, for the leachate recycle operated column C6, the addition of 1000 mg l^{-1} phenol did not seem to negatively affect the onset of anaerobiosis or the terminal process of methanogenesis.

In contrast, with Column C10 (Figure 3.2D) it appeared that the process of acidogenesis was not severely affected by the added phenol but the subsequent and interrelated processes of VFA catabolism and methanogenesis were significantly affected. These results agreed with the findings of Watson-Craik and Senior (1989c) who observed that acetic acid accumulations coincided with depressed methanogenesis.

The relatively low methane concentrations evolved by Column C10 did not result from substrate limitation since the column was characterised by high 'total' VFAs accumulations and low gross pH values. However, it is doubtful if the low methane generation resulted from low pH values *per se* as Column C2 was characterised by a methane concentration of 58% (v/v) despite a gross leachate pH of 5.4. Although it is now generally recognised that high concentrations of VFAs and low pH regimes can inhibit methanogenesis, the mechanism of this is not fully resolved (Kasali, Senior and Watson-Craik, 1990a; Senior, 1990). Sturz, Topel, Ali, Merrett and Robinson (1991), for example, reported that relatively high VFA concentrations of 23 000 mg l^{-1} were not inhibitory and, at pH values above 4.9, stimulated methanogenesis.

Several authors have indicated that landfills may be either nitrogen (Pacey, 1989b; Senior and Balba, 1987) or phosphorus (Senior, 1991) limited, which could severely inhibit methanogenesis and result in low methane evolution. However, this did not seem likely in this study as the leachates of Columns C2 and C6 both contained low (< 0.1 mg l^{-1}) phosphorus concentrations. Also, Watson-Craik (1987) did not record any mineral/nutrient limitations in comparable refuse column studies until $+/-$ 680 days of operation.

Another possible explanation for the low methane generation was competition with the nitrate and sulphate-reducing bacteria for common precursors (Senior and Balba, 1984). This possibility was supported by relatively high concentrations of 3.12 and 8.9 mg l^{-1} for the respective electron acceptors, nitrate and sulphate, prior to mineral salts addition.

In a study made to determine the effects of selected fermentation gases (H_2, CO_2, CH_4) on refuse acidogenesis and methanogenesis, Kasali, Senior and Watson-Craik (1990b)

observed that over pressures of these gases could affect fermentation balance changes. A culture headspace atmosphere of 100% CO_2 effected 98% inhibition of methanogenesis and partial suppression of acetogenesis while a gas mixture of CO_2-CH_4 (40:60 v/v) also exerted inhibitory effects on methanogenesis. These results seemed to indicate that the slow onset of methanogenesis resulted from either the slow development of a sufficiently low redox potential, approximately -400 mV (Oremland, 1988), dependent on the metabolic activities of, for example, sulphate-reducing bacteria (Senior and Balba, 1984), or gaseous overpressures. The latter would be the less likely as the experimental configuration facilitated gas ventilation although, at times, significant gas volume accumulations were recorded. It is also of interest to note that the nitrite concentrations also indicated an apparent inhibition of nitrite reduction since a concentration of 13.9 mg l^{-1} was recorded for Column C10 leachate in comparison with 0.11 and 0.22 mg l^{-1} for Columns C2 and C6, respectively.

It, therefore, appeared that the relatively low degradation rates of phenol may have resulted in the inhibition of the nitrate and sulphate reducers as well as the methanogens. Senior and Balba (1987), for example, reported that the anaerobic fermentation of aromatic compounds required sulphate-reducing or methanogenic activity to maintain low cultural concentrations of both acetate and hydrogen.

The methane concentrations of Column C10 clearly increased following the mineral salts addition although a beneficial effect on the pH was less evident as no significant change was apparent. The methane concentration increases, therefore, could have been due to the added carbonate promoting the hydrogenophilic methanogens. Also, the phenol resupplementations appeared to have enhancing effects on methane generation as Column C10 exhibited between 42 and 56% (v/v) methane following the second supplementation (RS2).

Batch Operated Columns

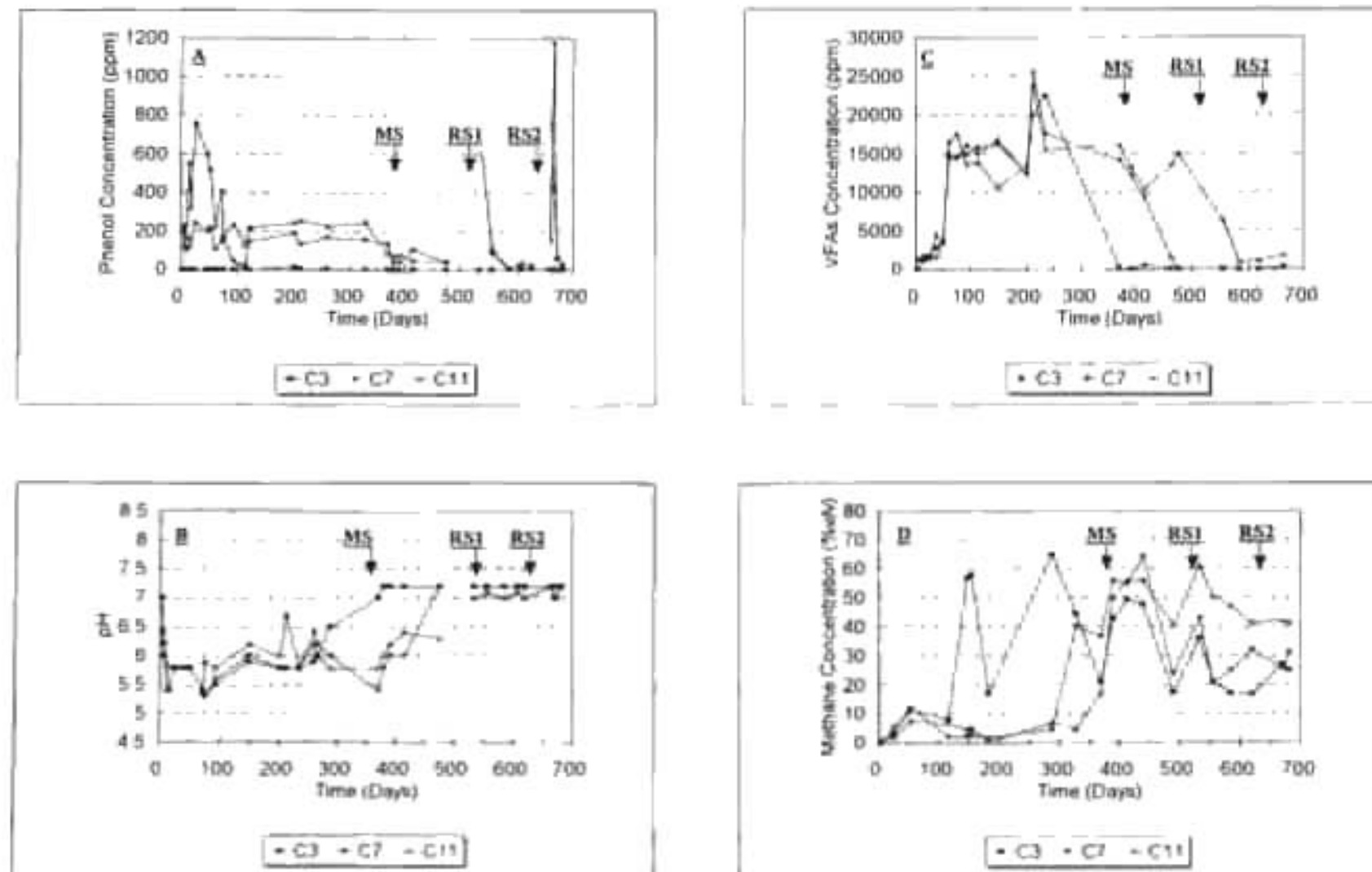The residual phenol concentrations of the batch reactor leachates are shown in

Figure 3.3 A. The control column (C3) at all times exhibited very low concentrations with a maximum of 14 mg l^{-1} detected on day 201. Despite the addition of 1000 mg l^{-1} phenol to Column C7 the highest concentration recorded in this column was 240 mg l^{-1} after 25 days. This apparent immobilisation of phenol, especially in the early stages, could have been as a result of physico-chemical phenomena such as adsorption to the refuse surfaces. This increased adsorption compared with the equivalent leachate recycle column (C6) could possibly have been due to the difference in operating conditions as the batch mode column facilitated extended contact between the solute and the adsorbent as a result of the lack of liquid movement. Despite the relatively low initial residual phenol concentrations, a protracted period of approximately 360 days was required to effect concentrations approaching zero and this was only attained after the mineral salts addition. Thus, attenuation could have necessitated a prolonged acclimation period for phenol degrading microflora and/or the removal of elemental limitations. Further, notwithstanding the initial protracted period for effective phenol catabolism, which was characterised by a mean removal rate of 0.00125g kg^{-1} d^{-1}, the phenol removal rate increased to 0.005g kg^{-1} d^{-1} following the first resupplementation (RS1).

The relatively low phenol concentrations recorded initially for Column C11 could also have resulted from adsorption although the residual phenol concentration rapidly increased to 761 mg l^{-1} after 25 days. Subsequently, the phenol concentrations decreased and plateaued around 200 mg l^{-1} until day 369 after which they declined following nutrient supplementation to an average phenol removal rate of 0.0029g kg^{-1} d^{-1}. After the first phenol resupplementation (RS1) the average phenol removal rate increased to 0.039g kg^{-1} d^{-1} which could have been due to the removal of elemental limitations, due to the mineral salts addition and/or microbial adaption. The increased removal rate exhibited by Column C11 compared to C7 was not unexpected as it has been shown that increased influent concentrations can effect increased phenol catabolism (Watson-Craik and Senior, 1990).

The apparently protracted period required for microbial acclimation was also not unexpected as Wang et al., (1993) reported that the acclimation period required for bacteria derived from digested sludge, from a municipal wastewater treatment plant, which were fed

Figure 3.3 Changes in Residual Phenol Concentrations (A), pH (B), Total Volatile Fatty Acids Concentration (C) and Methane Concentrations (D) of Phenol Co-Disposal Columns C3 (Control), C7 (1000ppm) and C11 (2000ppm) Operated in Batch Mode During an Incubation Period of 695 Days

MS Designates Mineral Salt Addition

RS1 Designates Phenol Reaustablisheration (1000ppm)

RS2 Designates Phenol Reaustablisheration (2000ppm)

a glucose-phenol (1000 mg l^{-1}) mixture was about 195 days. Further, it is known that increased phenol concentrations effect phenol degradation rates negatively. For example, Wang *et al.*, (1989) demonstrated in batch methanogenic experiments that increased acclimation periods and decreased phenol degradation resulted when the initial phenol concentration was increased above 600 mg l^{-1}. In this present study an acclimation period of approximately 360 days was required.

All the batch operated columns displayed an initial sharp decline in pH with subsequent increases (Figure 3.3 B). However, during the first 234 days Column C11 displayed higher pH values than both the control and the 1000 mg l^{-1} challenged column (C7) which, in turn, displayed higher pH values than the control (C3). The increased pH values recorded for Column C11 were unexpected as the column showed approaching equivalent VFA concentrations as the other columns over the same time period. After 360 days, however, Column C3 attained a neutral pH while the phenol supplemented columns required 475 (C7) and 533 (C11) days.

Following the period of decreased pH values for Column C3 the VFAs concentration decreased rapidly (Figure 3.3 C) with a concomitant increase in the pH value. A similar result was later recorded for Column C7 which could, in part, explain this column's slower recovery in pH values. Column C11 on the other hand displayed a slight decrease in the VFAs generation/release rate with a corresponding increase in the pH value following the mineral salts addition and a strong decrease following the first phenol resupplementation (RS1). Of importance, however, is the fact that both phenol resupplementation events did not effect any noticeable increases or accumulations of the 'total' VFAs.

The methane concentrations (Figure 3.3D) of Column C3 were variable although the, now, characteristic increase with subsequent decrease following mineral salts addition were again evident. During the first 288 days, similar methane concentration trends were recorded for Columns C7 and C11. Subsequently, a concentration of 40% (v/v) was recorded for Column C7 prior to the mineral salts addition, whereas Column C11 only

increased following the addition. The initial low methane concentrations were, therefore, not due to substrate limitation. Also, all the columns showed a general decrease in methane concentrations following the first phenol resupplementation (RS1) although it must be noted that the phenol co-disposal columns showed enhanced concentrations compared to the refuse only control with column C11, for example, exhibiting a concentration of 41% (v/v) at the termination of the study. The resupplementation (RS2) with increased phenol concentrations (≤ 4000 mg l^{-1}) did not appear to inhibit methanogenesis.

The positive effects of the mineral salts addition were also recorded for the electron acceptors nitrate and sulphate in Column C7. Prior to the addition the concentrations were 3.1 and 6.5 (mg l^{-1}), respectively. Subsequently, the concentrations were below detection limits. A further point of interest is that detectable concentrations of nitrite were recorded prior to the addition but not afterwards. Phosphate concentrations were at all times below detection limits. This seemed to indicate that the refuse could have been nutrient limited. Further circumstantial evidence of this deficiency was the relatively high concentrations of nitrate (8.41 mg l^{-1}) and sulphate (10.6 mg l^{-1}), compared to 0.37 and below detection for the control, immediately preceding the mineral salts addition. The concentrations of these electron acceptors rapidly decreased to below detection limits following the addition. For this column the phosphate concentrations were also at all times below detection limits.

The presence of the electron acceptors nitrate and sulphate as well as the VFAs prior to the mineral salts addition supported the view that phenol additions, especially 2000 mg l^{-1}, inhibited the bacteria that metabolically preceded the methanogens. A similar conclusion was also reached by Sulisti et al., (1996b) who supplemented methanogenically active batch cultures with a range of o-cresol concentrations (from 0 to 7mM) and reported that the addition of o-cresol even at high concentrations did not affect the production of methane compared to the control. These observations suggested that o-cresol did not inhibit methanogenesis directly but at concentrations ≥ 3.5mM it, possibly, affected microorganisms whose growth preceded those of the methanogens and induced redox conditions which were favourable for the methanogens. For example, Ferry and Wolfe (1976) showed that the methanogenic fermentation of benzoate required the cooperation of

several groups of bacteria and that the methanogens served only as the terminal organisms of the catabolism.

From these results it appeared that the refuse methanogenic fermentation was negatively affected by the addition of, especially 2000 mg l^{-1}, phenol prior to the addition of the mineral salts. Promotion of the fermentation could then have resulted from:

a.   The buffering effect of the added bicarbonate; and/or

b.   The addition of essential nutrients; and/or

c.   Although only small volume changes resulted, phenol dilution.

A possible buffering effect of the added bicarbonate was substantiated by the results of Figure 3.3 B since the pH recovered markedly following the addition. This, in turn, resulted in an increase in the methane concentration either due to the pH change or the provision of methane precursors. The beneficial effects of the addition were also evident in the decreases in the phenol concentrations of both Columns C7 and C11 although the effects on the VFAs of Column C11 were not sustained.

Simulated Rain Columns

The residual phenol concentrations of the control (C4) were at all times very low (< 4.8 mg l^{-1}) (Figure 3.4 A). In contrast, the 1000 mg l^{-1} co-disposed column (C8) exhibited very high concentrations of up to 840 mg l^{-1} after 7 days and these remained relatively constant at around 240 mg l^{-1} until day 45 after which a further reduction was recorded. It has been demonstrated that upon repeated wetting and drying of the adsorbate the adsorption of xenobiotics is significantly stronger and the subsequent desorption of the xenobiotics is reduced (C. A. du Plessis, personal communication). In this instance, however, it appeared that the operating regime resulted in rapid leaching and, therefore, desorption of the added phenol from the column resulted in very low phenol catabolism. Due to the initial perfusion regime and the subsequent addition of rain (180ml per week) it was calculated that approximately 81 % of the added phenol was leached from this column. This further appeared to indicate that the adsorption of phenol to refuse was very weak (See Chapter 6). For the 2000 mg l^{-1} co-disposed column (C12) a slightly longer time was required to attain

Figure 3.4 Changes in Residual Phenol Concentrations (A), pH (B), Total Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of Phenol Co-Disposal Columns C4 (Control), C8 (1000ppm) and C12 (2000ppm) Operated with Simulated Rain During an Incubation Period of 695 Days
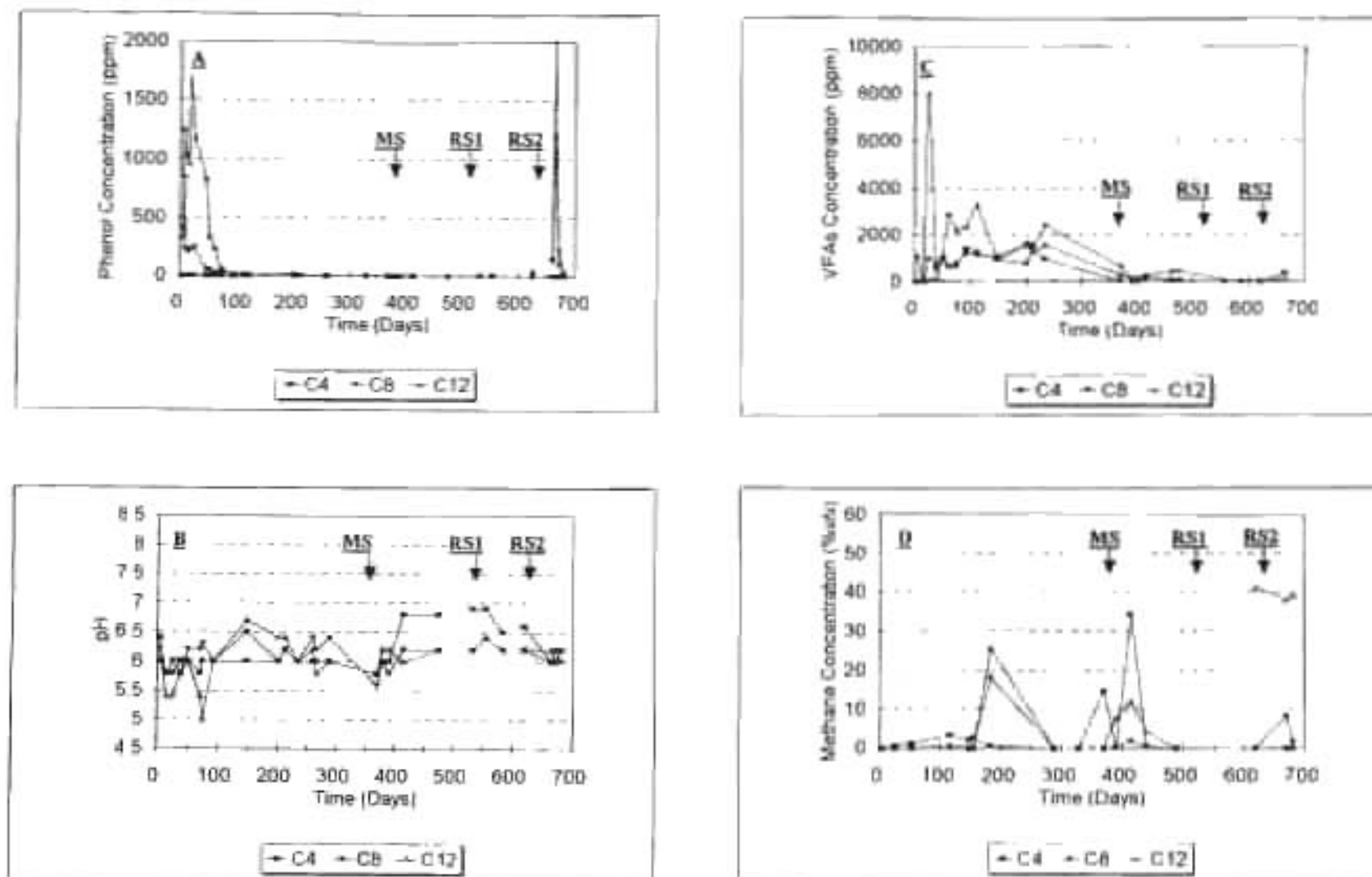
MS Designates Mineral Salts Additive

RS1 Designates Phenol Resupplementation (+1000ppm)

RS2 Designates Phenol Resupplementation (2000ppm)

very low phenol concentrations although significant leaching (79%) was also evident for this column. The phenomenon of weak adsorption was again recorded with the second phenol resupplementation and was evidenced by the rapid breakthrough and subsequent leaching.

The pH values (Figure 3.4 B) of all three columns were below 7 prior to the addition of mineral salts after which slight increases were recorded with the pH values again decreasing following the phenol resupplementations (RS1 and RS2).

The concentrations of 'total' fatty acids (Figure 3.4 C) were, irrespective of the influent, close to the same order over the entire length of the experiment although Column C12 was characterised by, initially, higher concentrations. The relatively low concentrations recorded for these columns could have been due to either low rates of fatty acid production or, more likely, as a result of the leaching out of these acids.

Although the methane concentrations recorded for these columns (Figure 3.4 D) were variable they were, in general, relatively low. The low 'total' VFAs concentrations could, therefore, not be accounted for by increased methanogenesis. Despite the addition of water, which has been shown to enhance methanogenesis, the intermittent wetting and drying appeared to inhibit the methanogenic process. The reasons for the enhanced methane concentrations recorded for Column C12 following the second phenol resupplementation were not clear as the fermentation indicators (pH and 'total' VFAs concentrations) remained relatively constant which would seem to indicate that microbial adaption to the phenol had occurred.

3.3.3 The Effects of the Perfusion Strategy on Phenol Co-Disposal and Refuse Degradation

Water Perfused Columns

As previously discussed, the control columns at all times displayed very low phenol

concentrations and, thus, did not appear to have been affected by the type of perfusion strategy employed. The residual phenol concentrations are shown in Figure 3.5 A.

All the columns were characterised by initial pH declines (< pH6) although, subsequently, the leachate recycle column (C2) and the batch reactor (Column C3) attained neutral pH values on days 148 and 369, respectively.

The pH values of the single elution (C1) and rain simulated (C4) reactors increased but only following the addition of mineral salts probably as a result of the added HCO_3 /CO_3^2. The lowered pH values of the refuse subjected to leaching have been attributed by Zinder, Anguish and Cardwell (1984) to the flushing out of buffer (HCO_3) thus decreasing the buffering capacity of the refuse. Supporting evidence for this was gained by the relatively low concentrations of VFAs in the columns which had also been subjected to leaching. The increase in the pH of Column C2 leachate was coincident with a decline in the 'total' VFAs concentrations. Column C3 also showed similar results but not until after the addition of mineral salts.

The significant effect of the perfusion strategy on methanogenesis is indicated in Figure 3.5 D. The low methane concentrations recorded for the single elution and rain simulated reactors could also have resulted from the low VFAs concentrations and/or low pH values. Further, the continuous leaching of the VFAs could have resulted in a slower establishment of a balanced refuse fermentation due to the removal of labile substrates. Finally, the physical effect of liquid channelling, through preferential pathways in the refuse, could have resulted in rapid flow regimes.

Unlike temperature and pH manipulation, increasing the moisture content and movement through a landfill is relatively simple and is usually effected by means of leachate recycle. Klink and Ham (1982) illustrated that moisture content *per se* and movement are separate variables affecting refuse degradation. The possible enhancing effects of leachate recycle on refuse catabolism and methanogenesis still remain to be resolved. For example, it has been reported that simple leachate recycling without some

Figure 3.5 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentration (D) of Refuse Columns Operated with Single Elution (C1), Leachate Recycle (C2), Batch Mode (C3) and Simulated Rain (C4) During an Incubation Period of 695 Days

MS Designates Mineral Salts Addition
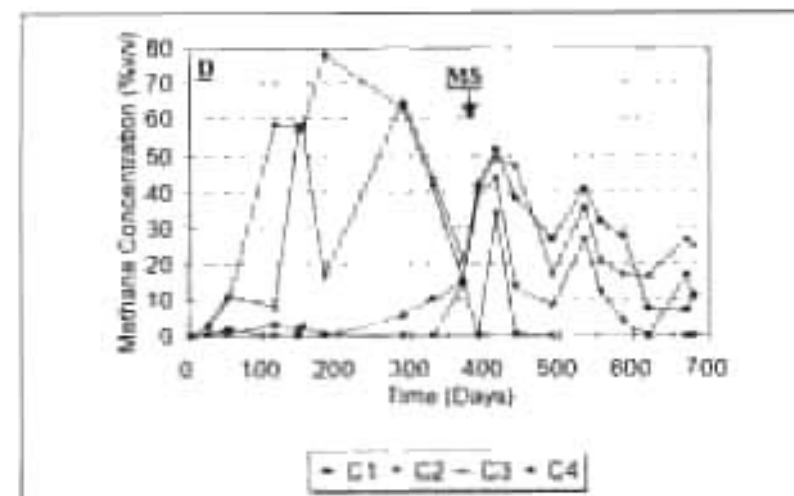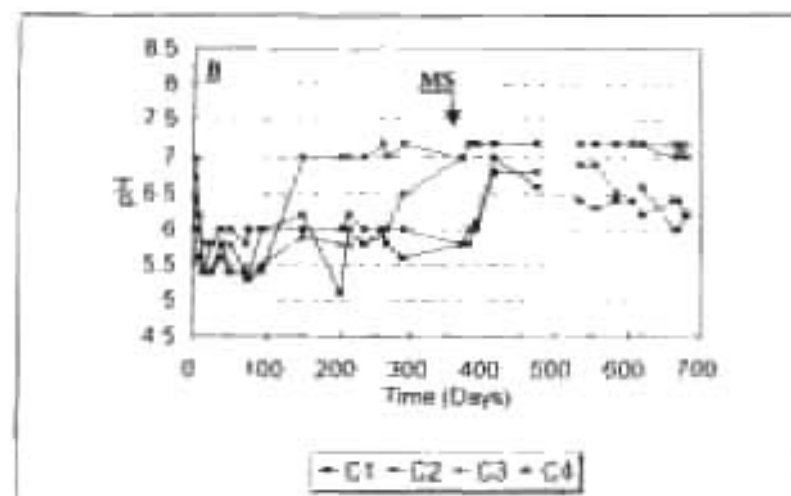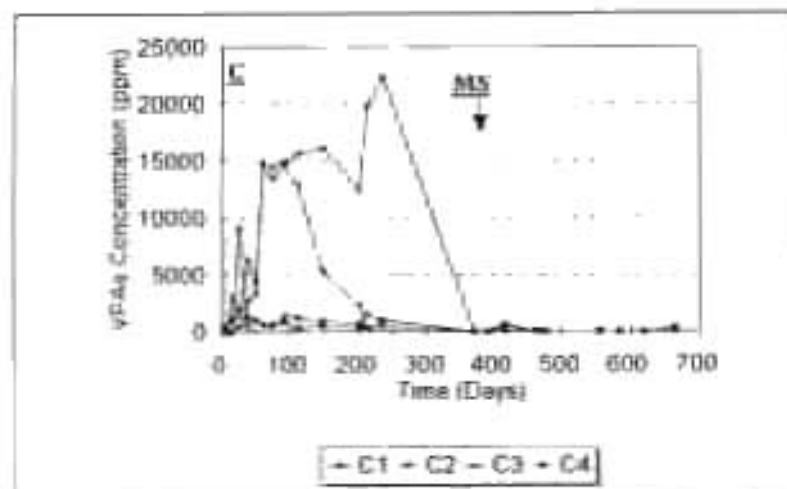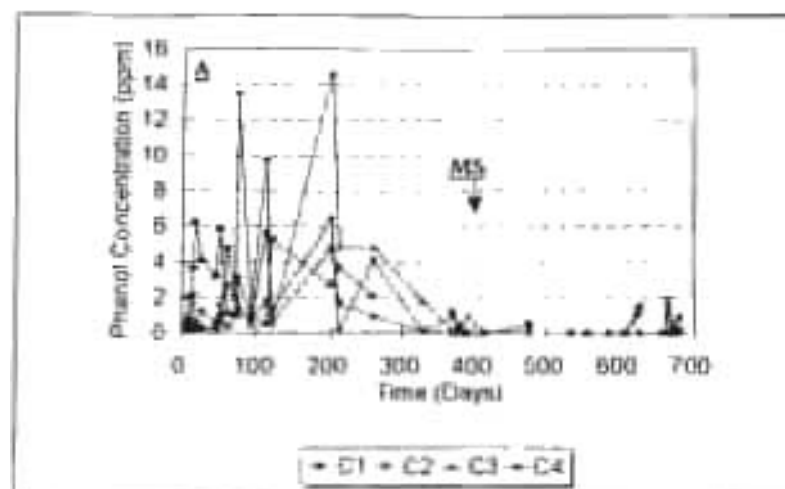
form of pH neutralization did not offer any advantages (Buivid *et al.*, 1981; Barlaz, Milke and Ham, 1987). Pohland (1989a;b), on the other hand, indicated that accelerated refuse stabilisation and conversion of labile substrates was evident in leachate recycle test cells compared to single elution cells. This enhancing effect was thought to be caused by one of the following:

a.   Maintenance of a desirable moisture content throughout more of the refuse sample;

b.   More uniform distribution of a suitable pH value;

c.   Recycling and distribution of nutrients and enzymes etc; and

d.   Dilution of inhibitory products (Buivid *et al.*, 1981; Westlake, 1990).

The temporary increases in the methane concentrations following mineral salts addition could have been due to the added carbonate which is one of the methanogenic precursors. These increases were, however, short lived and soon decreased.

1000 mg l⁻¹ Phenol Perfused Columns

The residual phenol, pH, 'total' VFAs and methane concentrations for Columns C5 to C8, recorded during this study are shown in Figures 3.6 A - 3.6 D. Initially, the single elution column (C5) showed relatively high residual phenol concentrations after which the values stabilised between 200 and 400 mg l^{-1}. Clearly, however, at all times the phenol concentrations of the single elution column were higher than the leachate recycle , batch and simulated rain columns. This was as expected although the phenol removal rate and, thus, the relative mass of phenol degraded, was higher in the single elution column than the other three columns (Table 3.3). In the leachate recycle column the residual phenol concentrations rapidly decreased to less than 200 mg l^{-1} (day 60) while 92 days were required by the batch columns to attain a similar residual phenol concentration. The leachate recycle column, thus, showed an enhanced phenol degrading capacity. Watson-Craik (1987); Watson-Craik and Senior (1989a) demonstrated similar enhanced methanogenic biodegradation of a model phenol-containing wastewater perfused through laboratory-scale refuse columns. This operating regime resulted in 100% attenuation of the influent phenol and increased methane release rates. Similar enhancing phenomena were recorded by

Figure 3.6 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of 1000ppm Phenol Co-Disposal Columns Operated with Single Elution (C5), Leachate Recycle (C6), Batch Mode (C7) and Simulated Rain if During an Incubation Period of 695 Days
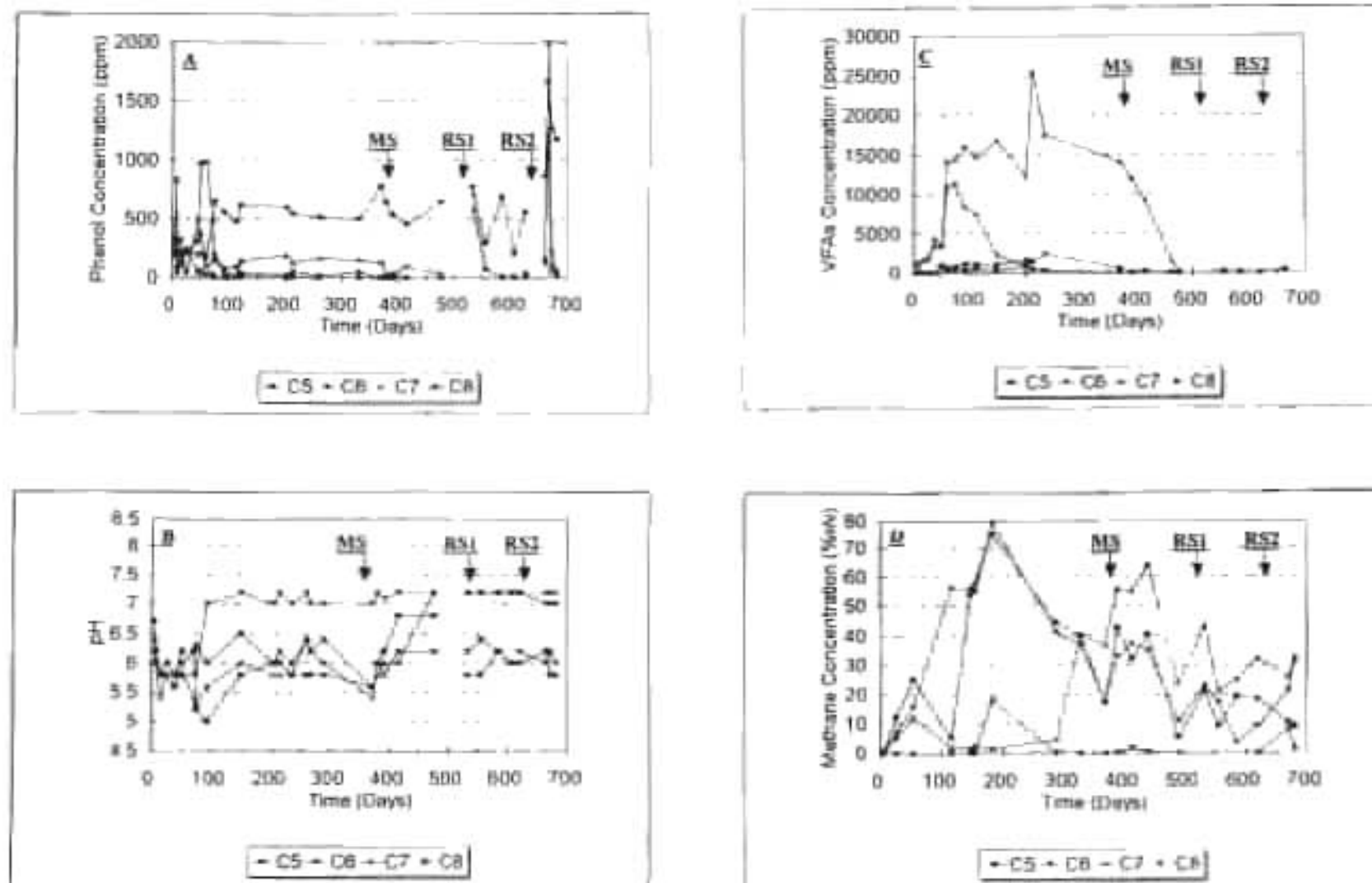
Sulisti (1996b) with the co-disposal of o-cresol and municipal refuse. These higher rates of methane release, as a result of leachate recycle, were attributed to:

a.   The reintroduction of sulphate ions and VFAs as electron acceptors and carbon sources, respectively;

b.   The role of leachate recycle in maintaining the buffering capacity, and maintaining leachate pH values suitable for the growth of methanogens; and

c.   The recycling of leachate containing high concentrations of organic acids which promoted the acid hydrolysis of refuse components and increased the pool size of substrates for acetogenesis, which was required for acetotrophic methanogenesis.

The simulated rain column showed rapid leaching and desorption of the phenol in the early stages with very low phenol removal accounted for by catabolism. Although no data have been presented on the effects of pH on the desorption of phenol from refuse, Arriola-Fortuny and Fuller (1982) reported accelerated desorption of phenol from soils under acidic conditions. Therefore, desorption in the simulated rain column could have been promoted in the early stages of the study by the low pH values.

The promoted refuse fermentation as a result of leachate recycle (C6) was also evidenced by the higher pH values since this was the only column to attain a neutral pH prior to the addition of mineral salts. Further, from Figure 3.6 B it can be seen that the pH values increased simultaneously as the 'total' VFAs concentrations decreased. The batch reactor (C7) appeared to be stressed as a result of the 1000 mg l^{-1} phenol co-disposal as indicated by the low pH values which remained low until after the mineral salts addition. Watson-Craik (1987) recorded a similar accumulation of VFAs with subsequent catabolism promoted by moisture movement. The severe effect that both single elution and simulated rain had was evident in the low pH values recorded for these two columns. Further, unlike the leachate recycle and batch operated columns, these two columns exhibited only transient increased pH values following the mineral salts addition. It is worth noting that the leachate recycle and batch operated columns continuously maintained pH values around neutral despite the two resupplementations (RS1 and RS2).

The single elution and rain simulated columns were, compared to the leachate recycle and batch operated columns, characterised by low VFAs concentrations (Figure 3.6 C) in the leachates. The continuously lowered pH values exhibited by the single elution and the simulated rain columns were, thus, most probably as a result of the leaching of the buffering capacity and not due to an accumulation of 'total' VFAs. However, the relatively slow recovery in pH values of the batch operated column can be ascribed to the increased 'total' VFAs concentrations which only significantly decreased following the mineral salts addition.

Notwithstanding the consistently low pH values and increased phenol concentrations a methane concentration of 54 % (v/v) was recorded for Column C5 on day 148 after which progressive decreases were recorded. One possible explanation for this enhanced methane release could be as a result of the enhancing effect of liquid movement. The leachate recycle column (C6) similarly demonstrated elevated methane concentrations which also decreased from a maximum of 78 % (v/v) on day 177. However, it is worth noting that following the first phenol resupplementation (RS1) there was a general decrease in the methane concentrations which then steadily increased again after the second resupplementation (RS2). In similar bioreactor studies, Fedorak and Hrudey (1984) reported phenol attenuation without severe methane inhibition, with phenol concentrations ≤ 2000 mg l^{-1}, although they suggested that the phenol-degrading acid formers were inhibited at concentrations ≥ 800 mg l^{-1}. It is, therefore, surprising that the batch operated column (C7) showed signs of inhibition as demonstrated by the low methane concentrations (Figure 3.6 D). Following the mineral salts addition progressive increases were recorded which seemed to suggest an alleviation of elemental limitation. It must also be recognised that the microorganisms in the refuse used had not been acclimated to phenol before being challenged which could possibly explain the protracted period of inhibition as indicated by the low methane concentrations. The general increases following the phenol resupplementations also tended to indicate the positive effect of microbial adaption. Although nitrate and sulphate concentrations of 3.1 and 10.6 mg l^{-1}, respectively were recorded prior to the mineral salts additions, the phosphate concentrations were below

detection limits which would seem to suggest that elemental limitations existed. The simulated rain column (C8) was characterised by continuously low methane concentrations (Figure 3.6D)

### 2000 mg l^{-1} Phenol Perfused Columns

The residual phenol, pH, 'total' VFAs and methane concentrations of the columns determined during the course of this study are given in Figures 3.7 A - 3.7 D. The single elution column (C9) showed the highest residual phenol concentrations followed by the leachate recycle column (C10) which, in turn, exhibited higher residual phenol concentrations than the batch operated column (C11). Both Columns C10 and C11 only exhibited phenol concentrations approaching zero after the addition of mineral salts (Figure 3.7 A). The simulated rain column (C12) again demonstrated that a significant concentration of phenol was leached from the refuse especially during the initial 80 days although low concentrations (≤ 54 mg l^{-1}) were recorded for a considerable time and values approaching zero were only recorded after 369 days. The remaining fraction has often been referred to as the 'resistant fraction' in previous desorption studies which have also demonstrated the difficulty of desorbing this fraction (Kan, Fu and Tomson, 1994).

Despite the "recovery" of the pH values for the 1000 mg l^{-1} leachate recycle column (C6) the equivalent 2000 mg l^{-1} column (C10) only attained a neutral pH after 664 days. Surprisingly, the addition of the mineral salts did not effect a significant pH increase for this column although a marked increase was evident following the second phenol resupplementation (Figure 3.7 B). The batch operated column (C11) also showed slight increases following the mineral salts addition with a neutral pH reached and maintained following the two phenol resupplementations (RS1 and RS2). The single elution and rain operated columns, with one exception, did not exhibit a pH value greater than 6.5 during the course of the experiment.

The relatively low pH values recorded for Columns C10 and C11, prior to the phenol resupplementations, can be explained, in part, by the continued high concentrations

Figure 3.7 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of 2000ppm Phenol Co-Disposal Columns Operated with Single Elution (C9), Leachate Recycle (C10), Batch Mode (C11) and Simulated Rain (C12) During an Incubation Period of 386 Days
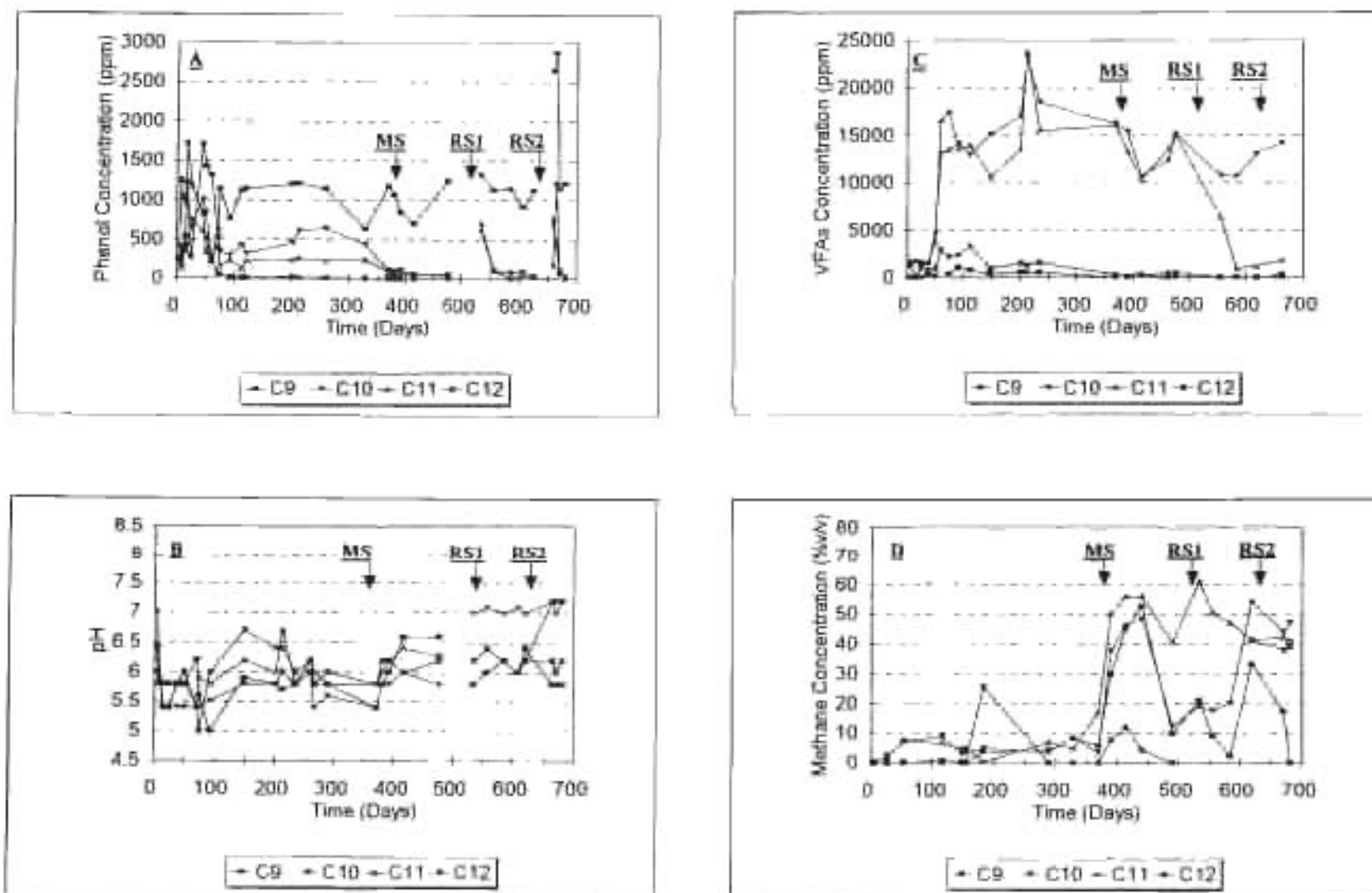
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (1000ppm)

RS2 Designates Phenol Resupplementation (2000ppm)

of VFAs (Figure 3.7 C). Unlike the batch operated column, the leachate recycle column, despite the neutral pH, constantly exhibited increased 'total' VFAs concentrations. The leaching from the single elution (C9) and simulated rain (C12) columns could be seen in the continuously low concentrations of VFAs in these columns. The low methane concentrations detected in the recycle (C10) and batch (C11) columns prior to the mineral salts additions were unlikely to have resulted from substrate limitation. A more likely explanation was the possible slow establishment of a suitable redox potential as, prior to the mineral salts additions, the leachate nitrate concentrations of Columns C10 and C11 were 3.12 and 3.1 mg l^{-1}, respectively while the equivalent sulphate concentrations were 8.9 and 6.5 mg l^{-1}. However, following the addition of mineral salts the concentrations of these alternative electron acceptors declined rapidly. Further, the nitrite concentrations prior to the additions were 139 and 8.4 mg l^{-1} but these also declined following the additions. It is encouraging to note, however, that, notwithstanding the addition of ≤ 4000 mg l^{-1} phenol, none of the fermentation indicators (pH, 'total' VFAs and methane) demonstrated significant inhibition due to shock loading.

The low methane concentrations in the single elution (C9) and simulated rain (C12) columns (Figure 3.7D) were ambiguous as the nitrate and sulphate concentrations approximated to the pre- and post-mineral salts addition concentrations. The mineral salts, however, had a significant effect on the methane generation and this could have been due to the added precursors such as bicarbonate. Displacement of such precursors would, however, have been characteristic of the single elution column (C9). Further, it must be pointed out that following the mineral salts addition the rain simulated column displayed a brief increase in methane concentrations although a dramatic increase to > 40% (v/v) was recorded after the second phenol resupplementation (RS2) The reasons for this positive enhancement of methanogenesis are not clear as no indication of enhanced refuse degradation was apparent from the catabolism parameters such as pH or 'total' VFAs. This would seem to indicate that microbial adaption was most likely responsible for the increased methane concentrations.

3.3.4 Conclusions

It can be concluded that, according to the refuse fermentation criteria of pH, 'total' VFAs concentrations and methane concentrations, the co-disposal of 1000 and 2000 mg l^{-1} phenol by single elution (Figures 3.1 A - 3.1 D) did not appear to significantly inhibit refuse catabolism. The enhancing effect of 1000 mg l^{-1} phenol co-disposal (C5) on methanogenesis was apparent although it was not clear how the added phenol effected such concentration increases as the concentration of the added phenol was, in all probability, low in comparison with more labile substrates. Significant accumulations of nitrate and/or sulphate were not recorded for this column although the re-direction of electron transport away from the nitrate or sulphate-reducing bacteria towards the methanogens could have been implicated. Watson-Craik, Sinclair, James, Sulisti and Senior (1993) reported similar increased methane productions resulting from differential inhibition and re-directed electron flow which were induced by phenol co-disposal in refuse columns. Also, the actual gas generation rate was not measured during the course of this study which would have facilitated a more accurate appraisal of the rates of methane production. To make a definitive assessment of the amounts of phenol conversion radio-labelled compounds would have to be used. Also, it is important to note the slight inhibition of these parameters, pH and 'total VFAs release, following the phenol resupplementations indicated a degree of microbial inhibition.

For the leachate recycle operated column (C6), the addition of 1000 mg l^{-1} phenol did not seem to negatively affect the onset of anaerobiosis or the terminal process of methanogenesis. In contrast, with Column C10 (Figure 3.2D) it appeared that although the process of acidogenesis was not severely affected by the added phenol, the subsequent and interrelated processes of VFA catabolism and methanogenesis were affected. These results agreed with the findings of Watson-Craik (1989a;c) who observed that acetic acid accumulations coincided with depressed methanogenesis.

Unlike the 1000 mg l^{-1} challenged column (C6) the increase in influent phenol concentration (2000 mg l^{-1}) did negatively affect the refuse fermentation and resulted not

only in a lowered pH and concomitant accumulation of 'total' VFAs and decreased methane concentrations but also in the inhibition of nitrate, and subsequent reduction of nitrite, as well as sulphate reduction. This seemed to indicate that other key members of the interacting associations could have been more vulnerable to the added xenobiotic and thus retarded the mineralisation of the perturbant. Senior and Balba (1987), for example, reported that the anaerobic fermentation of aromatic compounds required sulphate-reducing or methanogenic activity to maintain low cultural concentrations of both acetate and hydrogen.

The batch operated columns (1000 and 2000 mg l^{-1}) exhibited suppressed methane concentrations compared to the refuse only control prior to the mineral salts additions after which, and despite the phenol resupplementations, the co-disposal columns exhibited increased methane concentrations. Further, the increased phenol removal rates exhibited by Column C11 compared to C7 (Table 3.3) were not unexpected as it has been shown that increased influent concentrations can effect increased phenol catabolism (Watson-Craik, 1987). As with the leachate recycle columns, accumulations of the electron acceptors, nitrate and sulphate, demonstrated the sensitivities of the bacteria which precede the methanogens.

It appeared that the simulated rain operating regime resulted in rapid leaching and, therefore, desorption of the added phenol with concomitant low phenol catabolism.

For the 1000 mg l^{-1} challenged columns the operating regime with the highest phenol removal rate was undoubtedly single elution (Table 3.3). However, this method resulted in decreased pH values. Thus, if this method of operation was practised at a site mobilisation of heavy metals could result.

Taking into consideration the methane release rates, pre- and post-mineral salts addition, leachate recycle appeared to be the best practical option. The batch mode column indicated that this operation resulted in refuse fermentation inhibition when pH, 'total' VFAs concentrations and methane concentrations were used as analytical criteria.

However, following the minerals salts addition and the phenol resupplementations this method, compared to the leachate recycle column, gave comparable increased phenol removals. In a study made by Watson-Craik (1987) a leachate recycle strategy maintained a rate of attenuation higher than a single elution column.

From the results of the present study it was apparent that the operating regime employed plays a greater role in both refuse degradation and phenol catabolism than the concentration of the xenobiotic co-disposed. However, following the phenol resupplementations, with the exception of the single elution columns, the leachate recycle and batch operated columns exhibited phenol removal rates of the same order which seemed to indicate that at that stage the operating conditions played a less significant role (Table 3.3).

Also, the results seemed to indicate that full-scale co-disposal operations should endeavour to enhance the liquid flux through the refuse. This conclusion has been stressed by the U.K. Department of the Environment (1994) who stated that flushing bioreactor conditions are needed for successful co-disposal. An environment conducive both to microbial activity and the removal of solubilised materials depends on adequate moisture content and flux through the wastes (Department of the Environment, 1994). This then leads to a separate requirement for containment, and possibly recirculation. The hydraulic retention time, which is site specific, should, therefore, be assessed with specific attention being given to channelling or impermeable zones.

3.3.5 The Effects of Mature ('Aged') Refuse on Phenol Co-Disposal

Refuse disposed in landfills passes through sequential stages of degradation before all of the labile matter has been stabilized (Barlaz, Schaefer and Ham, 1989a; Westlake, 1990). The landfill is considered to be stable as soon as no further degradation takes place and when the catabolic products in the leachate and gas are minimal (Chapman and Ekama, 1991).

Numerous researchers have attempted to classify the different decomposition stages which refuse must pass through to give the terminal end products of CH_4 and CO_2 (Barlaz, Ham and Schaefer, 1990; Large, 1983). One of the first such classifications was presented by Farquhar and Rovers (1973) who relied mainly on gas composition data to characterize refuse decomposition. The process of degradation was divided into four stages. The stages identified were designated as Phase 1, Aerobic; Phase 2, Anaerobic Non-Methanogenic; Phase 3, Anaerobic Methanogenic Unsteady; and Phase 4, Anaerobic Methanogenic Steady.

Present landfilling techniques, in general, result in numerous individual cells or layers being formed each in its own phase of degradation. Further, due to the longevity of modern and deep landfills the major proportion of the refuse will be 'mature' before the site is closed. This implies that the relevant microbial associations will have established and the mineralisation of organic compounds will be proceeding. The process of refuse degradation to the terminal end products, methane and carbon dioxide, is a dynamic and highly complicated process and clearly cannot be reduced into simplistic compartments (Senior and Balba, 1990).

In this study the phenol challenged columns (C5-12) were used as the controls for the experimental 'aged' refuse columns (D1-D6).

Single Elution Columns

From Figure 3.8 A it can be seen that the residual phenol concentrations of the single elution, 1000 mg l^{-1} perfused, column (D1), over a period of 350 days, did not notably differ from those recorded for Column C5 with mean removal efficiencies recorded of 39 (0.21g kg^{-1} d^{-1}) and 42% (0.24g kg^{-1} d^{-1}), respectively. After 80 days operation the pH values recorded for the 'aged' refuse leachate were lower than the values recorded for Column C5 (Figure 3.8B). This was not unexpected as Watson-Craik (1987) demonstrated that 'fresh' (1 month) refuse had a buffering capacity which was considerably higher than in mature (2.3 years) refuse samples. The slight increase in the pH of D1 after 70 days,

compared to Column C5, could have resulted from both a relatively low 'total' VFAs concentration (Figure 3.8C) and a greater inoculum size in the mature refuse. The latter explanation appeared less likely as the headspace methane concentrations recorded for Column D1 progressively declined as the experiment progressed (Figure 3.8D). The decline in methanogenic activity in Column D1 could have resulted from an exhaustion of methane precursors although the 'total' VFAs concentrations (Figure 3.8 C) did not support this possibility. Following the phenol resupplementation (RS2), however, the methane concentrations increased again.

It, therefore, appeared that 'aged' and metabolically active refuse did not offer any advantages over fresh refuse as regards phenol attenuation and degradation.

Similarly, the results of the 2000 mg l^{-1} perfused columns operated with single elution (D4 and C9) were comparable during the initial 350 days (Figure 3.8 A). The average phenol removal of Column D4 was 56.8% (0.48g kg^{-1} d^{-1}), while the equivalent values for Column C9 were 57.3% (0.57g kg^{-1} d^{-1}). From these results it can be seen that with an increase in the influent phenol concentration, from 1000 mg l^{-1} to 2000 mg l^{-1}, a concomitant increase in phenol degradation resulted. Watson-Craik and Senior (1990) similarly indicated elevated removal rates with increased organic loading rates although these rates decreased with influent phenol concentrations ≥ 565 mg l^{-1}.

Despite the lower pH values of the 'fresh' refuse column (C9) (Figure 3.8 B) compared with Column D4 both columns exhibited, during the first 370 days, pH values of $<$pH6. The lower pH values in Column C9 were not unexpected since this column was characterised by significantly higher VFAs concentrations (Figure 3.8 C). Notwithstanding the higher VFAs concentrations, the methane headspace concentrations were comparable with both columns showing low ($<$8% v/v) methane concentrations. The lower VFAs concentrations recorded with the mature refuse were probably due to the previous degradation of labile substrates rather than phenol inhibited acidogenesis.

Figure 3.8 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of Phenol Co-Disposal with 'Aged' (D1 and D4) or 'Fresh' (C5 and C9) Refuse in Columns Operated with Single Elution During an Incubation Period of 386 Days
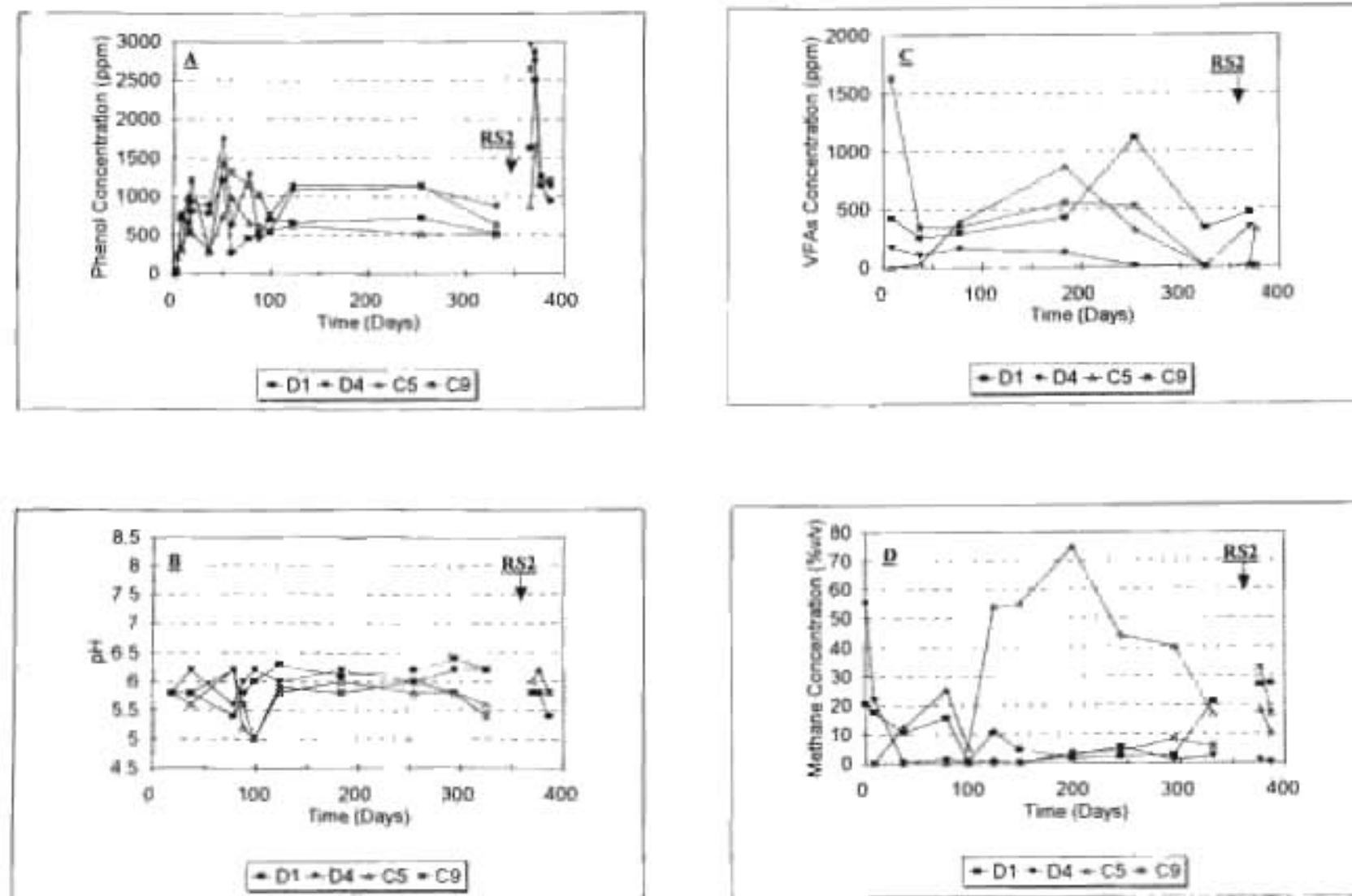
Leachate Recycle Columns

The initial (days 1 to 50) residual phenol concentrations of Column D2 were indicative of the apparent inability of 'aged' refuse to readily adsorb phenol (Figure 3.9 A). Knox (1989) similarly indicated that the amount of xenobiotic (phenol) adsorbed was dependent not only on the type of compound but also to a great extent on the age of the refuse. Despite the apparent decline in phenol adsorption, however, the residual phenol concentrations rapidly decreased after approximately 50 days from which point Columns D2 and C6 demonstrated comparable concentrations. This increased desorption of phenol in Column D2 together with the mean phenol removal rate of $0.0033g$ kg^{-1} d^{-1} compared to C6 $(0.0042g$ kg^{-1} $d^{-1})$ suggested that the mature refuse did not invariably act as an efficient source of inoculum for phenol degradation. Similar results were reported by Watson-Craik (1987), who indicated that "mature" (>3 years) refuse effected lower phenol removal rates compared with "active" (4 month) or "fresh" (1 month) refuse.

Although, like the single elution columns, the initial pH values of the mature refuse leachate were lower than the 'fresh' refuse equivalent the subsequent rapid decline in residual phenol concentrations appeared to coincide with gradual increases in the pH values (Figure 3.9B). The 'fresh' refuse column on the other hand demonstrated a rapid increase in pH, possibly due to its higher buffering capacity, and this coincided with decreases in the 'total' VFA concentrations (Figure 3.9 C). The VFAs concentrations of the mature refuse suggested that the labile acids had been catabolized. This was as expected since the establishment of a balanced fermentation often results in dramatic pH and VFA concentration changes. For example, Robinson (1989) indicated that at the Crompton Bassett Landfill site in Southern England, U. K. that the leachate exhibited a dramatic pH rise from 5.7 to 7.9 with a concomitant decrease in the COD value from 64 000 mg l^{-1} to 2 600 mg l^{-1} as the volatile fatty acids were metabolised over a 12 month period.

Despite the relatively low concentrations of VFAs recorded in Column D2 leachate the methane concentrations remained remarkably high ($+/- 45\%$ v/v) until the phenol resupplementation when a decrease was recorded (Figure 3.9 D). The higher methane

Figure 3.9 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of Phenol Co-Disposal with 'Aged' (D2 and D5) or 'Fresh' (C6 and C10) Refuse in Columns Operated with Leachate Recycle During an Incubation Period of 386 Days
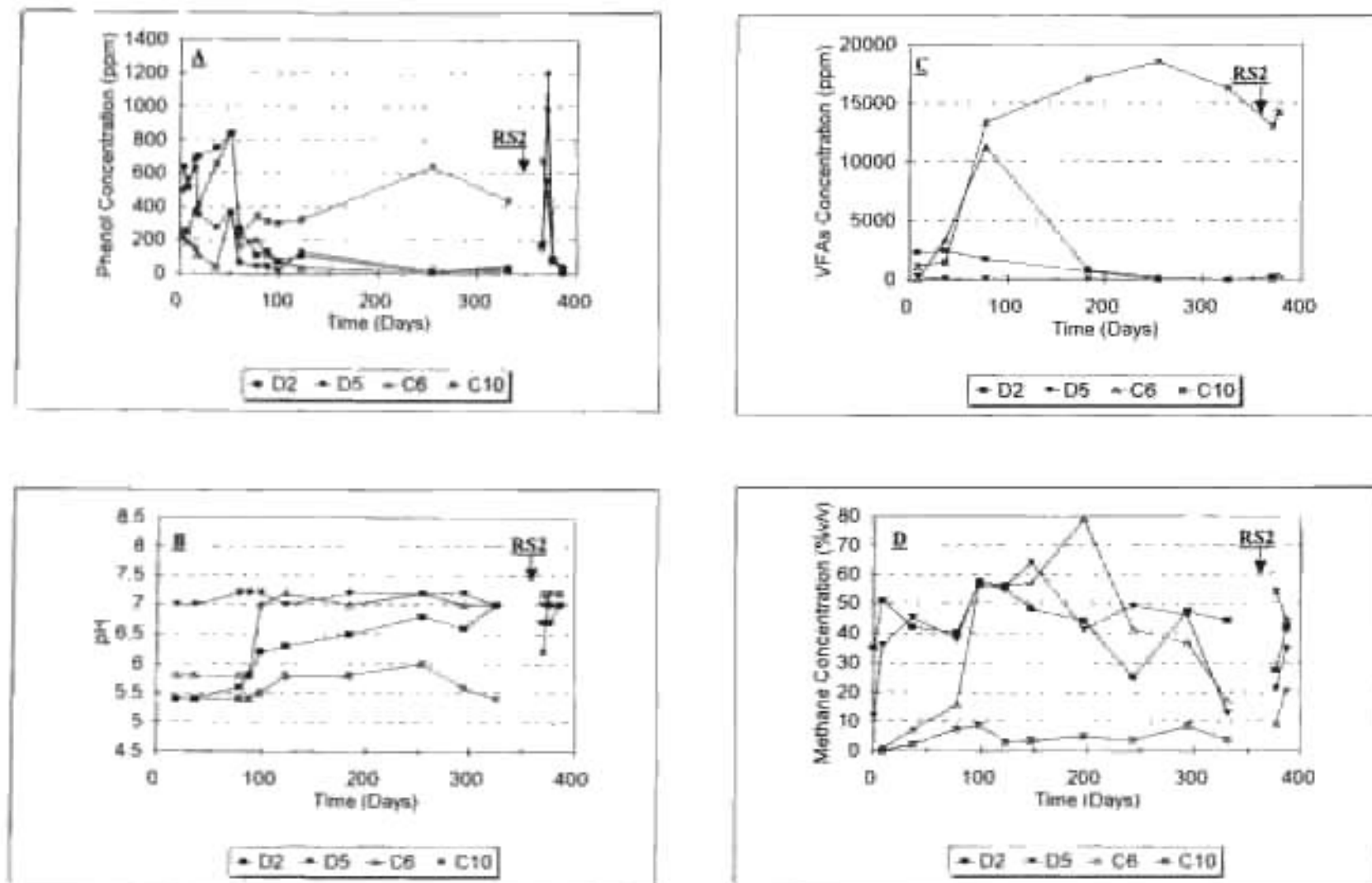
RS2 Designates Phenol Resupplementation (200 ppm)

concentrations in the early stages of the experiment were possibly due to, initially, higher numbers of anaerobic and methanogenic bacteria in the mature refuse. The headspace methane concentrations of Column C6 on the other hand showed dramatic rises to a maximum of 75% (v/v) on day 155, only to decline to a minimum of 16 % on day 329. These decreases in methane concentrations have been discussed previously (Section 3.3.2) where it was concluded that the rapid degradation of labile substrates would subsequently result in lower methane generation. This conclusion was supported by the apparent decreases in the 'total' VFAs concentrations after approximately 75 days. The results from the mature refuse column did not, however, support this view.

The 2000 mg l^{-1} phenol perfused column (D5) also showed higher initial (day 1 to 20) residual phenol concentrations although these rapidly declined to reach a concentration of 63 mg l^{-1} after 59 days and maintained a removal rate of 0.007g kg^{-1} d^{-1} compared to the 'fresh' refuse column (C10) removal rate of 0.0013g kg^{-1} d^{-1}. This catabolism could also have been facilitated by anomalously high pH values which were at all times above 7 (Figure 3.9 B). This enhanced removal of phenol and neutral pH values could have resulted from the higher inoculum size in the 'aged' refuse and the rapid establishment of a stable fermentation. However, the higher pH values also reflected the lower 'total' VFAs concentrations (Figure 3.9 C).

The dramatic effects that mature, compared to "fresh", refuse, in the presence of phenol perfusion, had on the methane concentrations can be seen in Figure 3.9 (D). Column D5 evolved methane at a concentration of 45.2% (v/v) after 36 days. This enhanced methane concentration could have resulted from a combination of the increased pH, and lower phenol and VFAs concentrations as well as increased microbial activity and numbers.

From these results the enhancing effect that the combination of leachate recycle and mature refuse had on refuse catabolism and phenol removal was evident.

Initially, the batch operated, mature refuse, column (D3) showed increased residual phenol concentrations (Figure 3.10 A) probably due to low adsorption. Subsequently, a dramatic concentration reduction was apparent after approximately 50 days. After approximately 75 days the mature refuse column demonstrated lower residual phenol concentrations than the "fresh" refuse control column (C7), possibly due to the inherent number of catabolic microorganisms in the refuse, and averaged a phenol removal rate of 0.0076g kg^{-1} d^{-1}. Barlaz, Schaefer and Ham (1989b) reported that of all the bacterial groups required for anaerobic mineralisation of refuse, it is the methanogenic bacteria which are the most lacking in 'fresh' refuse, probably as a result of their oxygen sensitivity. Therefore, the metabolically active mature refuse probably had a greater inoculum size which resulted in a more rapid progression of the fermentation. The 'fresh' refuse column (C7) maintained an average phenol removal rate of approximately 0.0012g kg^{-1} d^{-1} and the phenol concentration in this column after 350 days was still 132 mg l^{-1}.

The decreases in residual phenol concentration of Column D3 resulted despite relatively low pH values which, subsequently, increased markedly after approximately 100 days (Figure 3.10B). The 'fresh' refuse column, on the other hand, demonstrated consistently low pH values over the experimental period of 350 days. The initial decreases in pH of the mature refuse leachate could have been due to the excavation and packing processes which could have disrupted the fermentation balance although no notable accumulation of VFAs was recorded (Figure 3.10 C).

The decreases in phenol concentrations and the subsequent pH value increases of Column D3 could have resulted from an active microbial population as evidenced by the sustained high methane concentrations (Figure 3.10D). These results are consistent with those of Senior and Balba (1987) who indicated that phenol degradation required an active methanogenic population. The consistently lower pH values of Column C7 leachate could have been due, partly, to the relatively high 'total' VFAs concentrations (Figure 3.10 C).

Figure 3.10 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of Phenol Co-Disposal with 'Aged' (D3 and D6) or 'Fresh' (C7 and C11) Refuse in Columns Operated in Batch Mode During an Incubation Period of 386 Days
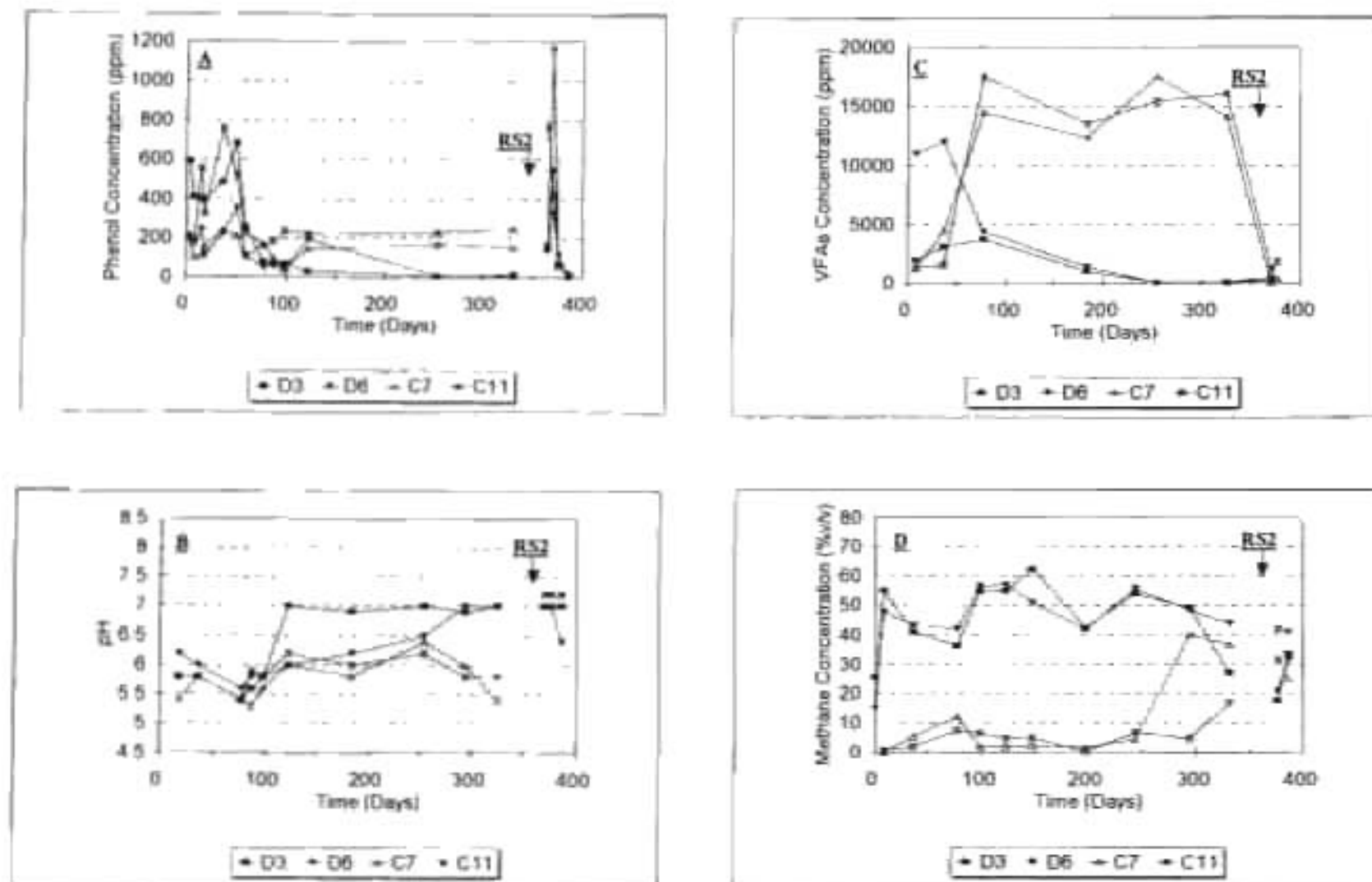
Therefore, the lower methane concentrations recorded for Column C7 (days 1 to 200) were not due to substrate limitation.

Unlike the other mature refuse columns, Column D6 at all times was characterised by lower phenol concentrations than the 'fresh' refuse control (C11) and exhibited an average phenol removal rate of 0.005g kg^{-1} d^{-1} which, unlike the 'fresh' refuse columns, was lower than the equivalent 1000 mg l^{-1} perfused column. This phenomenon could be ascribed to a greater initial microbial population which was able to rapidly adapt to the perturbant and thus effect phenol catabolism. Further, the slightly higher pH values of the mature refuse could also have been a contributory factor. The lower microbial inoculum size in the "fresh" refuse resulted in protracted periods of both adaption and phenol catabolism as well as the establishment of a balanced fermentation as evidenced by a consistently low (< 6.2) pH regime (Figure 3.10 B). This was further indicated by the enhanced acidogenesis in relation to acidotrophy. Despite the higher concentrations of 'total' VFAs recorded for Column C11 (unlike Column D6), the methane concentrations remained low and attained a concentration of 16% (v/v) after 331 days. The enhanced microbial activity in the mature refuse appeared to effect a short lag phase and elevated methane concentrations as well as enhanced phenol removal rates. These could have been due to either higher numbers of microorganisms, especially methanogens, or to adaption to the xenobiotic. The latter explanation is possible since refuse from the Umlazi landfill site could have been exposed to phenol due the operation of the site as a co-disposal landfill.

In general, it can be concluded that the mode of operation in relation to the use of mature and "fresh" refuse was critical. For example, the single elution columns exhibited no significant enhanced refuse and phenol degradation in the 'aged' refuse columns. However, with the leachate recycle and batch operated columns phenol catabolism was promoted by co-disposal with mature refuse.

3.3.6 The Effects of Perfusion Strategy on Refuse and Phenol Catabolism in Mature Refuse

1000 mg l^{-1} Phenol Perfused Columns

Columns D1-D3 were challenged with 1000 mg l^{-1} phenol and operated as detailed in Section 3.2. From Figure 3.11 A it can be seen that the phenol concentrations of both the recycle (D2) and the batch (D3) operated columns decreased rapidly from initially high values while the single elution column (D1) remained above 600 mg l^{-1} for most of the study period. The single elution column averaged a phenol attenuation of 38.5% which was equivalent to 0.21g kg^{-1} d^{-1}. The batch and recycle columns, on the other hand, averaged phenol removal rates of 0.0033 and 0.0076 g kg^{-1} d^{-1}, respectively. From these results it can be seen that single elution was the most effective strategy if the objective was to maximise the removal of phenol. One reason for this could be that in the recycle and batch operated systems the leachate phenol concentrations progressively decreased with time thus the recirculation column (D2) was operated at sub-maximum removal rates for much of the study. Although the column was competent to remove 1000 mg l^{-1} phenol from the influent (at a mean dilution rate of 0.03 h^{-1}), a removal rate of 0.006 g kg^{-1} wet refuse d^{-1} was recorded during the first 98 days while towards the latter part of the experiment (days 98 and 331) the mean removal rate decreased to 0.0004 g kg^{-1} wet refuse d^{-1}. A similar result was obtained by Watson-Craik (1987) who reported that the continuously declining influent phenol concentration resulted in a mean rate of removal decrease from 0.12 to 0.087 mg phenol cm^{-3} refuse d^{-1} between days 122 and 531 of her study.

The columns were all initially characterised by low pH values (Figure 3.11 B) although pH increases were recorded after about 100 days. Unlike in the equivalent 'fresh' refuse column (C7) the batch column (D3) attained a neutral pH on day 122 while the leachate recycle column (D2) required 325 days before the same value was reached. This was unexpected as the 'fresh' refuse columns all demonstrated increased pH values in the presence of leachate recycle. Further, Watson-Craik and Senior (1991) reported that leachate recycle actually increased and maintained the buffering capacity of refuse.

Figure 3.11 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of 1000ppm Phenol Co-Disposal with 'Aged' Refuse in Columns Operated with Single Elution (D1), Leachate Recycle (D2) and Batch Mode (D3) During an Incubation Period of 386 Days

RS2 Designates Phenol Resupplementation (2000ppm)
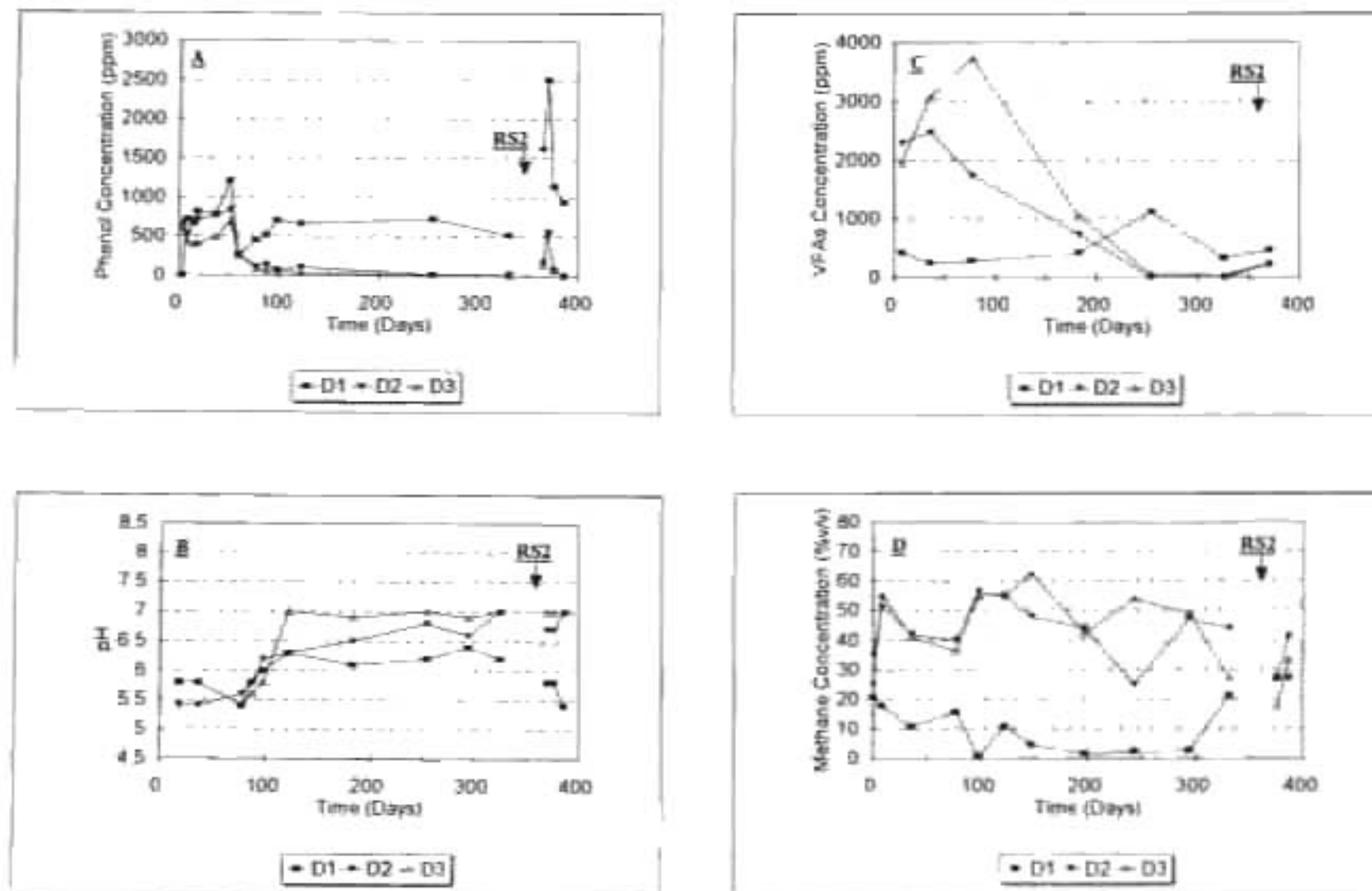
The 'total' VFAs concentrations also appeared to contradict these higher pH values in D3 as this column showed higher VFAs concentrations than the recycle column (D2). Both these columns, in turn, exhibited higher concentrations than the single elution column (D1) particularly in the early stages.

The enhancing effect that recycle and batch mode operation had on refuse catabolism was also supported by the methane concentration results (Figure 3.11 D) which were at all times higher than in the single elution column.

2000 mg l^{-1} Phenol Perfused Columns

The fermentation parameters recorded for these columns are detailed in Figures 3.12 A - D. The single elution column perfused with 2000mg l^{-1} phenol (D4) was characterised by an average removal of 56.8% which was equivalent to a removal rate of 0.48g kg^{-1} d^{-1}. As with the "fresh" refuse columns, the mature refuse columns also demonstrated increased phenol catabolism with increased influent concentration. The leachate recycle (D5) and batch (D6) columns on average maintained removal rates of 0.007 and 0.005 g kg^{-1} d^{-1}, respectively. As with the 1000 mg l^{-1} challenged columns, these columns recorded the greatest phenol removal when operated with single elution while, unlike the 1000 mg l^{-1} perfused columns, the leachate recycle column demonstrated greater removal capacity compared to the batch operated column.

From Figure 3.12 B it can be seen that only Column D5, which was operated with leachate recycle, experienced a neutral pH from the early stages of the experiment. The batch operated column (D6) showed a gradual increase in pH to attain a neutral pH on day 325. Despite these lower pH values recorded with Column D6, this column, like the recycle column, showed enhanced but comparable methane concentrations throughout the study (Figure 3.12D). The continued high concentrations of residual phenol and the low pH values appeared to inhibit methanogenesis in the single elution column (D4). The 'total' VFAs concentrations of Column D6 were relatively high in the early stages of the experiment, thus indicating a pool of labile substrates for the methanogens. In contrast, the

Figure 3.12 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of 2000ppm Phenol Co-Disposal with 'Aged' Refuse in Columns Operated with Single Elution (D4), Leachate Recycle (D5) and Batch Mode (D6) During an Incubation Period of 386 Days
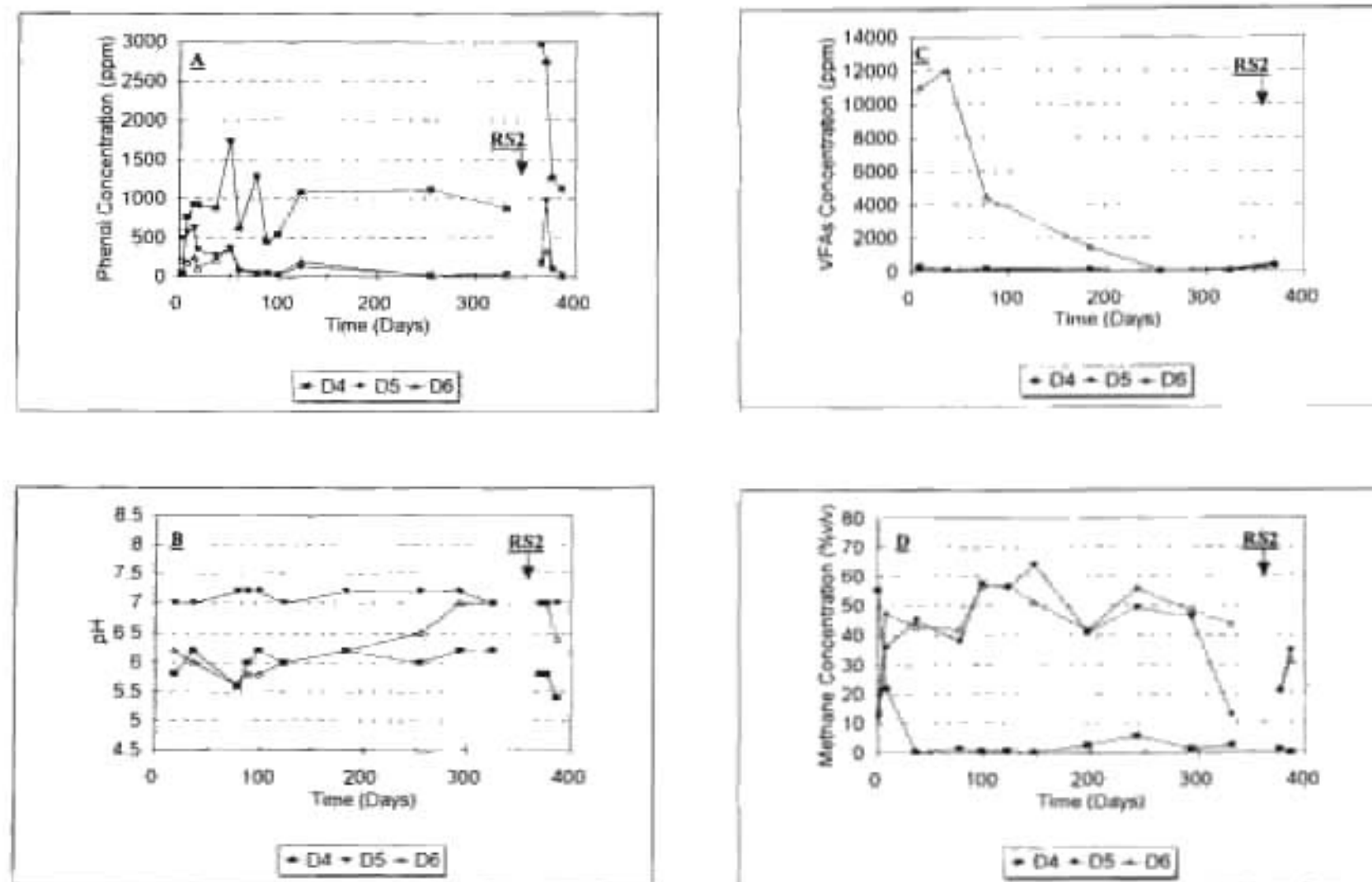
RS2 Designates Phenol Resupplementation (2000ppm)

leachate recycle column (D5) at all times showed low concentrations. Similarly, the single elution column (D4) was characterised by low 'total' VFAs concentrations thus, possibly, explaining the low concentrations of methane recorded (Figure 3.12 D).

3.3.7 Conclusions

As discussed for the 'fresh' refuse experiments, the results of the mature equivalent refuse challenged with either 1000 or 2000 mg l^{-1} phenol indicated that the perfusion regime determined the effectiveness of the refuse fermentation and/or phenol catabolism when pH, 'total' VFAs concentrations and methane concentrations were used as the analytical criteria.

Despite the seemingly inhibited refuse catabolism, the average phenol removal rates, for both the 1000 and 2000 mg l^{-1} challenged columns, prior to the phenol resupplementation (Table 3.5) were highest in the single elution operated columns. Similarly to the 'fresh' refuse columns, a significant increase in the phenol removal rate was evident with increased influent concentrations. However, the equivalent 'fresh' refuse single elution columns (C5 and C9) (Table 3.3) demonstrated increased removal rates compared to the mature refuse columns (D1 and D4) (Table 3.5). The mean phenol removal rates following the phenol resupplementation were not calculated as, unfortunately, insufficient data points were recorded for accurate scrutiny.

It must be noted that subsequent to the phenol resupplementation the single elution column, unlike the leachate recycle and batch operated columns, exhibited strong decreases in pH values. For a full-scale co-disposal landfill site an equivalent reduction in pH could be potentially hazardous with regards to the mobility and desorption of xenobiotics, especially heavy metals.

In contrast, leachate recycle through the refuse mass clearly demonstrated enhanced refuse catabolism as well as higher phenol removal rates when compared to the batch operated systems.

Table 3.5   Mean Phenol Removal Rates for the Single Elution, Leachate Recycle and Batch Operated Columns, Packed with Mature Refuse, During an Incubation Period of 386 Days

Column Number	Operating Regime	Mean Phenol Removal Rate Prior to Phenol Resupplementation ($g\ kg^{-1}\ d^{-1}$)
D1	Single Elution	0.21
D2	Leachate Recycle	0.0033
D3	Batch Mode	0.0076
D4	Single Elution	0.48
D5	Leachate Recycle	0.007
D6	Batch Mode	0.005

Further, the benefit of mature refuse for the purpose of phenol (1000 and 2000 mg l^{-1}) co-disposal in conjunction with leachate recycle and batch operation regimes was clear since significantly increased pH values and methane concentrations resulted. The mean phenol removal rates indicated that the leachate recycle and batch operated columns, with the exception of Column D2, with mature refuse (Table 3.5) maintained enhanced rates compared to the equivalent 'fresh' refuse columns (Table 3.3). Also, and possibly more importantly, the leachate recycle as well as the batch operated columns did not demonstrate any marked microbial inhibition with the phenol resupplementation. In a landfill situation this could be extremely important as the co-disposed compound will, in all probability, be added at frequent intervals which could, in effect, challenge the microorganisms repeatedly. Further, due to the apparent robustness of the leachate recycle and batch operated columns it was thought that these columns would also be more tolerant of the simultaneous and sequential co-disposal of different xenobiotics.

CHAPTER 4

ASSESSMENT OF THE IMPACTS OF ANAEROBICALLY DIGESTED SEWAGE SLUDGE CO-DISPOSAL WITH REFUSE ON REFUSE CATABOLISM

4.1   Introduction

The increasing need for efficient and environmentally friendly processes for sewage sludge disposal has been highlighted in Section 1.8. Furthermore, as discussed earlier (Section 1.8.12), the potential benefits that can be accrued from sewage sludge co-disposal with refuse are still under debate. The objectives for this study were, firstly, to determine the effects of anaerobically digested sewage sludge co-disposal on refuse catabolism and, secondly, to determine the most effective co-disposal strategy to optimise refuse catabolism.

4.2   Experimental Procedure

Sewage sludge was added to refuse in discrete layers as indicated in Figures 2.1 and 2.2 to model, as closely as possible, actual field conditions where sludge is usually poured into a trench, excavated in the refuse mass, and subsequently backfilled (U.K. Department of Environment, 1988).

For convenience, the columns (A1-A12) operated with a refuse:sludge loading of 4.5:1 were designated "high loading" columns, while the columns (B1-B12) operated with a loading of 9:1 were designated "low loading" columns.

The column headspaces were not initially overgassed and thus the redox potential of the refuse mass was allowed to self generate through the activities of the *in situ* microorganisms. The initial perfusion regimes (Section 3.2) and "start up" procedures (Section 3.3) were identical to the phenol co-disposal columns and mineral salts additions were again made (Section 3.2.1).

The anaerobically digested sewage sludge used, unless otherwise indicated, is referred to simply as "sludge". A summary of the modes of operation as well as the sludge ratios and eluents used are given in Table 4.1

Table 4.1   Perfusion Strategies and Operating Conditions for Individual "High" and "Low" Load Dual Co-Disposal Columns

Column Number and Sludge Ratio	Eluent	Mode of Operation
A1 (4.5:1); B1 (9:1)	Distilled Water	Single Elution
A2 (4.5:1); B2 (9:1)	Distilled Water	Leachate Recycle
A3 (4.5:1); B3 (9:1)	Distilled Water	Batch
A4 (4.5:1); B4 (9:1)	Distilled Water	Simulated Rain
A5 (4.5:1); B5 (9:1)	1000 mg l^{-1} Phenol	Single Elution
A6 (4.5:1); B6 (9:1)	1000 mg l^{-1} Phenol	Leachate Recycle
A7 (4.5:1); B7 (9:1)	1000 mg l^{-1} Phenol	Batch
A8 (4.5:1); B8 (9:1)	1000 mg l^{-1} Phenol	Simulated Rain
A9 (4.5:1); B9 (9:1)	2000 mg l^{-1} Phenol	Single Elution
A10(4.5:1); B10 (9:1)	2000 mg l^{-1} Phenol	Leachate Recycle
A11(4.5:1); B11 (9:1)	2000 mg l^{-1} Phenol	Batch
A12(4.5:1); B12 (9:1)	2000 mg l^{-1} Phenol	Simulated Rain

4.3   Results and Discussion

4.3.1 Microcosm Operation

In practice, the maximum concentration of sludge which can be disposed of in any landfill site depends on various site specific factors such as the refuse type and age, and the refuse and sludge moisture contents (U.K. Department of Environment, 1988). In a study made at the Coastal Park landfill site, Cape Town it was determined that the highest ratio

which could be used without compromising the operative working conditions was 4.5:1 (w/w) (Novella, 1992). This was, therefore, the highest sludge loading used in the present study.

4.3.2 Refuse and Anaerobically Digested Sewage Sludge Co-Disposal

Single Elution Columns

The single elution columns (A1, B1 and C1) (Figure 4.1 A), in general, were characterised by relatively low residual phenol concentrations (≤ 31 mg 1^{-1}) although, especially in the early stages (< 120 days), the sludge-supplemented columns (A1 and B1) showed slightly higher residual phenol concentrations than the refuse control (C1). These increased phenol concentrations, in the presence of sludge additions, could have been due to either phenol concentrations inherent in the sludge or increased phenol release due to higher degradation rates of phenol-containing compounds such as lignin. Unfortunately, the former was not verified as no sludge controls were included while the latter could have been due to the microbial inoculum added with the sludge. Increased microbial activity resulting from sludge additions has been noted by Blakey (1991) who reported enhanced refuse degradation rates due to sludge additions. Further, over the experimental period the "high load" column was characterised by higher residual phenol concentrations than the "low load" column which, in turn, had higher concentrations than the control. Following these early, and relatively high, concentrations all the columns showed residual phenol concentration decreases after 120 days.

During the first 50 days, the columns were all characterised by decreased effluent pH values (Figure 4.1 B) although the control column (C1) recovered to attain a pH of 6 on day 92, at which point, Columns A1 and B1 had pH values of 4.7. The mineral salts addition on day 359 resulted in pH increases for the effluents of Columns C1 and B1 and neutral pH values were attained after 414 and 432 days, respectively. These increases could, however, in part, be attributed to the carbonates added with the mineral salts medium and, in turn, could also explain the subsequent decline in pH values as the added

MS: Designates Mineral Salts Addition
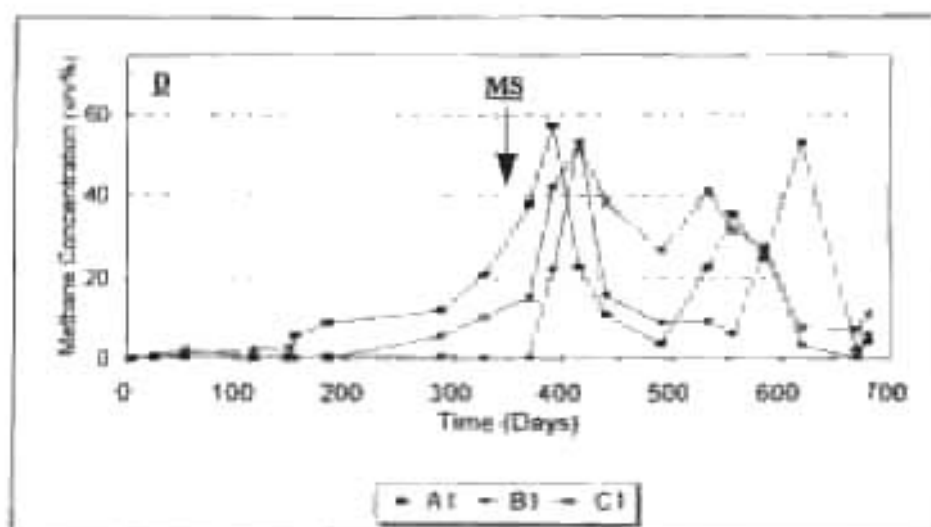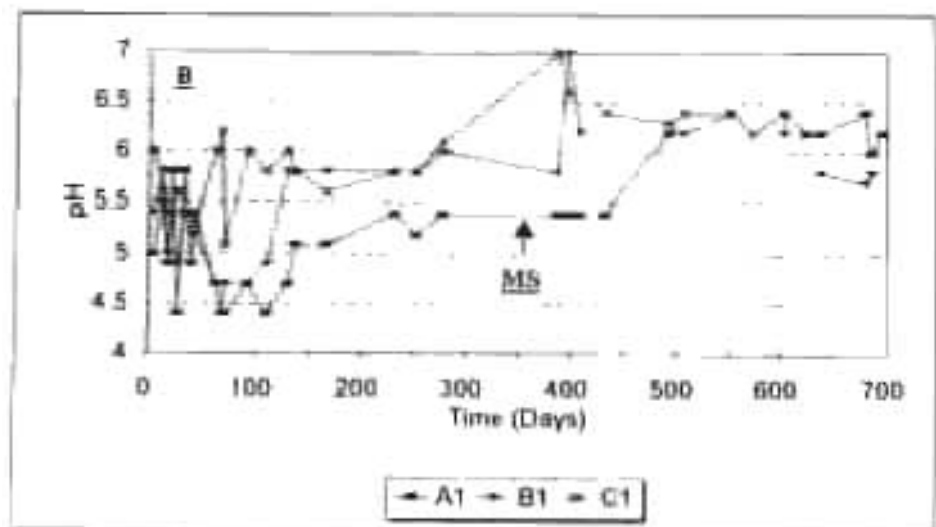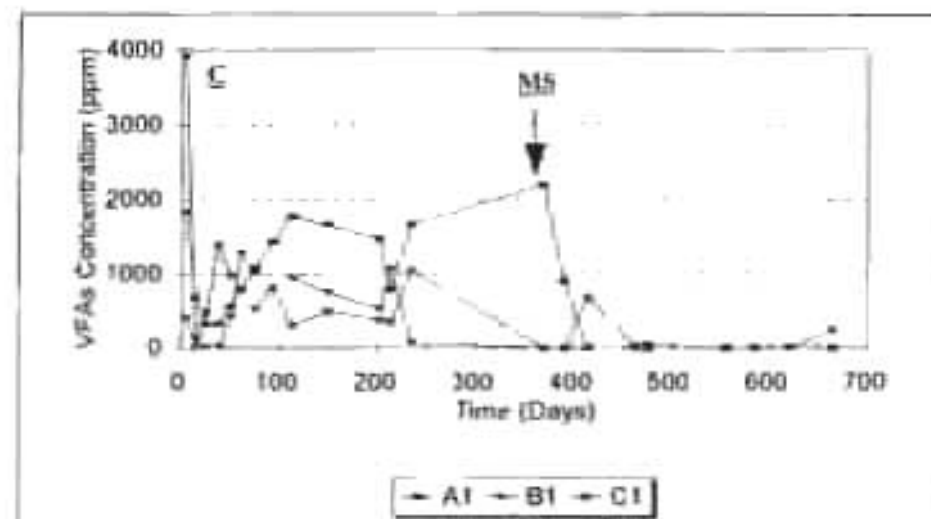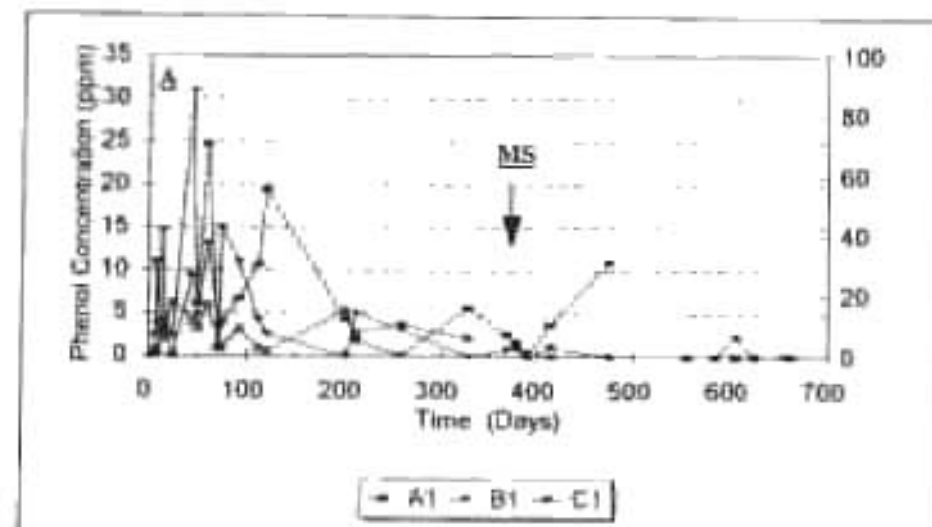
carbonate was utilised. Column A1 did not appear to respond to the same extent as C1 and B1 to the mineral salts addition and never attained a pH greater than 6.2

These lower effluent pH values as a result of sludge co-disposal were unexpected as Chapman and Ekama (1991) calculated that anaerobically digested sludge co-disposal added between 1800 and 2300 mg l^{-1} H_2CO_3 alkalinity (as $CaCO_3$). Further, in bioreactor studies with anaerobically digested sewage sludge co-disposal, Daneel (1993) recorded marked increases in the pH for sludge supplemented bioreactors compared to refuse controls. With activated sewage sludge co-disposal a similar result was obtained by Sinclair (1994) who reported a direct relationship between the sludge loading and the length of time for which the pH remained at its most acidic. The addition of activated sludge in high loadings resulted in the most rapid attainment of stable (more neutral) pH values. In contrast, Wise *et al.*, (1986) reported that sewage sludge additions in relatively low concentrations (75-400 mg l^{-1}) stimulated gas production although higher concentrations inhibited the degradation process. It must be stressed, however, that it is difficult to compare the results of studies relating to sludge addition as different studies have used different refuse:sludge ratios and different sludge types and age.

The low pH values recorded for the "high load" column (A1) could, in part, have been due to the increased 'total' VFAs concentrations (Figure 4.1 C) which, until day 220, were higher than in the "low load" column (B1) which, in turn, were higher than the refuse control column (C1). The 'total' VFAs concentrations of the effluents of Columns B1 and C1 declined after approximately 220 days and coincided with increases in the leachate pH values. A similar decline for Column A1 was not recorded until after the mineral salts addition.

The increases in VFAs concentrations as a result of sludge additions have also been recorded by other researchers. For example, Chapman and Ekama (1992) reported that sludge co-disposal resulted in VFAs concentration (determined as acetic acid) increases of between 2000 and 5000 mg l^{-1} in comparison with refuse controls. These increases, in turn, resulted in a net lower leachate pH of 5.5 (with sludge addition) in comparison with

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6.0 (without sludge addition). The authors also showed that the sludge-supplemented lysimeters initially (< 8weeks) were characterised by higher alkalinity concentrations than the refuse control but these, subsequently, rapidly decreased as the VFAs concentrations increased and the pH values decreased. Similar trends can be seen in Figure 4.1 for Column A1. In general, as the VFAs concentrations of Column A1 increased from day 1 to day 100 there was a concomitant decrease in the effluent pH value to a minimum of 4.2. Although not examined here, it has been shown (Kasali, 1986) that there is a correlation between 'total' VFAs concentration and pH.

Typically, acetate constituted the greatest part of the 'total' VFAs. Also, for all the columns the valerate and hexanoate concentrations were low and rarely exceeded 300 mg l^{-1}. Concentrations of these higher molecular weight fatty acids in leachate are usually significantly lower than the shorter-chain acids such as acetate and propionate which are the end products of the β-oxidation of fatty acids.

From these results, therefore, it appeared that despite an alkalinity increase through sewage sludge addition, enhanced acidogenesis resulted in increased effluent VFAs concentrations and lower pH values. A similar phenomenon was observed by Coutts, Dunk and Pugh (1990), who measured VFAs pool sizes at depths of 5, 10 and 15m in samples obtained from boreholes in six test cells and recorded both acetate and butyrate concentrations > 20mM in all cells. However, in a cell which had received sewage sludge, the acetate concentrations exceeded 60mM at a depth of 15m.

The types of potential pollutants resulting from sludge co-disposal with refuse can be placed into five main categories (Watson-Craik, Sinclair and Senior, 1992a):

a.   Gases;

b   Volatile fatty acids;

c.   Metals;

d.   Nitrogenous compounds; and

e.   Pathogens.

The primary gaseous products present in landfill gas (LFG) are, generally, considered to be methane and carbon dioxide (Letcher, Jarmain and Daneel 1994; Senior, 1986). Although, LFG production has, traditionally, been regarded as a liability and a problem, due to the risk of explosions and fires, and the necessity to install gas venting systems, present emphasis on gas utilisation has changed this thinking.

Despite the lower pH values and increased VFAs concentrations, the "high load" sewage sludge column was characterised by elevated methane concentrations (Figure 4.1 D). From this figure it is clear that for the "high load" column, the addition of sewage sludge affected the time required for the onset of methanogenesis. A similar, but more dramatic, decrease in the lag phase was also recorded by Blakey (1991) who concluded that sewage sludge supplementation not only greatly improved the quality of the methane content but also promoted the methane generation rates by a factor of up to 10. The increased methane concentrations recorded for the "high load" column could have resulted from either the increased concentrations of labile (acetic acid) substrates, added or generated, or the addition of greater numbers of methanogens.

The "low load" columns did not appear to benefit from the added sludge in contrast with the results of Wise et al., (1986) who reported that sewage sludge additions in relatively low concentrations (75-400 mg l^1) stimulated gas production, although higher concentrations inhibited the degradation process. Watson-Craik, Sinclair and Senior (1992b) also showed that with increased activated sludge:refuse ratios, the relative enhancing effect declined, as indicated by the lower methane generation rates. The reason for these apparently contradictory results was not clear as no evidence of microbial inhibition was recorded for the "low load" column when pH and VFAs release were used as the analytical criteria.

Of interest, however, was the apparent brief period of active methane production in the co-disposal reactors. From Figure 4.1 D it can be seen that the methane concentrations of Column A1 decreased after attaining a maximum of 57% (v/v) on day 398 to 3.5% (v/v) on day 507. This short period could have been due to numerous factors but in particular the

depletion of the labile substrates. A similar conclusion was drawn by Blakey (1991) who demonstrated initially high methane concentrations (50-60% v/v) and generation rates of 0.45 m^3 t^{-1} volatile solids d^{-1} in co-disposal experiments with subsequent cessation of gas generation recorded between six and twelve months after commencement of the study. The refuse controls, on the other hand, showed increasing gas generations over the same time period.

In contrast, in the present study, the "low load" column appeared to be inhibited as very low methane concentrations were recorded during the first 289 days although significant increases were evident following the mineral salts addition. Similarly the control (C1) was also characterised by low methane concentrations until after the minerals salts addition which again effected increased concentrations.

Other components of LFG, such as ammonia and hydrogen sulphide, can be generated under specific conditions and are, potentially, toxic. In general, gaseous ammonia should not pose a significant problem since leachate pH values appear to vary between 3.7 and 8.5 (Rees, 1980) with the result that most of the ammonia should be present in solution in the ionised form (NH_4^+). Significant concentrations of this nitrogen component can, however, be present in a landfill system. For example, Sinclair (1994) showed that the ammonium ion is the predominant inorganic nitrogen species present in activated sewage sludge and can approximate to 50% of the total available-N. Thus, the practice of anaerobically-digested sludge co-disposal should further increase the concentration of this ion. The presence of high ammonia/ammonium concentrations in sewage sludges, particularly those of industrial origin, could be problematic and could actually compromise further treatment/disposal options (Sinclair, 1994). The bactericidal or bacteriostatic effects of high NH_3/NH_4^+ concentrations is not clear as Jarrell and Saulnier (1987) indicated that at a pH of 6.5 monocultures of methanogens could withstand ammonia concentrations far in excess of 3000 mg l^{-1}. Similarly, Senior and Balba (1987) reported concentrations ≤ 2000 mg l^{-1} in leachates from microbially active landfills which indicated that the microbial populations were capable of tolerating high concentrations. However, Soubes, Muxi, Fernandez, Tarlera and Queirolo (1995) indicated that 4000 mg l^{-1}

of NH$_4^+$ effected a 50% reduction in methanogenesis and that the inhibitory effect of ammonia appeared to change according to the different bacterial species. For example, *Methanosarcina*-like cells were unaffected by a seven day exposure to 8000 mg l^{-1} of NH$_4^+$ whereas a *Methanothrix*-like population was reduced by 99% under the same conditions. Of particular interest was the finding that the residual activity, determined after the removal of the toxic ions, showed that the inhibition caused by a seven day exposure to 500 mg l^{-1} of Cr^{3+} was partially (66%) reverted whereas, for ammonia inhibition, the effect was permanent (Soubes *et al.*, 1995).

In this present study, Column A1 at all times exhibited higher 'total' ammonia concentrations (until day 386). No notable differences between the "low load" column and the refuse control (Figure 4.2A) were recorded. This was unexpected as Blakey (1991) recorded that the masses of total organic carbon (TOC), BOD, COD, sulphate, iron, manganese, zinc, nickel and lead leached from co-disposal bioreactors were lower than from the refuse control although the releases of ammonia (NH$_4^+$) and total phosphorus were higher in the co-disposal columns. Watson-Craik, Sinclair and Senior (1992b) reported similar increases in leachate 'total' ammonia concentrations in co-disposal columns compared with refuse controls with the amount of NH$_3$/NH$_4^+$ increasing as the ratio of sludge to refuse also increased. As additional NH$_3$/NH$_4^+$ would have been added with the co-disposed sludge the comparable concentrations of the "low load" column and the control could have been due to the following:

a. Dissimilatory nitrate reduction to ammonia which could be a protective mechanism against nitrite accumulation (Brock and Madigan, 1991). In this regard it is interesting to note that prior to the mineral salts addition the nitrite concentrations in the co-disposal columns were below detection limits while for the refuse control concentrations of 1.7 mg l^{-1} NO$_2^-$ were recorded. Further, according to Senior (1991) the C:N ratio in the organic fraction of refuse often exceeds 50:1 with the result that aerobic, and even anaerobic, dissimilation may be nitrogen limited. This situation of high organic contents and limited nitrogen sources has, according to Tiedje (1988).

Figure 4.2 Changes in Residual 'Total' Ammonia Concentrations for the "High Load" (A1, A2, A3), "Low Load" (B1, B2, B3) and Refuse Only (C1, C2, C3) Columns Operated with Single Elution (A), Leachate Recycle (B) and Batch Mode (C) During an Incubation Period of 308 Days
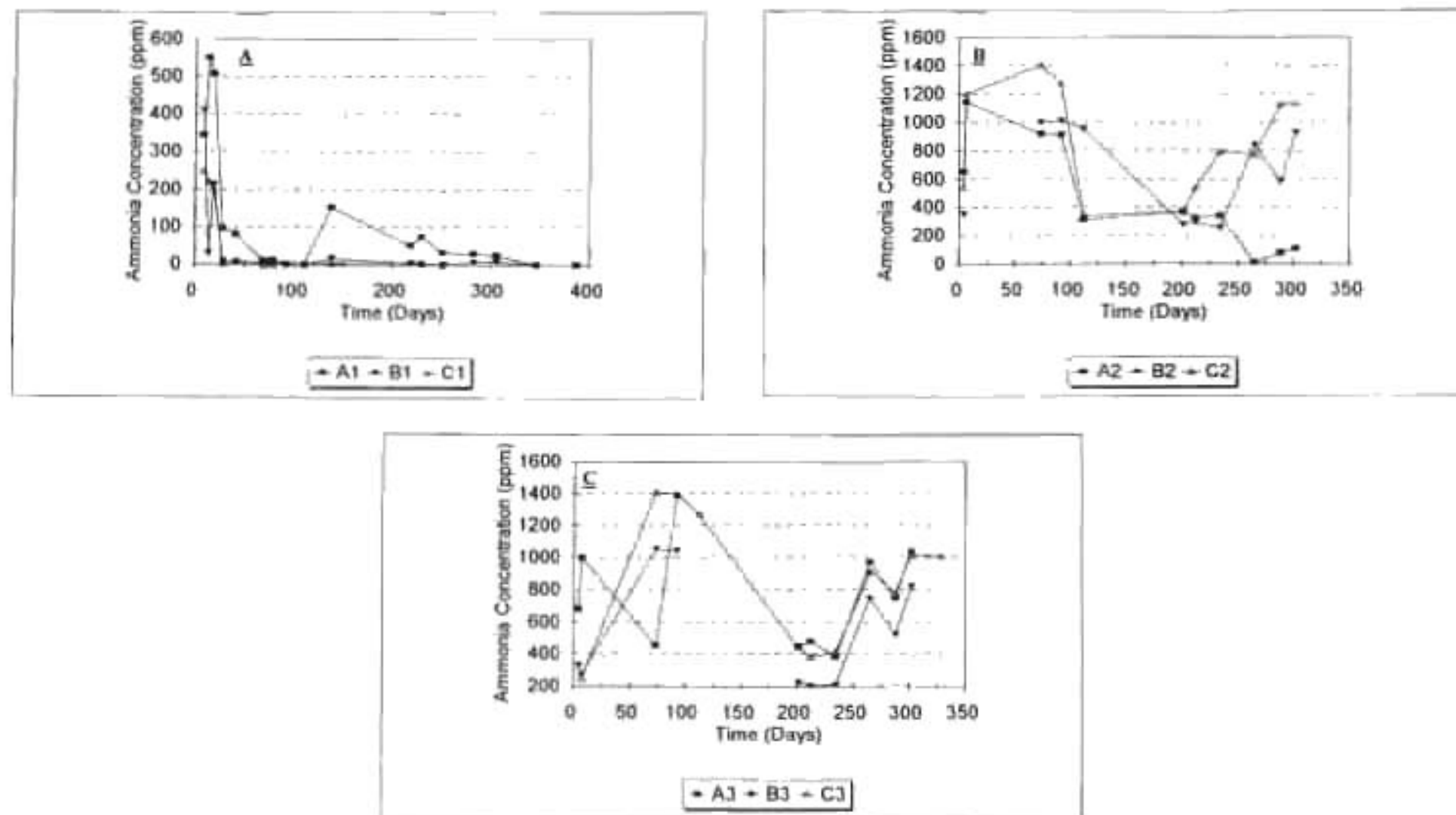
been shown to often favour dissimilatory nitrate reduction to ammonia which could explain the relatively high ammonia concentrations in the refuse control;

b. Nitrification is an oxidative process which could be active in refuse, especially during the early stages when pockets of air are likely to be trapped. As the columns were not overgassed with an inert gas air could have been trapped within the refuse Also, during the packing of the columns with the sewage sludge no effort was made to maintain anaerobiosis which, in turn, could have facilitated significant nitrification rates; and

c. The type of sludge used is also of paramount importance as well as the type and degree of pretreatment. The sludge used in this study could have been low in ammonia due to nitrification as a result of pretreatment steps.

However, according to Sinclair (1994) until the factors which influence the ultimate fate of nitrate in a landfill ecosystem are identified, it is difficult to speculate on the effects which sludge additions to refuse have on nitrate reductive processes.

The low concentrations of nitrate determined for all the microcosm effluents were not unexpected. Despite the possibility of adding additional nitrate with the sludge, this ion acts as an efficient electron acceptor during denitrification (Tiedje, 1988; Sinclair 1994). Further, the release rates of the sulphate and phosphate ions were found to be independent of the loading strategy

Apart from the potential pollutants, such as NH_3/NH_4^+ and heavy metals, added as a result of co-disposal, this practice could also introduce essential nutrients, especially nitrogen and phosphorus, which could be beneficial to the landfill ecosystem as it has been indicated that refuse masses may be either nitrogen (Senior and Balba, 1987) or phosphorus (Senior, 1991) limited. In view of this, the potential of sludge to supply these nutrients has been recognised (Buivid et al., 1981; Barlaz et al., 1990). For example, in a laboratory study, Leuschner (1989) operated six lysimeters under various conditions of buffer,

nitrogen, phosphorus, anaerobically digested sludge and septic tank residue additions. The author reported that the reactors seeded with both nutrients (nitrogen, as urea, and phosphorus) and buffer exhibited methanogenic lag phases approximately 70 days shorter than control reactors which had no added nutrients. Further, the continuous addition of nutrients following the initiation of methanogenesis did not appear to improve the rate of methane formation compared to buffer only controls which suggested that the process exerted a different nutrient demand during different stages of the fermentation, with the greatest nutrient requirement during the initial stages. The addition of sewage sludge to refuse also resulted in a reduced lag phase before the onset of methanogenic activity which then occurred at a significantly higher rate than in any of the other reactors. It was suggested that the most likely reason for this was the addition of nutrients present in the sludge, with the increased microbial inoculum cited as the major reason for the improved gas generation rates. Due to the possibility of refuse masses becoming nitrogen limited it is difficult to speculate whether nitrogen additions would constitute a positive improvement or impact the environment negatively by nitrate and/or NH_3/NH_4^- toxicity.

Leachate Recycle Columns

The residual phenol concentrations of the leachate recycle columns (A2, B2 and C2) are given in Figure 4.3 A which indicates that relatively low phenol concentrations (between 5 and 20 mg l^{-1}) were detected during the first 200 days. It is interesting to note that no distinct variations in the residual phenol concentrations between the sludge loaded and refuse only columns were detected.

It is currently accepted that leachate recycle can significantly enhance refuse degradation and this has been discussed earlier (Section 3.3.3). However, depending on the loading ratio, leachate recycle appeared to affect the refuse/sludge degradation processes differently as indicated by the pH values recorded. For example, for the sludge supplemented columns, the "high load" column (A2) appeared to benefit more from the leachate recycling and attained a near neutral pH on day 201 while the equivalent "low load" column (B2) required 670 days (Figure 4.3 B). Both the sludge supplemented
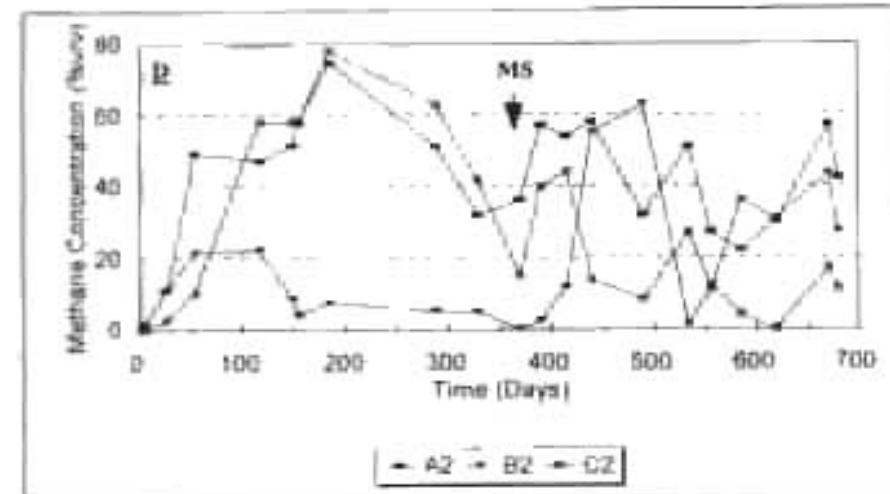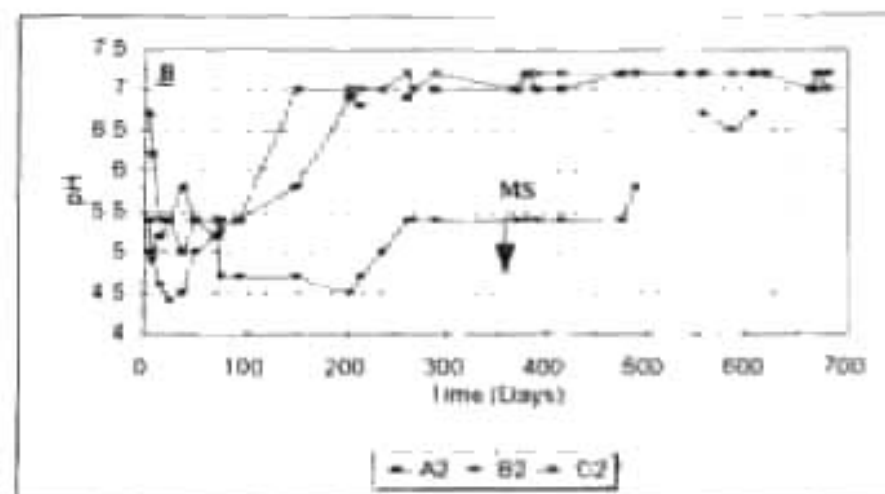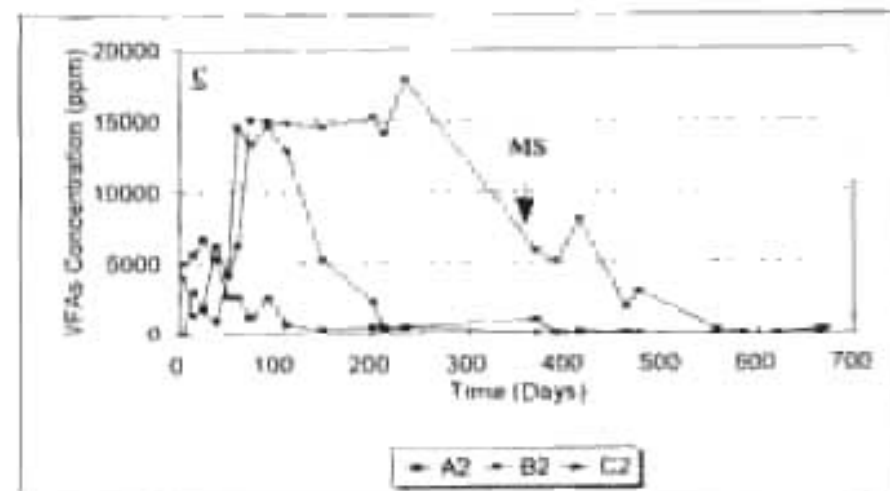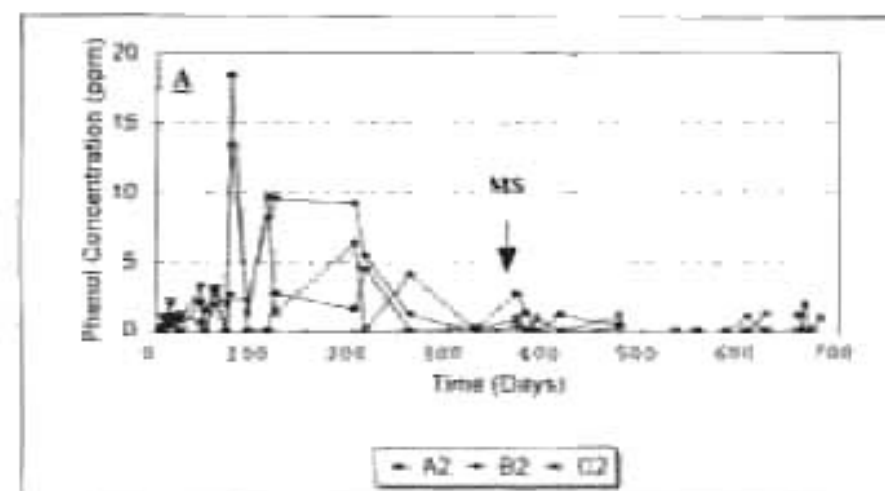
columns showed lower pH values than the control. Similar depressed pH values resulting from anaerobically digested sewage sludge addition were noted by Leuschner (1989) who also reported that the sludge addition resulted in negligible methane production. In contrast, in a lysimeter which contained sludge and buffer ($CaCO_3$) additions both the pH and methane production increased rapidly. It was also highlighted that the sludge addition did not enhance methane production but did significantly increase the rate of refuse degradation since the lysimeter which received sludge plus buffer and nutrients was characterised by a very shortened lag phase (Leuschner, 1989). On the other hand. Sinclair (1994) reported that for both leachate recycle and single elution columns, which were subjected to a second addition of activated sludge, a significant rise in pH was effected. The author attributed this directly to the concomitant increases in leachate 'total' ammonia concentrations.

The increases in pH of the refuse control column (C2) were coincident with decreases in the 'total' VFAs concentrations as shown in Figure 4.3 C. From this figure it is also clear that the increases in pH values of the "high load" column (A2) could, in part, be ascribed to the low 'total' VFAs concentrations detected after approximately 50 days. In contrast the 'total' VFAs concentrations of the "low loaded" column (B2) increased and only decreased following the mineral salts addition. These decreases in 'total' VFAs concentrations did not immediately effect increases in the pH values for this column which was surprising as Kasali (1986) showed a correlation between 'total' VFAs concentration and pH.

The reasons for the higher VFAs concentrations, compared to the refuse control, detected in the "low load" column leachate (Figure 4.3 C) were not clear although a few possibilities exist. For example, fatty acid production, in excess of catabolism, may have been greater in the presence of the low sludge loading. Leuschner (1989) mathematically estimated substrate decay rates based on the conversion of the biodegradable volatile solids to methane and volatile acids. The author indicated that the addition of anaerobically digested sewage sludge increased the decay rate from 0.228×10^3 d^{-1} to 3.24×10^3 d^{-1} and, hence, stimulated the hydrolysis of particulate material.

The most striking difference in the individual fatty acid concentrations of the various treatment leachates was observed for propionic acid (Figure 4.4 B). Both the high sludge loading and refuse control columns were characterised by similar initial high concentrations which decreased after approximately 120 days. The low sludge loading, on the other hand, exhibited a progressive increase in concentration. It seemed likely, therefore, that with the low loading the addition of the sludge had a stimulatory effect on the generation of the intermediate propionate which then accumulated due to its slower turnover rate (van Lier, Grolle, Frijters, Stams and Lettinga, 1993). Similar transient propionate accumulations were observed in other refuse column studies (Watson-Craik and Senior, 1989b).

During the initial stages the "high load" column (A2) showed increased methane concentrations (Figure 4.3 D) compared with the control and reached a concentration of 42% (v/v) after approximately 50 days. Following this, Columns A2 and C2 exhibited similar trends and attained their respective maximum methane concentrations of 74.5 and 78% (v/v) after 183 days, after which the methane concentrations declined. Although the methane concentrations of A2 recovered following the mineral salts addition and remained relatively high until the termination of the study, the refuse control, after first increasing, progressively decreased to a terminal concentration of 11.1% (v/v) after 695 days. The "low load" column (B2) also initially (< 50 days) showed increased methane concentrations compared to C2 although, subsequently, the concentrations declined to near zero prior to the mineral salts addition, following which marked increases were again recorded. The enhancing effect which the "high load" (A2) had on microbial catabolism of refuse, as recorded by increased pH values and methane concentrations, could be ascribed to one or a combination of the following:

a.   The increased inoculum size;

b.   The higher concentration of added buffer; and

c.   The added substrate in the form of labile organic material in the sludge.

Figure 4.4 Changes in Residual Propionic Acid Concentration for the "High Load" (A1, A2, A3, A4), "Low Load" (B1, B2, B3, B4) and Refuse Only Control (C1, C2, C3, C4) Columns Operated with Single Elution (A), Leachate Recycle (B), Batch Mode (C) and Simulated Rain (D) During an Incubation Period of 695 Days
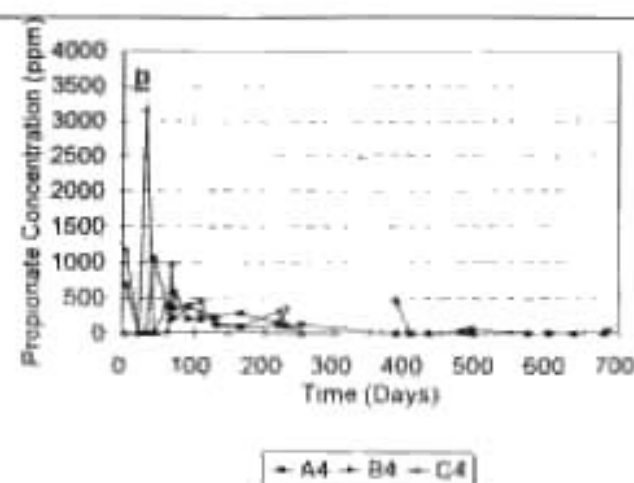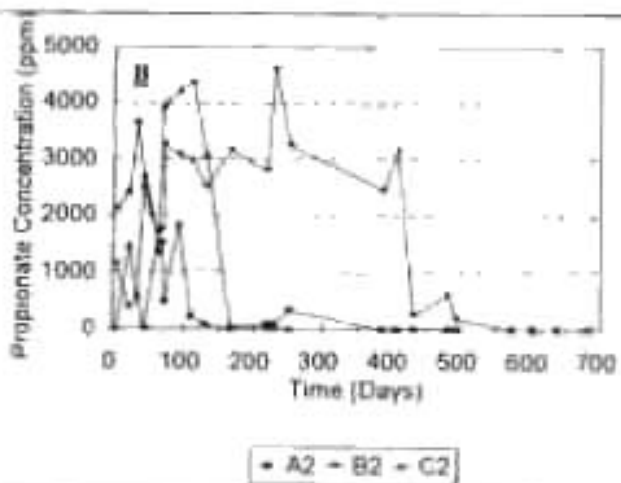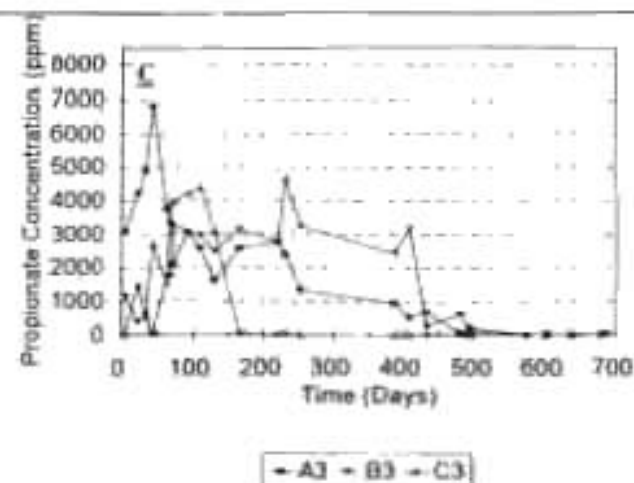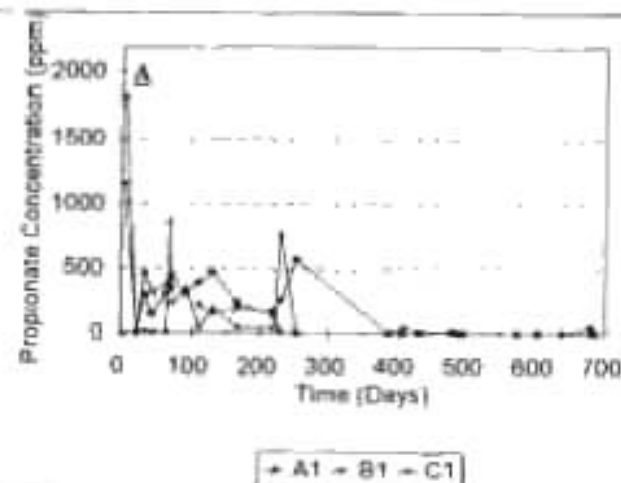
However, the low sludge loading did not appear to effect a similar increase in microbial activity and low methane concentrations resulted. A similar observation was reported by Stegmann and Spendlin (1989) who stated that the addition of sewage sludge with a low solids content was probably insufficient to act as an inoculum. Leuschner (1989) showed in six laboratory-scale digesters that the addition of sewage sludge was an excellent source of microbial inoculum, although the reactors which received sludge together with buffer and nutrient additions recorded enhanced refuse degradation and methane production. In the present study, the benefit of adding nutrients, which included carbonate (as $CaCO_3$), was apparent for the "low load" column which indicated the possibility of nutrient deficiency and/or low buffering capacity. Despite the addition of nutrients which are inherently present in sewage sludge the results of this study, as well as the six digesters studied by Leuschner (1989), seemed to indicate that either insufficient labile nutrients were present in the sewage sludge or the nutrients were recalcitrant. The mineral salts addition contained not only macro-nutrients but also micro-nutrients such as vitamins which may not have been present in the added sludge. For example, Speece and Parkin (1986) reported that a domestic sewage digester at Baltimore, MD, was not operating properly and had a 'total' fatty acid concentration of 2000 mg l^{-1}. When a sample of the sludge was "spiked" with 50 mg l^{-1} $FeCl_3$, the methane production rate increased by 167% compared with a control. This stimulation with $FeCl_3$ addition was noted in spite of the fact that the soluble iron concentration was 12.4 mg l^{-1}. The authors speculated that many anaerobic digesters suffer from trace metal deficiency even when the feed stock is rich in trace metals.

The residual 'total' ammonia plots (Figure 4.2 B) for the leachate recycle columns indicated that the refuse control microcosm (C2) was characterised by the highest residual concentrations in the early stages with a decrease recorded after 107 days followed by an increase. In contrast, the "high load" column (A2) showed a decrease after 112 days which did not subsequently increase. The observation of higher 'total' ammonia concentrations in the refuse control column was contrary to what was expected since sludge co-disposal should add NH_3/NH_4^+ to the system. Sinclair (1994) also reported a similar phenomenon

and found a higher overall loss/mobilisation of 33% nitrogen from an activated sludge co-disposal system compared with a refuse control (21%).

The results for the control and the "low load" columns illustrated two phases of 'total' ammonia release ie. initial (+/- 100 days) flushes of high concentrations after which declines were noted only to be followed by further increases. Similar 'total' ammonia release phenomena were recorded by Sinclair (1994) who noted that the initial "flush" of ammonia tended to last for about two weeks and this he attributed to three factors:

a.   The presence of "free" ammonia NH_4^+/NH_3 in either the sludge or refuse;

b.   The presence of labile N-containing compounds which generated NH_4^+/NH_3 during catabolism; and/or

c.   The dissimilatory reduction of nitrates within the system to form ammonia.

The leachate nitrate concentrations of all the columns were at all times very low which was not unexpected as nitrate is an effective electron acceptor. Similar low concentrations of nitrate were observed by Barlaz et al., (1989a) in refuse-filled laboratory lysimeters and were attributed to the utilisation of nitrate for the oxidation of available sugars to carbon dioxide and water via denitrification. Therefore, the importance of factor number three above is expected to be relatively minor.

Batch Operated Columns

The residual phenol concentrations (Figure 4.5 A) of the refuse control column (C3) indicated, in general, low (<5 mg l^{-1}) values with a maximum concentration of 14 mg l^{-1} recorded on day 201. In general, the sludge-supplemented columns also demonstrated relatively low phenol concentrations although, in the early stages, the "high load" column showed the higher concentrations (≤34 mg l^{-1}).

Column C3 was characterised by a sharp decline in pH (Figure 4.5 B) from near neutral to pH 5.4 over a period of 15 days. The pH then remained depressed until

Figure 4.5 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of "High Load" (A3), "Low Load" (B3) and Refuse Only Control (C3) Columns Operated in Batch Mode During an Incubation Period of 695 Days
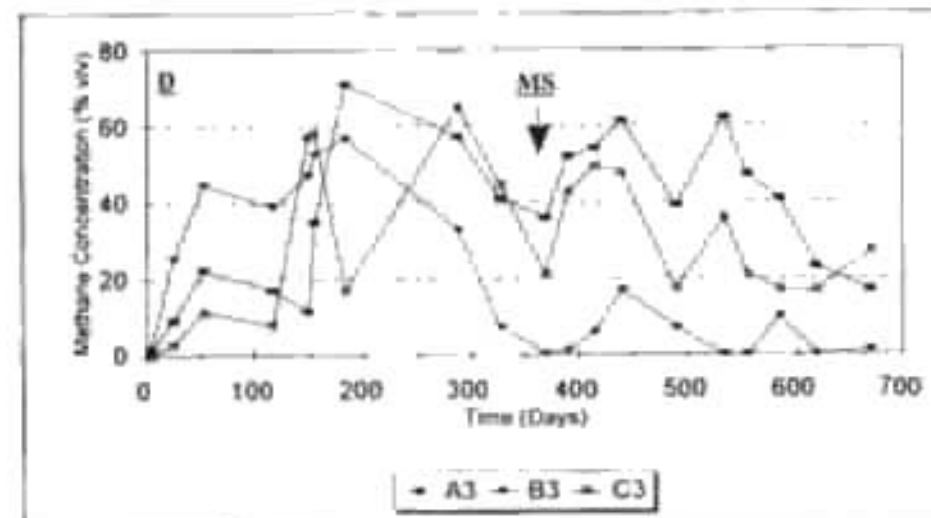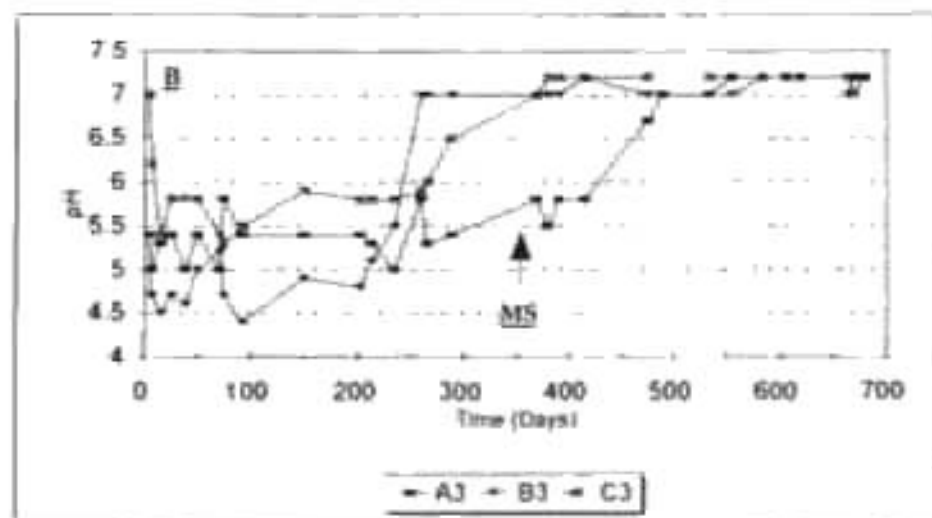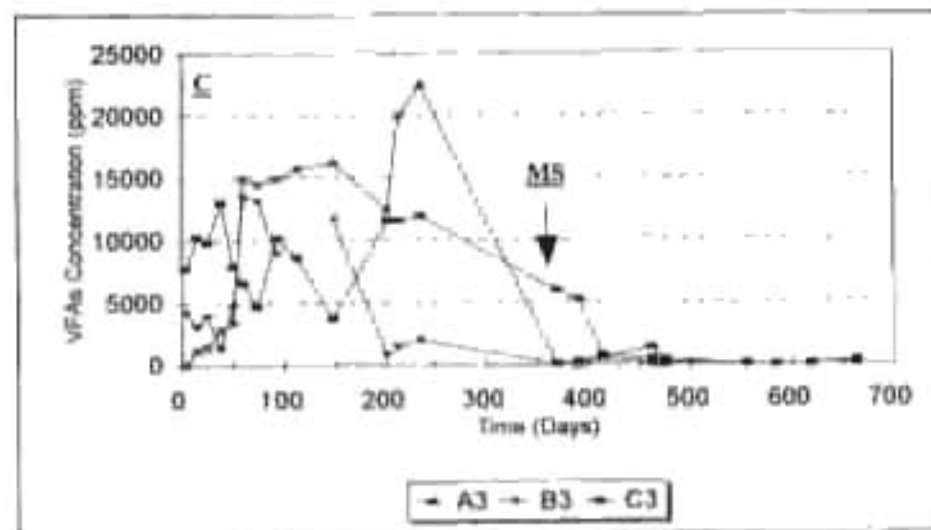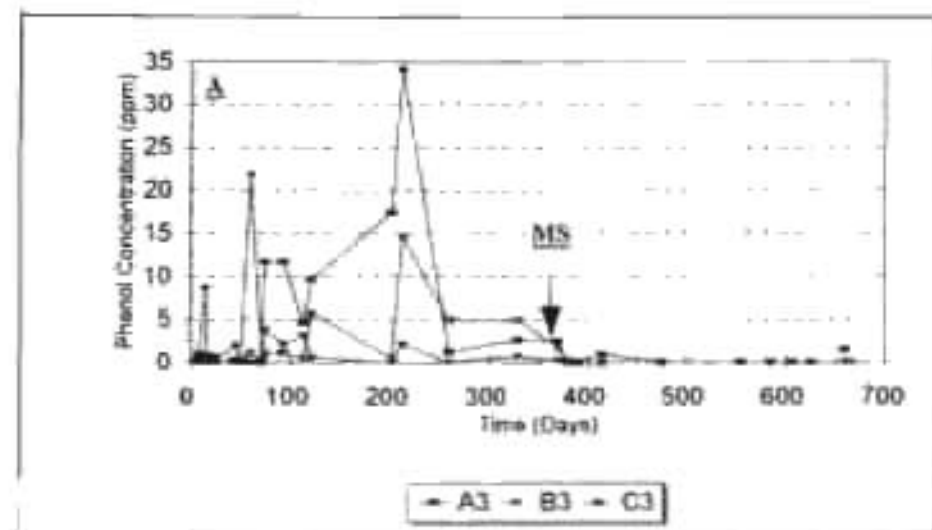
MS Designates Mineral Salts Addition

approximately day 230 after which a progressive increase was evident which reached a neutral pH after 370 days. From the initial pH value of 5.5 of the "low load" column (B3) there was a further decrease to a minimum of 4.4 on day 90 before increases were recorded to a neutral value on day 252. During the first 240 days, however, the pH values of Column B3 were significantly lower than the control (C3). The "high load" column (A3) also exhibited lower pH values than the control during the first 212 days although these values were higher than the "low load" column.

The effects that the added sludge had on the 'total' VFAs concentrations are shown in Figure 4.5 C. From this figure it can be seen that the 'total' VFAs concentration at the start of the experiment was near zero in Column C3. In contrast, the initial concentration increased as the sludge loading increased. This seemed to imply that the fatty acids were either generated to a great extent from the added sludge, a phenomenon also observed by Sinclair (1994), or the generation was promoted by the sludge addition. Although the pH values of Column B3 during the first 50 days were lower than A3 the VFAs concentrations were the converse. Further, it is interesting to note that despite the very low initial 'total' VFAs concentrations of the refuse control, the concentrations increased to values greater than both the sludge-supplemented columns and decreased only after the mineral salts were added.

As with the leachate recycle columns, transient accumulations of propionic acid were also recorded with these batch operated systems (Figure 4.4 C).

The methane concentrations (Figure 4.5 D) of C3 tended to be erratic although a concentration of 58% (v/v) was recorded after 148 days after which the concentrations progressively declined. Concomitant with, and despite, the initial low pH values of the "low load" column (B3) methane concentrations of 44% (v/v) were recorded after 52 days. Surprisingly, however, these relatively high methane concentrations soon decreased to near zero after 282 days. These decreases coincided with a rapid decrease in the 'total' VFAs concentration although it is doubtful whether this could have been the sole reason for the decline in methanogenesis. Notwithstanding the protracted period of depressed pH values

recorded for Column A3 a methane concentration of 71 % (v/v) was recorded on day 153. Of interest, however, was the initial (<148 days) higher methane concentrations recorded for the "low load" microcosm compared to the equivalent high sludge loaded column. Why enhanced methanogenic microbial activity was more evident with the lower loading was unclear. With the higher loading no apparent evidence of inhibition was observed when accumulations of VFAs, pH values or electron acceptor concentrations were used as analytical criteria. This more rapid onset of methanogenesis as a result of sewage sludge additions has been noted by other researchers (Wise *et al.*, 1986; Leuschner, 1989) and implies that a more balanced refuse fermentation had been attained. As a result of a balanced refuse fermentation an increase in methanogenesis is usually concomitant with a rise in the pH values (Senior, Watson Craik, Sinclair and Jones, 1991) which, together with the buffering capacity afforded by sludge co-disposal, could explain the early attainment of a neutral pH for the "low load" column.
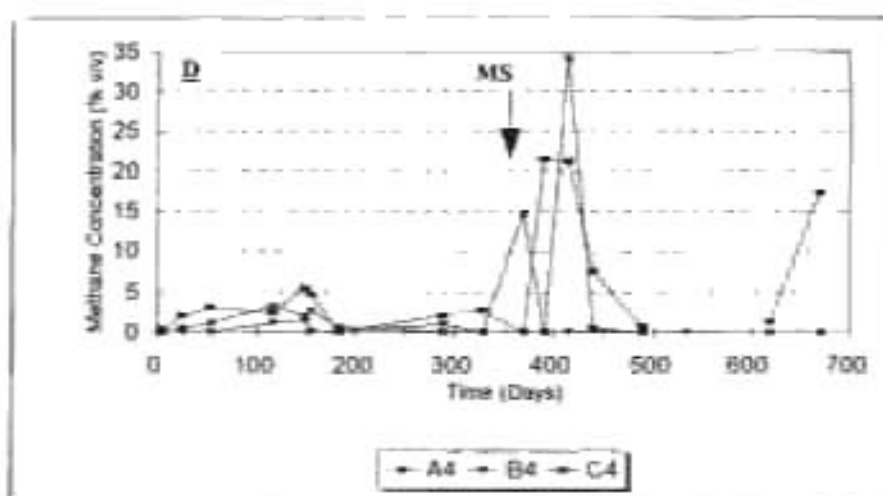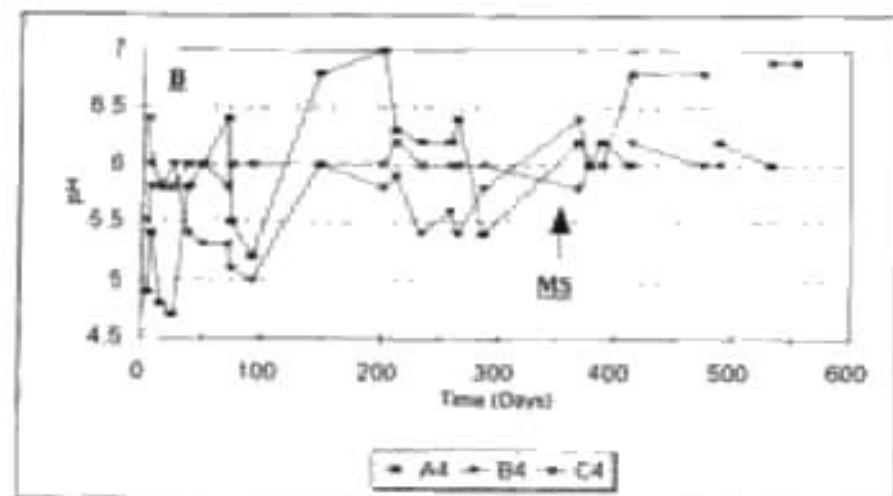
Simulated Rain Columns

The residual phenol concentrations for the "high", "low load" and refuse control columns are given in Figure 4.6 A. Low (≤5 mg l^{-1}) concentrations of phenol were recorded for the refuse control (C4) and, in general, for the "low load" column (B4) while the "high load" column (A4) showed, particularly in the early stages, slightly increased phenol concentrations.

All the rain simulated columns exhibited similar lowered pH values (Figure 4.6 B) which were, with a few exceptions, all below pH 7 prior to the mineral salts additions. Following these additions, Columns A4, B4 and C4 showed slight increases in pH while only C4 maintained pH values > 6.5. The relatively low leachate pH values of these columns were unexpected as the 'total' VFAs concentrations (Figure 4.6 C) were, in general, low for all these microcosms. One possible explanation for this could be the leaching effect of the water which would tend to remove the inherent buffering from the refuse/sludge columns. Therefore, even in the presence of relatively low fatty acid concentrations, large fluctuations in leachate pH would be possible.

Figure 4.6 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations (D) of 'High Load' (A4), 'Low Load' (B4) and Refuse Only Control (C4) Columns Operated with Simulated Rain During an Incubation Period of 695 Days
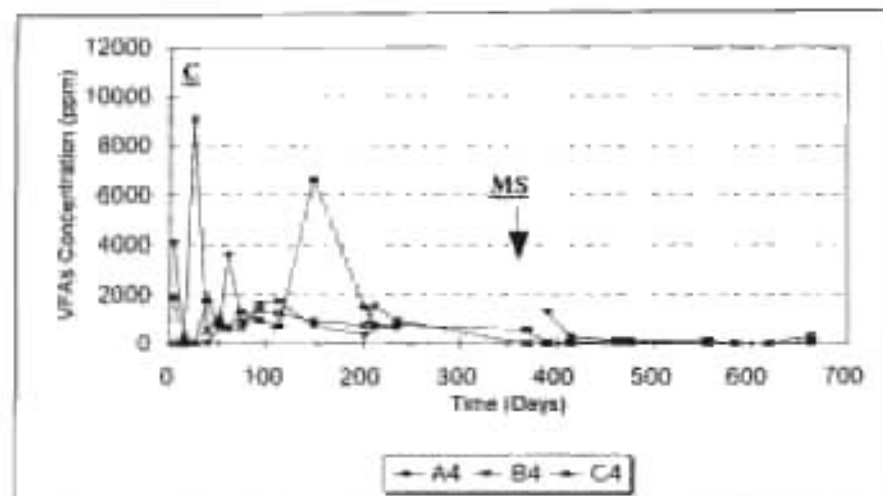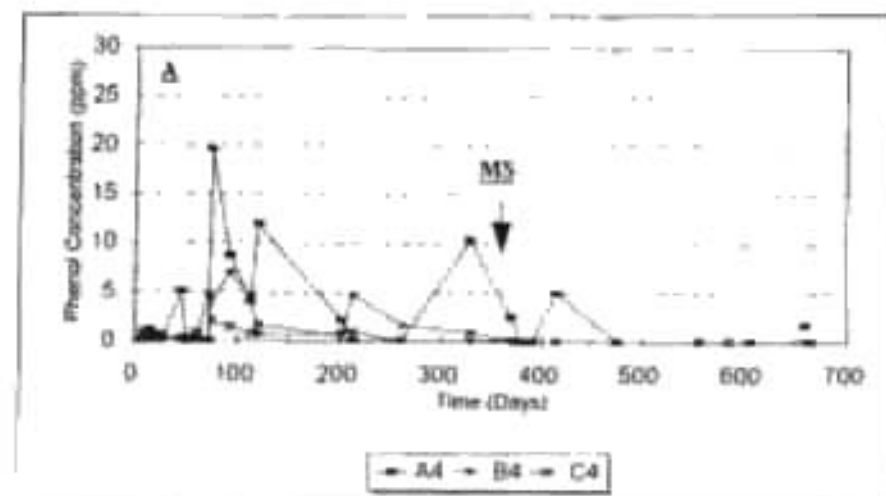
MS Denotes Mineral Salts Addition

The methane concentrations recorded during the operation of these columns are given in Figure 4.6 D which shows that all the rain simulated columns, irrespective of the co-disposal ratio, were characterised by low methane concentrations. Following the addition of the mineral salts the refuse control and the "high load" columns gave positive responses which were, however, transient and the concentrations rapidly declined to near zero.

The influence that rain has on the methanogenic fermentation of a refuse mass is not clear and may only be of relevance near the surface of the site. Clearly, in this region refuse degradation and the resulting fermentation balances will be very different compared with the majority of the refuse mass as air ingression could result in the maintenance of aerobic or microaerophilic conditions. Further, interacting variables such as moisture, temperature, pH and redox potential may also be very different compared with the bulk of the refuse mass and could effect significantly different refuse/sludge degradation processes.

4.3.3 Effects of the Perfusion Strategy in Anaerobically Digested Sewage Sludge Co-Disposal with Refuse

"High Load" Columns

In general, it can be seen from Figure 4.7 A that all the "high load" columns (A1 to A4) displayed erratic residual phenol concentrations which subsequently declined to very low concentrations following the mineral salts additions. However, the single elution column (A1) displayed the highest residual phenol concentrations during the first 70 days. These elevated concentrations could have been due to the increased desorption of the phenol due to the increased fluid movement through this column. The single elution and leachate recycle columns were subjected to different hydraulic loadings as described in Section 2.5.1.

Figure 4.3   Changes in Residual Phenol Concentrations (A), pH (B), Total Volatile Fatty Acids Concentrations (C) and Methane Concentrations of the "High Load" Columns Operated with Single Elution (A1), Leachate Recycle (A2), Batch Mode (A3) and Simulated Rain (A4) During an Incubation Period of 695 Days.
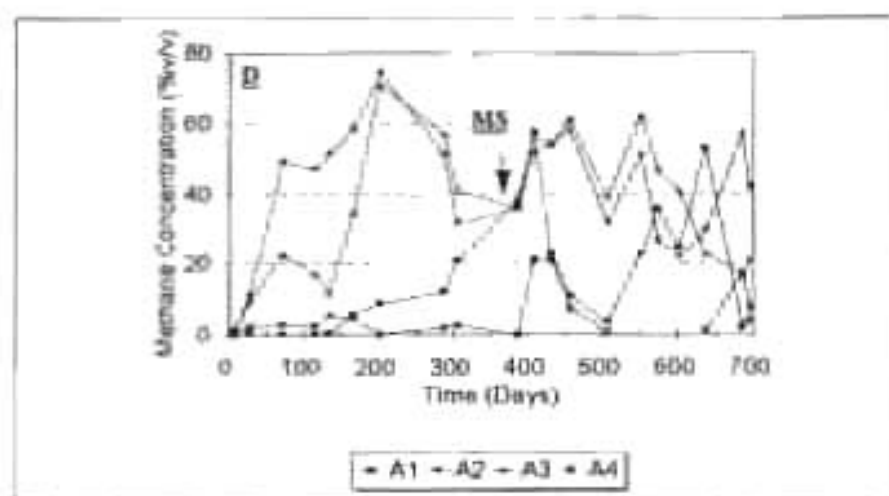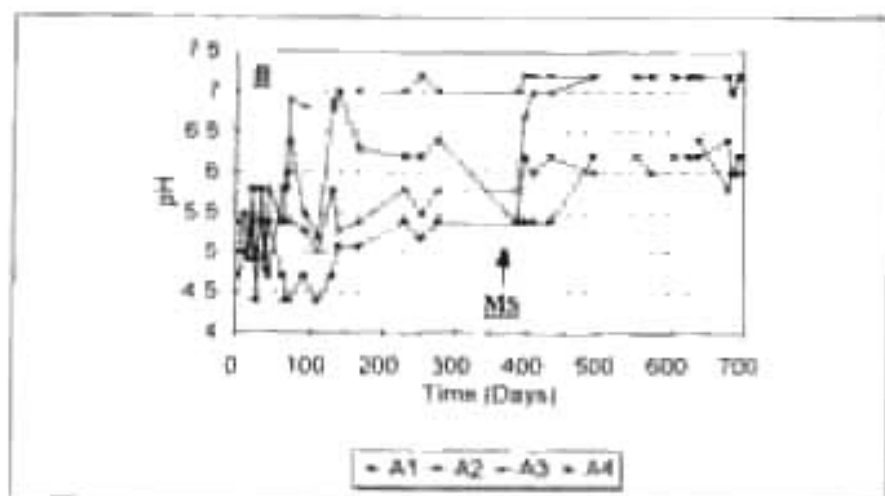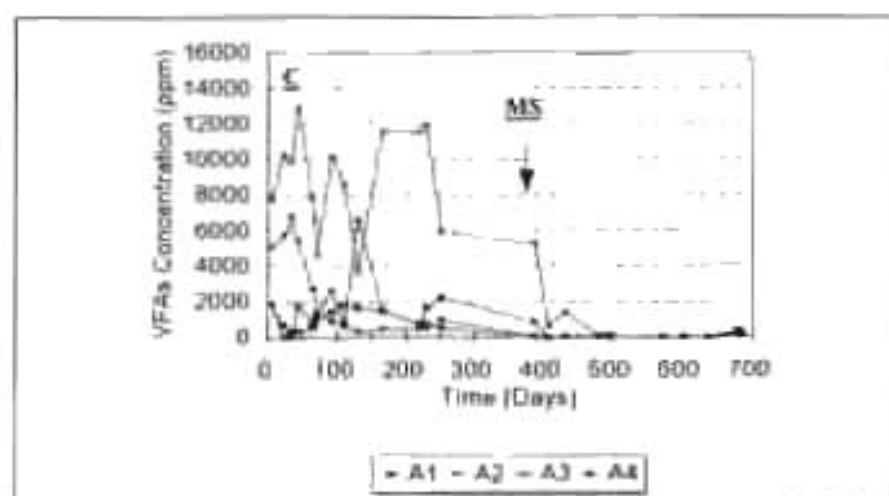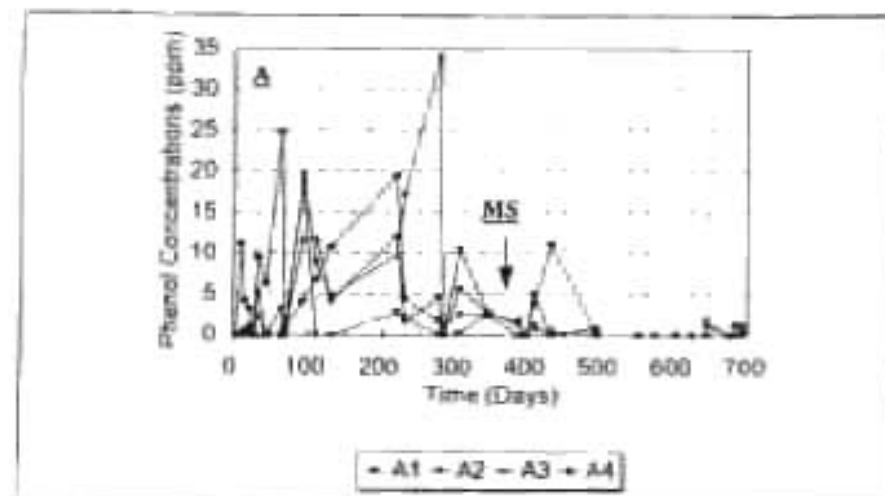
MS Designates Mineral Salts Addition

During the first 50 days, as indicated in Figure 4.7 B, all the microcosms were characterised by pH values below 5.7 and this phenomenon has been fully discussed earlier (Section 3.3.2). The single elution column (A1) leachate, with one exception, was characterised by pH values < 5.5 prior to the mineral salts addition following which there was an increase to pH values > 6. The column operated with leachate recycle (A2), on the other hand, demonstrated a rapid increase in pH to attain a value of 6.9 after 71 days. The batch-maintained microcosm (A3) showed decreased pH values until the addition of the mineral salts after which a significant increase was noted and a neutral pH was attained on day 408. Perhaps surprisingly, the rain simulated microcosm (A4) gave erratic results although a neutral pH was recorded on day 277. This pH value was, however, not maintained and the pH progressively declined to 5.4 following the mineral salts addition only to increase again and stabilise around pH 6.

The low pH values recorded for Column A1, especially prior to the mineral salts addition, could be ascribed, in part, to relatively high 'total' VFAs concentrations (Figure 4.7 C). Although decreases in the 'total' VFAs concentrations were observed before the mineral salts addition, major decreases were only evident following the addition. This seemed to indicate that the refuse fermentation was still in the acetogenic stage. It is, however, important to again note that recorded discrete fatty acid concentrations represent balances between genesis and trophy and provide only limited information about turnover rates. An accumulation of fatty acids could be detrimental to the fermentative process. The reason for this is that the acid-forming bacteria have higher growth rates than the acetoclastic bacteria which could result in 'souring' of the system and, eventually, inhibition of the methanogens. This could be detrimental to full-scale co-disposal operations with regards to methanogenesis as well as phenol catabolism. For example, Godsy, Goerlitz and Grbić-Galić (1992) demonstrated that the cooperation of several bacteria utilising different substrates and electron acceptors are required for phenol catabolism to methane and carbon dioxide.

The column operated with leachate recycle (A2) exhibited moderately high acid concentrations which rapidly decreased after approximately 45 days and were subsequently

lower than the single elution column. This was unexpected as the mode of operation of the leachate discard columns would be expected to effectively reduce the fatty acid concentrations. Sinclair (1994) reported that leachate produced from recycle columns packed with refuse or refuse/sludge mixtures, contained higher concentrations (usually by a factor of 2-3) of individual fatty acids than those generated from corresponding columns with single elution operation. However, in the present study, it was not possible to determine whether this was due to increased fatty acid generation or inhibition of fatty acid decomposition within the recycle columns.

One possibility for the higher 'total' VFAs concentrations in the single elution column leachate, after approximately 70 days, could have been the continued low pH values coupled with the relatively low methane generations which would otherwise utilise the fatty acids. The methane concentrations (Figure 4 7 D) recorded during the first 162 days for Column A1 were relatively insignificant but slowly increased to attain a concentration of 21 % (v/v) prior to the mineral salts addition. Subsequently, increases in the methane concentrations to a maximum of 57.4 % (v/v) on day 408 were recorded after which the concentrations decreased to 3 7% (v/v) on day 507 before again increasing. Unlike the single elution column which was characterised by a relatively short period of elevated methane concentrations, Column A2 showed continuously high methane concentrations with 42 % (v/v) recorded after 695 days.

It is interesting to note the relatively short period of active methane generation as recorded in the headspace concentrations of the single elution column compared with both the leachate recycle and batch mode operated columns. For example, Column A3 attained a maximum concentration of 71 % (v/v) on day 201, although there was a steady decline to 36 % (v/v) on day 387 which coincided with the mineral salts addition. These increased methane concentrations contrasted the depressed pH values of below 6. Following the mineral salts addition further increases were again noted although they were not maintained and a methane concentration of 16.1 % (v/v) was recorded on day 685.

The rain simulated column, despite initial neutral pH values, exhibited low methane concentrations and only briefly exhibited concentrations greater than 20% (v/v) following the mineral salts addition (Figure 4.7 D).

"Low Load" Columns

The residual phenol concentrations, pH values, 'total' VFAs concentrations and methane concentrations of the "low load" columns subjected to leachate discard (B1), leachate recycle (B2), batch mode (B3) and simulated rain (B4) are detailed in Figures 4.8 A - 4.8 D. During the early stages (< 100 days) the columns exhibited relatively high residual phenol concentrations with subsequent decreases after approximately 280 days. During this initial period, and similar to the "high load" columns, the single elution column (B1) showed the highest leachate phenol concentrations.

All the "low load" columns, with the exception of B1, showed the characteristic rapid decline in initial pH values. In contrast to the "high load" column subjected to leachate recycle (A2) when a neutral pH was recorded on day 68 the equivalent "low load" column required 624 days to attain the same pH. The "high load" batch operated column (A3) did not exhibit a pH > 5.8 prior to the mineral salts addition whereas with the "low load" column (B3) a neutral pH was reached on day 157. It is difficult to speculate whether the recorded differences resulted from microbial activity or were due to the variability and heterogeneous nature of the refuse. Following the mineral salts additions, however, a general increase in the pH values of all the columns was recorded. It is also interesting to note that, in general, the period required for pH increases to occur in the "high load" columns was seemingly shorter than in the equivalent "low load" columns which could have been due to the increased buffer capacity added with the higher sludge load.

The steady increase to a neutral pH, for the batch operated column (B3), was coincident with a decrease in the 'total' VFAs concentrations. Likewise, the slow recovery in pH for the leachate recycle column was concomitant with increased fatty acid concentrations which only decreased significantly following the mineral salts addition. On

Figure 4.8 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' Volatile Fatty Acids Concentrations (C) and Methane Concentrations of the "Low Load" Columns Operated with Single Elution (B1), Leachate Recycle (B2), Batch Mode (B3) and Simulated Rain (B4) During an Incubation Period of 695 Days
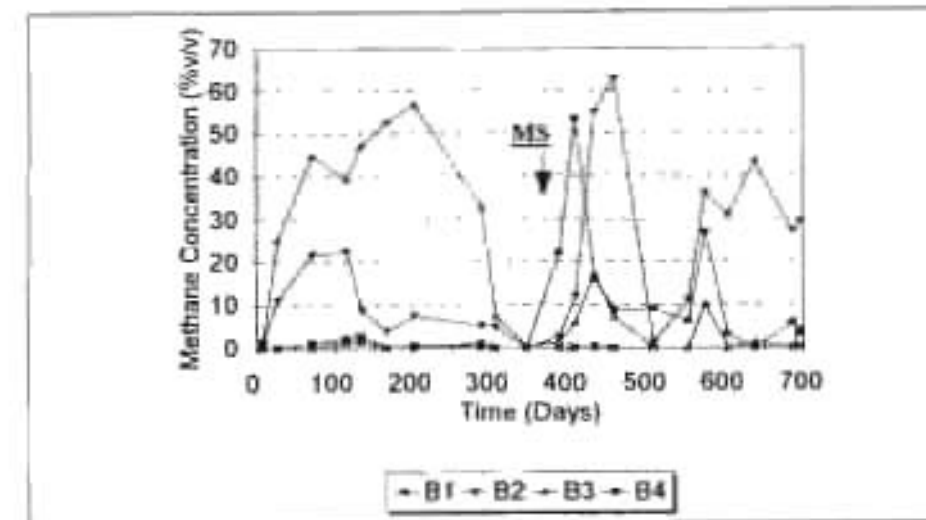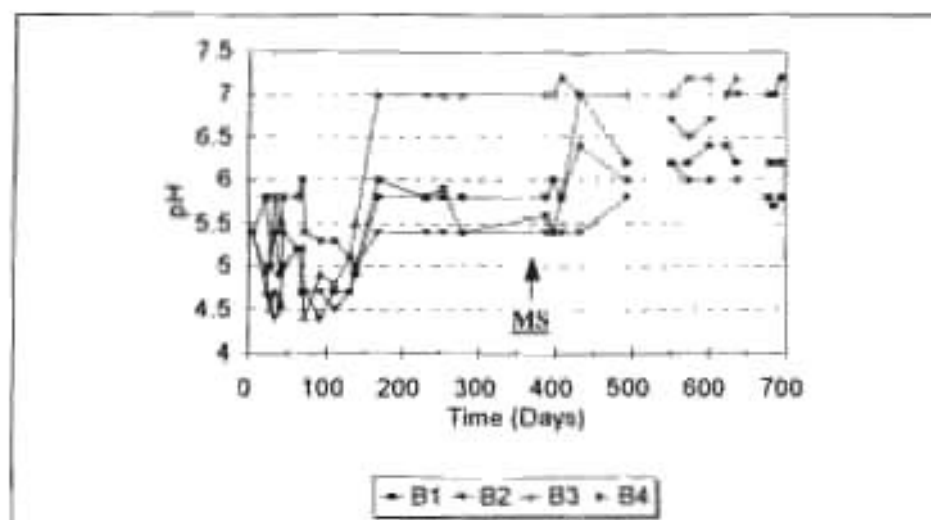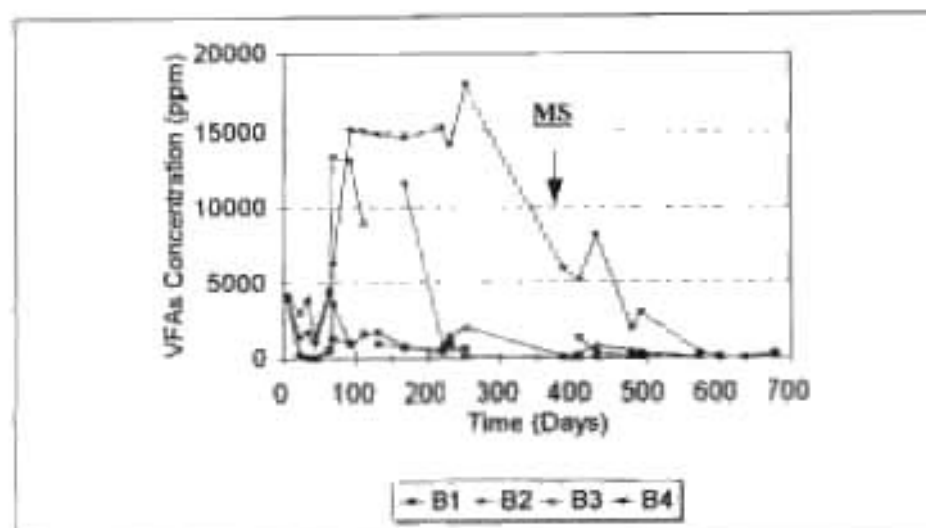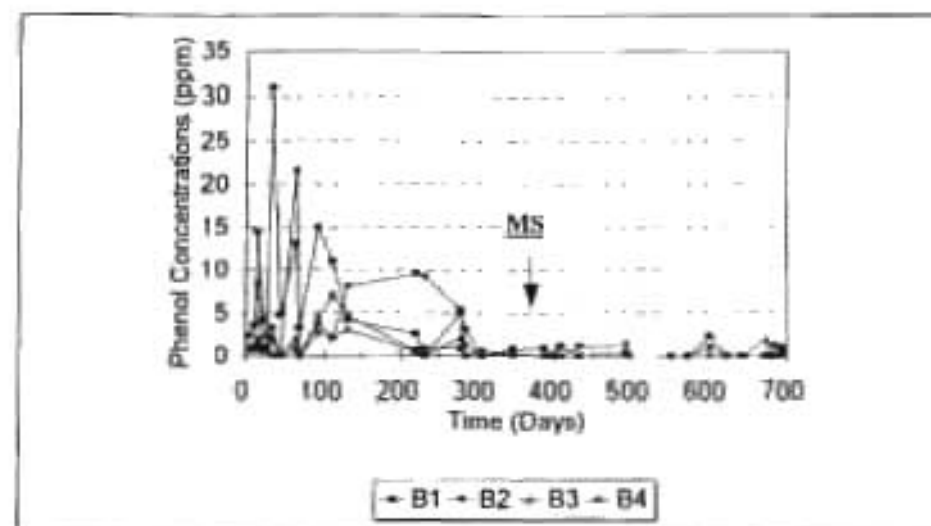
the other hand, the single elution and simulated rain operated columns were characterised by low 'total' VFAs concentrations possibly as a result of the increased leaching inherent in the operating regimes.

The negative influences that the single elution and simulated rain regimes had on methanogenesis are clear from Figure 4.8 D although brief increases were recorded following the mineral salts additions. The enhanced methane concentrations as a result of leachate recycle, and especially batch operation, were clear although the reason for the low methane concentrations of the latter, following the mineral salts addition, remains to be resolved.

4.3.4   Conclusions

The results of the sludge co-disposal study in the presence of a single elution regime indicated the importance of the sludge:refuse ratio. For example, the "high load" column exhibited reduced pH values whereas the pH values of the "low load" column leachate did not differ markedly from the refuse control. Therefore, the alkalinity present, as a result of the sludge additions to Columns A1 and B1, was insufficient to act as a buffer against the high 'total VFAs concentrations recorded. Similarly, the methane concentrations of the "high load" column, despite the lower pH values, were enhanced while the "low load" column concentrations were depressed. Another difference between the two loadings was the increased 'total' ammonia leached from the "high load" column in comparison with the "low load" column which, in turn, was lower than the refuse control. The low pH values combined with higher 'total' VFAs concentrations of the "high load" columns suggested that refuse masses subjected to single elution regimes and "high" sludge loads could be inhibitory although methanogenesis could be slightly enhanced.

The leachate recycle columns, similarly, demonstrated different effects depending on the loading rate. The "high load" column was characterised by enhanced refuse catabolism as indicated by the increased pH values and methane concentrations which coincided with lower 'total' VFAs concentrations. The "low load" column, on the other

hand, did not appear to benefit from the sludge addition as the pH values and methane concentrations were reduced. Following the mineral salts additions the co-disposal columns showed markedly enhanced methane concentrations. It is also interesting to note that the leachate ammonia concentrations, for both sludge loadings, were lower than the refuse control. It may be concluded that, unlike the single elution column, the "high" sludge load tended to enhance refuse catabolism in the presence of leachate recycle.

The two sludge loadings of the batch operated columns exerted similar effects on the refuse fermentation. For example, both the "high" and "low" load columns initially showed lower pH values than the refuse control. Also during the first 62 days the co-disposal columns recorded increased 'total' VFAs concentrations which subsequently decreased below the refuse control concentrations. The onset of methanogenesis was shortened for both the co-disposal columns. For optimization of refuse catabolism, the "low load" regime with leachate recycle should be the method of choice. However, for maximum methane generation the "high" sludge load appeared to be a better alternative.

Taking into consideration the fermentation parameters of pH, VFAs release and methane concentrations, the addition of sludge to refuse in the presence of simulated rain did not appear to significantly affect the fermentation.

The movement of liquid through the single elution and simulated rain "low load" columns did, in effect, decrease the 'total' VFAs concentrations, the pH values and the methane concentrations. Leachate recycle and batch operation, on the other hand, gave shorter methanogenesis lag phases and pH recovery times. It must, however, be noted that for these parameters the batch operated column showed enhanced refuse degradation compared to the leachate recycle column.

For the "high load" columns the single elution and simulated rain regimes inhibited refuse mineralisation as indicated by the continuously low pH values and methane concentrations. The leaching of fatty acids was also prevalent. In this instance, unlike the "low" load columns, the leachate recycle column gave a more rapid attainment of a neutral

pH and enhanced methane concentrations which coincided with reduced 'total' VFAs concentrations. It can, thus, be concluded that, depending on the sludge:refuse ratio, the operating regime is extremely important in optimising refuse degradation processes. In general, leachate recycle appeared to be the most favoured method of operation. This is in agreement with Boari, Mancini and Spinosa (1989) who reported that studies in Germany showed that the time required before stable methanogenesis commenced doubled in the absence of recirculation.

From the study made by Sinclair (1994) it was concluded that the optimum stage for sludge addition to landfilled refuse, to minimize the polluting strength of the leachate, was at a point when the leachate fatty acid concentrations were beginning to decrease. At this stage there should be sufficient available carbon to allow microbial uptake/conversion of any NH_4^+/NH_3 generated from the sludge, whilst the fatty acids generated from the sludge should not significantly lower the pH of the microenvironment to cause a "souring" of the fermentation. However, due to modern landfilling techniques, where sludge is most often poured into pre-excavated trenches, it is difficult to envisage how it would be possible to limit sludge disposal to refuse to a stage of the fermentation. The alternative technique of creating individual cells could possibly facilitate this co-disposal method although, at present, few landfills in South Africa, are operated in this mode.

Numerous researchers have indicated, although not investigated in the present study, that a further benefit of sludge co-disposal is the reduction of leachate heavy metal concentrations (Hill, 1989; Beker and van den Berg, 1989; Blakey, 1991). Also, Lichtensteiger, Brunner and Langmeier, (1989) reported that a few months after disposal, the Kjedahl nitrogen content of co-disposed liquid sludge decreased by about 50% while after 13 years significant concentrations of xenobiotics such as polychlorinated biphenyls and alkylbenzenesulphonates could be determined. This serves to illustrate that xenobiotics which are often given little attention, due probably to low concentrations, could possibly constitute a long term problem.

CHAPTER 5

ASSESSMENT OF THE IMPACTS OF THE DUAL CO-DISPOSAL OF ANAEROBICALLY DIGESTED SEWAGE SLUDGE AND PHENOL WITH REFUSE ON REFUSE CATABOLISM

5.1   Introduction

Sewage sludge, which has usually undergone various degrees of treatment, has often been used as a source of inocula for numerous microbial studies (Ferry and Wolfe, 1976; Mountfort and Bryant, 1982; Grbić-Galić, 1985; Flyvbjerg, Jorgensen, Arvin, Jensen and Olsen, 1993). Therefore, the dual co-disposal of sewage sludge together with phenol could, potentially, alleviate some of the problems inherent in the disposal of both these wastes. Thus, the objective of this component of the research programme was to examine the effects of dual co-disposal of sewage sludge and phenol with refuse on refuse catabolism.

5.2   Experimental, Results and Discussion

5.2.1 Microcosm Operation

In the dual co-disposal experiments the two sludge:refuse ratios used were: a. Ratios of 1:4.5 which were designated "high load" columns; and b. Ratios of 1:9 which were designated "low load" columns (Section 3.2). Together with the two sludge ratios two phenol concentrations (1000 and 2000 mg l^{-1}) were co-disposed. The columns were subjected to mineral salts additions as described in Section 3.2.1. Further, phenol resupplementations were made with the first phenol resupplementation (RS1) concentration equal to the initial phenol perturbation concentration. This resupplementation was made when the residual phenol concentrations of the leachate recycle and batch columns had approached zero. The second resupplementations (RS2) which were double the initial phenol concentration were also made when the residual phenol concentrations approached

zero. Therefore, the single elution columns, which were continuously perfused, were only subjected to an increased phenol concentration during the second phenol resupplementation. The simulated rain columns, due to operational difficulties, were not subjected to the first phenol resupplementation (RS1).

Due to the number of variables examined in this study, as well as the number of columns used, the results and discussion are structured as follows: a. Firstly, sewage sludge, "high" and "low" loadings, together with either 1000 or 2000 mg l^{-1} phenol co-disposal with refuse are considered in relation to the microcosm mode of operation (single elution, leachate recycle, batch and simulated rain); b. Subsequently, the effects of the perfusion strategy (water, 1000 and 2000 mg l^{-1} phenol) for both the "high" and "low" loadings are considered.

5.2.2 "High Load" and "Low Load" Anaerobically Digested Sewage Sludge and 1000 mg l^{-1} Phenol Dual Co-Disposal with Refuse

Single Elution Columns

The changes in leachate concentrations of the residual phenol, pH, 'total' VFAs concentrations and headspace methane concentrations for the "high load" column (A5), "low load" column (B5), phenol co-disposal column (C5) and refuse control column (C1) are shown in Figures 5.1 A - 5.1 D, respectively.

During the first 70 days the sewage sludge and phenol co-disposal columns (A5 and B5) exhibited marked increases in leachate phenol concentrations of between 400 and 900 mg l^{-1}, compared with the phenol co-disposal column (C5) where phenol concentrations of approximately 270 mg l^{-1} were recorded (Figure 5.1 A). The increases in the residual phenol concentrations could have been due to inherently higher phenol concentrations as a result of the sludge addition or higher degradation rates of phenolic compounds. Unfortunately, the former could not be verified as no sludge only controls were used while the latter could have resulted from increased microbial activity. Alternatively, the addition

Figure 5.1   Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for "High Load" (A5), "Low Load" (B5), Phenol Co-Disposal (C5) and Refuse Only Control (C1) Columns Operated with Single Elution During an Incubation Period of 695 Days
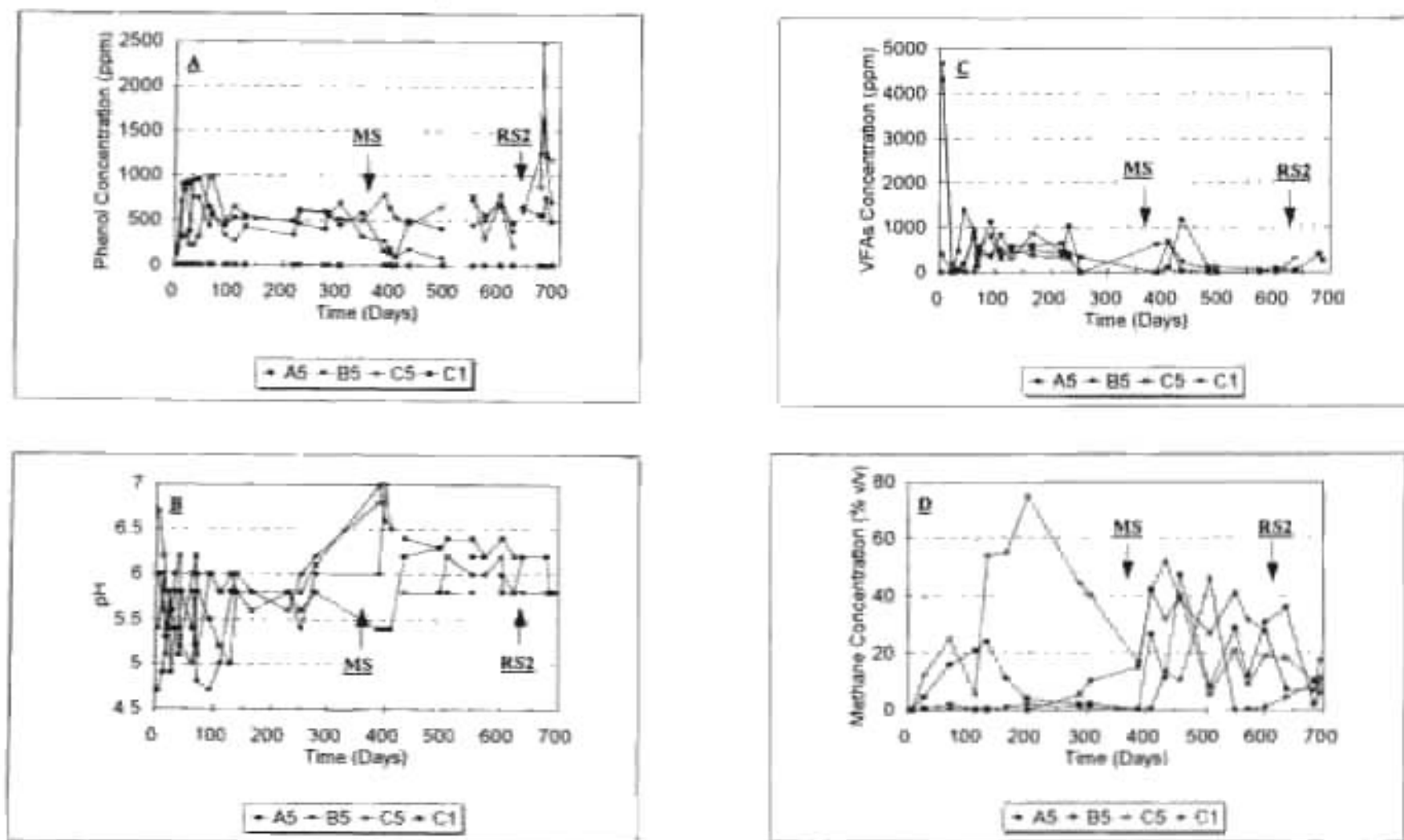
MS Designates Mineral Salts Addition

RS2 Designates Phenol Concentration Increase (2500ppm)

of sewage sludge could have limited the adsorption of the phenol to the refuse which could also explain the rapid breakthrough of the phenol. Following this period of relatively high residual phenol concentrations, all the columns exhibited decreases and stabilised around 500 mg l^{-1}. After the addition of the mineral salts, however, the sewage containing columns (A5 and B5) recorded further decreases in the residual phenol concentrations while the phenol co-disposal column C5 exhibited a slight increase. These decreases, in Columns A5 and B5, were not maintained and by day 551 all the columns again indicated leachate concentrations of approximately 500 mg l^{-1}. The "high load" (A5), "low load" (B5) and phenol co-disposal (C5) columns averaged phenol removal rates of 0.297, 0.31 and 0.29g kg^{-1} d^{-1}, respectively. Following the influent phenol concentration increase (2000 mg l^{-1}), the leachate residual phenol concentrations of the "high load" column (A5) did not appear to increase significantly while the equivalent "low load" column recorded sharp increases. Subsequently, however, these high concentrations decreased to the pre-increase concentrations. The phenol co-disposal column, following the influent concentration increase, exhibited similar rapid breakthroughs of phenol with, subsequent, decreases. From the results, prior to the influent concentration increase, it appeared that the dual co-disposal of sewage sludge and phenol with refuse did not effect a marked increase in phenol catabolism compared with the phenol co-disposal column. Following the concentration increase increases in the phenol removal rates were recorded for the "high load", "low load" and phenol co-disposal columns, to 0.73, 0.46 and 0.59g kg^{-1} d^{-1}, respectively. From these results, however, the positive effect of sludge addition was evident for the "high load" column but not the "low load" column since the latter exhibited a lower removal rate than the phenol co-disposal column.

The sludge-amended columns (A5 and B5), in general, were initially (<17 days) characterised by lower pH values compared with the phenol co-disposal column (C5). By day 150, however, all the column leachates exhibited comparable pH values of approximately 5.7. Following the mineral salts additions the pH values of Columns B5, C5 and C1 approached or attained neutrality. These elevated pH values were, however, not maintained and subsequently decreased. The influent phenol concentration increase, in general, resulted in slight decreases in the pH values. It is interesting to note that the refuse

control. Column C1, which was not challenged by phenol, demonstrated similar reduced pH values as the co-disposal columns. This seemed to suggest that the co-disposal *per se* of sludge and/or phenol was not responsible for the lowered pH values. Therefore, the operating regime was the major determinant of pH. Also, one possible reason for the increased phenol removal rates, prior to the concentration increase, of the "low load" column (B5) compared with the "high load" column (A5) could have been due to the higher pH values immediately prior to and following the mineral salts addition. The critical role of pH on phenol catabolism has been discussed previously (Section 3.3.2).

From Figure 5.1 C it can be seen that the continuously low pH values, prior to the mineral salts addition, could not have been due to 'total' VFAs accumulations. Thus, the transient increases in pH values described above, following the additions, were most likely as a result of the buffering compounds added with the mineral salts. Also, the low 'total' VFAs concentrations were most probably as a result of the leaching effect of the perfusion regime rather than low rates of acidogenesis. The lower pH values recorded for the sludge amended columns, during the first 20 days, could have been due to the higher 'total' VFAs concentrations. These results were in agreement with the VFA release trends reported by Sinclair (1994) who concluded that the added sewage, in refuse columns, was responsible for the major contribution to the 'total' VFAs concentration. After the influent concentration increase there was a slight accumulation of 'total' VFAs in the co-disposal columns which seemed to suggest a measure of microbial inhibition. The inhibiting effect of phenol perfusion can result in VFA accumulation. For example, Satsangee and Ghosh (1990) reported that an anaerobic microbial association required 24 hours to metabolise a 2% (w/v) glucose solution as a carbon source although when 100 mg l^{-1} phenol were added to a similar glucose solution the microorganisms required 48 hours and there was a large accumulation of volatile fatty acids.

The methane concentrations recorded for these columns during the study are given in Figure 5.1 D. The "high load" column (A5) showed progressive increases in methane concentrations to 24% (v/v) on day 137 with subsequent decreases. The reasons for the subsequent decreases are not clear although labile substrate limitation could be implicated.

Notwithstanding the erratic nature of the methane concentrations of Column A5, following the mineral salts additions, it can be seen that the influent concentration increase resulted in decreases in the methane concentrations to 6.1 % (v/v) at the termination of the study. However, it must be pointed out that at this stage the refuse (C1) and the phenol co-disposal (C5) columns recorded concentrations of 8.7 and 11.3% (v/v), respectively. The "low load" column (B5) exhibited suppressed methane concentrations until the mineral salts addition following which the methane concentrations increased although they were erratic in nature. One possible explanation for the sharp decreases in methane concentrations of Column B5 after 547 days could have been the subsequent declines in pH values following the mineral salts addition. More interesting, however, is the observation that subsequently, and especially following the influent concentration increase, the methane concentrations again increased. This could possibly have been due to microbial adaption to the co-disposed molecule or, alternatively, the establishment of favourable environmental conditions such as redox potential. Circumstantial evidence of this was the accumulation of nitrite (168 mg l^{-1}) in Column B5 prior to the mineral salts addition with the disappearance of this compound following the addition. Therefore, it seemed that, prior to the addition, the reduction of nitrate to the terminal compound nitrogen gas was limited. The reduction of nitrate, as well as sulphate, facilitates the establishment of reduced conditions. The negative effect of the dual co-disposal of sewage sludge and phenol compared to the phenol co-disposal column (C5) was clear from the latter's enhanced methane concentrations prior to the mineral salts additions. However, the sludge supplemented columns, and especially Column A5, recorded higher methane concentrations following the salts additions.

The fermentation trends determined during this study seemed to indicate that, compared with the refuse control column (C1), negligible microbial inhibition resulted, when using pH and 'total VFAs concentrations as the analytical criteria, from the dual co-disposal of sludge and phenol. Also, microbial activity, and especially methanogenesis, was enhanced with high sludge loadings.

Leachate Recycle Columns

The leachate, phenol concentrations, pH values, 'total' VFAs concentrations and the methane concentrations of the leachate recycle columns (A6, B6, C6, C2) are shown in Figures 5.2 A - 5.2 D.

During the first 40 days the sludge amended columns (A6 and B6) exhibited residual phenol concentrations of between 200 and 870 mg l^{-1} compared with <200 mg l^{-1} for the phenol co-disposal column (C6). This phenomenon of increased breakthrough of phenol could be ascribed to lower adsorption rates for phenol as a result of sewage sludge addition which could have altered the various refuse surfaces. Sinclair (1994), for example, reported that following an experimental period of 300 days similar co-disposal columns were dismantled and the columns which had contained sewage sludge were more slimy in nature than the refuse controls.

Following this initial stage of high residual phenol concentrations, the "high load" column (A6) was characterised by increased phenol catabolism and by day 112 displayed very low concentrations. Average phenol removal rates of 0.006 and 0.033g kg^{-1} d^{-1} were recorded for the pre- and post-resupplementation (RS1 and RS2) periods, respectively. The equivalent "low load" column (B6), on the other hand, required 387 days to attain similar low phenol concentrations and, likewise, recorded increases in the phenol removal rate from 0.0016 to 0.023g kg^{-1} d^{-1} following the resupplementations (RS1 and RS2). During the first 44 days the residual phenol concentrations of Column C5 were lower than both the sludge amended columns although 227 days were required to attain concentrations approaching zero. Average phenol removal rates of 0.005 and 0.06g kg^{-1} d^{-1} were recorded for the pre- and post-resupplementation (RS2) periods, respectively. From these results it appeared that the addition of sludge at a refuse:sludge ratio of 4.5:1 increased the rate of phenol catabolism and that the equivalent "low load" column did not effect the same promotion. These results are in agreement with those of Stegmann and Spendlin (1989) who concluded that sewage sludge co-disposal in low ratios did not supply sufficient inocula to enhance biochemical processes. The resupplementation (RS1) with 1000 mg l^{-1}

Figure 5.2 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for "High Load" (A6), "Low Load" (B6), Phenol Co-Disposal (C6) and Refuse Only Control (C2) Columns Operated with Leachate Recycle During an Incubation Period of 695 Days
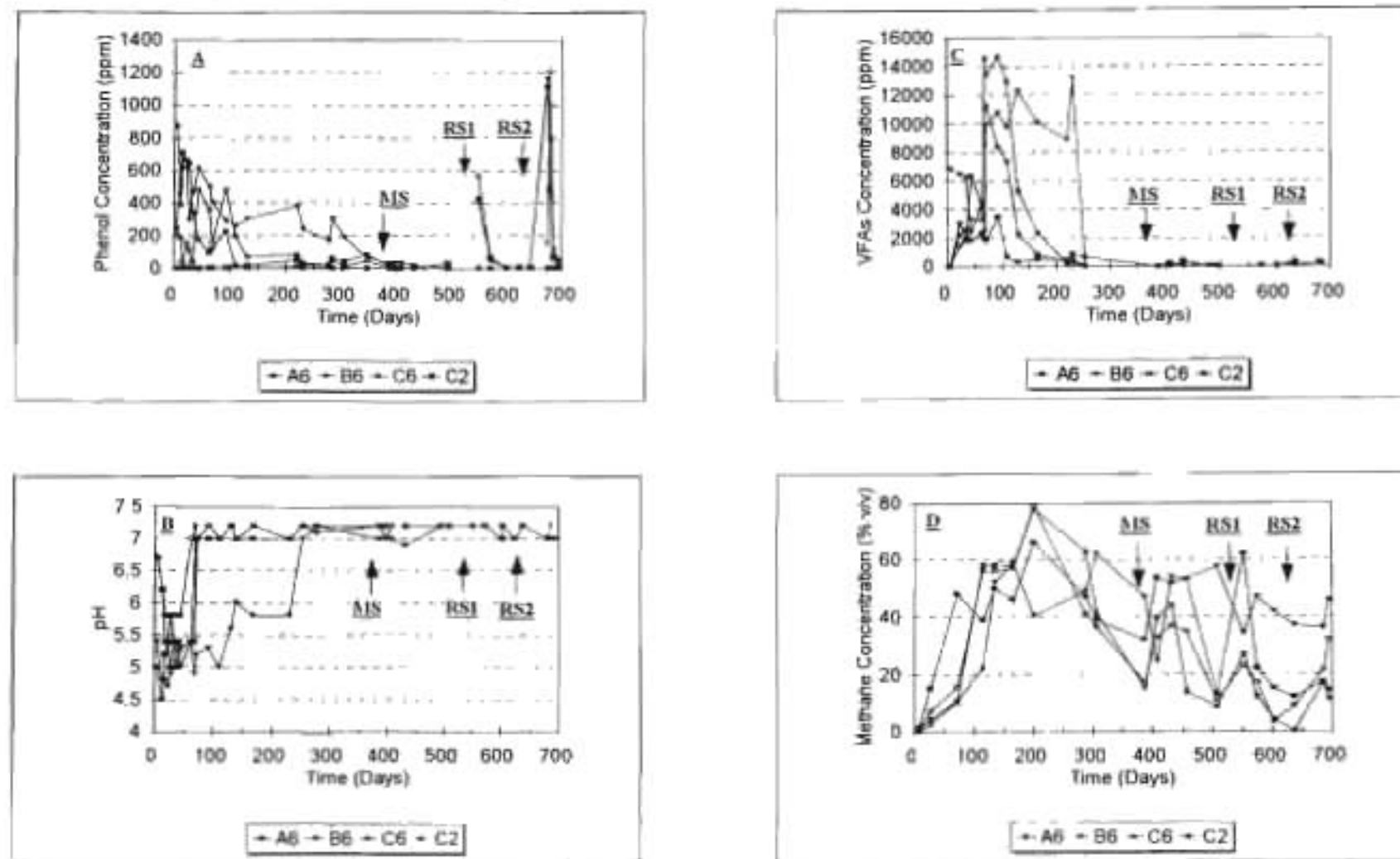
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (1000ppm)

RS2 Designates Phenol Resupplementation (2000ppm)

phenol effected a rapid breakthrough of phenol in both the sludge amended and phenol co-disposal columns but was followed by rapid decreases. After the resupplementations, phenol catabolism and, thus, phenol removal rates, increased possibly due to acclimation of the microorganisms to the added phenol. A similar pattern of increased removal rates was reported by Watson-Craik and Senior (1989a) who found that following five individual phenol resupplementations of co-disposal columns the rate increased with each. The authors concluded that the possible continued phenol selection pressure, together with the reintroduction of displaced microbial species and essential metabolites, due to leachate recycling, resulted in the selective enrichment and subsequent uniform distribution of phenol-catabolising populations with resultant improved bioreactor efficiencies. Also, in anaerobic digestion studies, without leachate recycle, Satsangee and Ghosh (1990) concluded that an increased time of acclimation of the mixed microbial population significantly increased the phenol degradation rate.

After the second phenol resupplementation (2000 mg l^{-1}) similar rapid breakthroughs and subsequent declines were recorded in Columns A6, B6 and C6. These removals most probably resulted from phenol catabolism although physico-chemical factors could also have influenced the retention of phenol. Retention may be influenced by a range of factors (Artiola-Fortuny and Fuller, 1982; McBride 1994) of which only surface area and pH were likely to have changed during the course of the study. However, with both phenol resupplementations (RS1 and RS2) the time from resupplementation to the attainment of very low phenol concentrations was short and it was, therefore, expected that the surface area would not have changed significantly. Further, as indicated in Figure 5.2 B, following the phenol resupplementations the pH values did not change sufficiently to effect such a dramatic difference in phenol degradation. A similar conclusion was reached by Watson-Craik and Senior (1989a).

Despite the initial declines in pH values for all the columns the pH values (Figure 5.2 B) of the sewage sludge amended columns (A6 and B6) were, during the first 14 days, lower than the phenol co-disposal (C6) and refuse (C2) columns. By day 68 only the "low load" column (B6), which required 249 days, had not attained a neutral pH. Furthermore,

it should be noted that despite the phenol resupplementations all the columns maintained this status.

As reported previously (Section 4.3.2) the VFA releases during the first few days (< 20 days) were the highest in the sludge amended columns and could have been due to the higher concentrations added in the sludge. Also, the "low load" column (B6) recorded protracted elevated 'total' VFAs concentrations compared to the phenol co-disposal (C6) and refuse (C2) columns. Noteworthy, however, was the occurrence of low 'total' VFAs concentrations for the "high load" column which, unlike the other columns, did not show a subsequent rise in 'total' VFAs concentrations after approximately 71 days. The longevity of the more acidic conditions in the "low load" column (B6) can, therefore, be explained, in part, by the increased 'total' VFAs concentrations of this column until day 227. Subsequently, significant concentration reductions were recorded which coincided with steady increases in the pH values. Despite the phenol resupplementations (RS1 and RS2) there was no resulting accumulation of the 'total' VFAs.

The highest initial (< 100 days) methane concentrations were recorded with the "high load" column although by day 140 all the columns had attained concentrations >50% (v/v). Watson-Craik and Senior (1989a,c) reported that the perfusion of a refuse column with 376 mg l^{-1} phenol resulted in headspace methane concentrations of 0.17% (v/v) and an accumulation of acetate (870 mg l^{-1}). It was, therefore, suggested that the methanogens were more susceptible than the acetogens to the inhibitory effects of phenol. In this present study, however, the addition of 1000 mg l^{-1} phenol did not effect a similar methanogenic inhibition or 'total' VFAs accumulation in the long term. The methane concentrations for all the columns following the mineral salts additions as well as the phenol resupplementations were erratic in nature. However, in general, the sludge amended columns, and especially the "low load" column, recorded greater methane concentrations than both the phenol co-disposal and refuse columns. These results seemed to indicate the possible beneficial effects of adding additional methanogens with the sludge.

It can, therefore, be concluded that the initial perfusion of the columns with 1000 mg l^{-1}, in the early stages, resulted in slightly decreased pH values for the "low load" column with a concomitant transient accumulation of 'total' VFAs. In general, however, the leachate recycle operated columns, with dual co-disposal of sewage sludge and phenol, did not, despite the phenol resupplementations, show any protracted inhibition when pH, 'total' VFAs and methane concentrations were the analytical criteria. Finally, the "high load" operated column exhibited not only enhanced phenol catabolism but also increased refuse fermentation and methane generation.

Batch Operated Columns

The residual phenol concentrations, pH, 'total' VFAs concentrations and methane concentrations for the batch operated "high load" (A7), "low load" (B7), phenol co-disposal (C7) and refuse (C3) columns are given in Figures 5.3 A - 5.3 D.

As recorded for the leachate recycle columns the batch operated columns, amended with sewage sludge (A7 and B7), initially (< 64 days), displayed higher residual phenol concentrations compared with the phenol co-disposal and refuse columns (Figure 5.3 A). This more rapid breakthrough of phenol could have been due to a decreased adsorption capacity as described above. The "low load" column (B7) averaged phenol removal rates of 0.0014 and 0.023g kg^{-1} d^{-1} pre- and post-phenol resupplementations (RS1 and RS2), respectively. Therefore, the "low load" column exhibited slightly higher leachate phenol concentrations than the equivalent "high load" column which maintained phenol removal rates of 0.0012 prior to and 0.046g kg^{-1} d^{-1} following the phenol resupplementations (RS1 and RS2). The phenol co-disposal column (C7) recorded phenol removal rates of 0.0012 and 0.05g kg^{-1} d^{-1} pre- and post-phenol resupplementations (RS1 and RS2), respectively. Therefore, despite the higher residual phenol concentrations recorded for the sludge supplemented columns (A7 and B7) the three experimental columns exhibited remarkably similar phenol removal rates. The additions of the mineral salts clearly had a positive effect on phenol catabolism as, following the additions, the three experimental columns revealed steady decreases (Figure 5.3 A). The phenol resupplementations resulted in rapid

Figure 5.3 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for "High Load" (A7), "Low Load" (B7), Phenol Co-Disposal (C7) and Refuse Only Control (C3) Columns Operated in Batch Mode During an Incubation Period of 695 Days
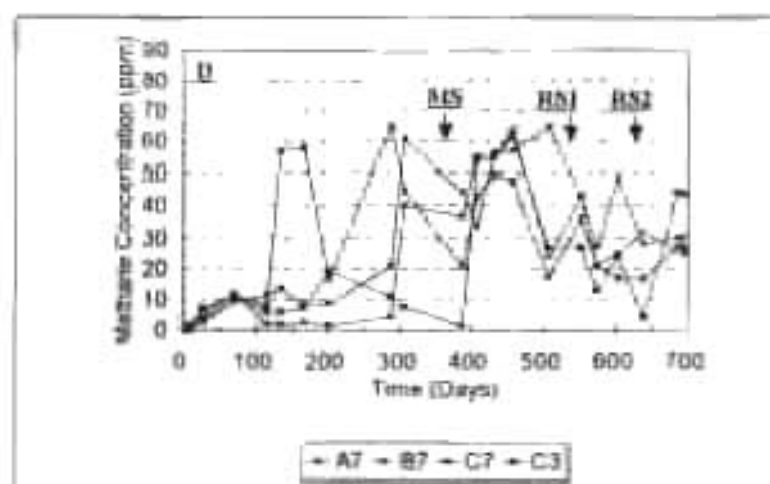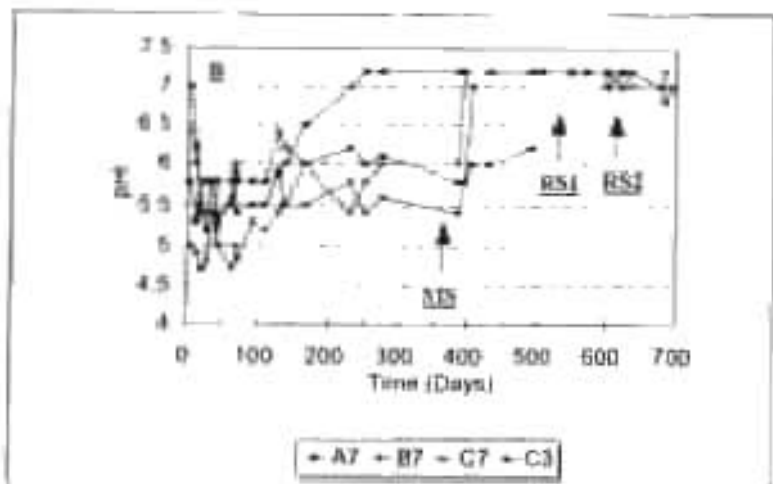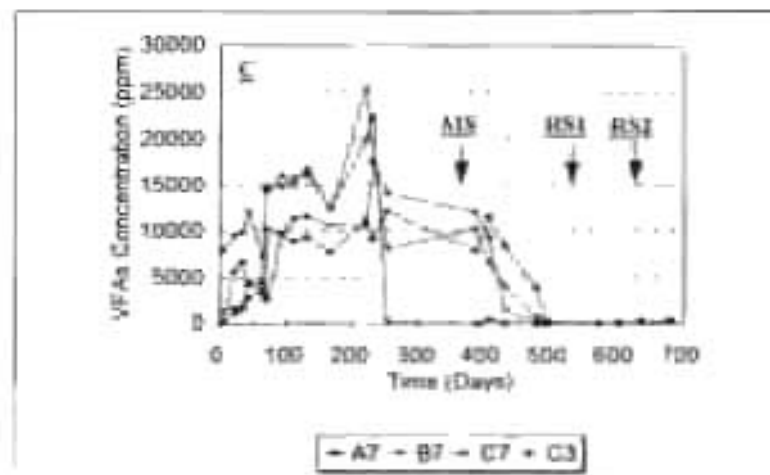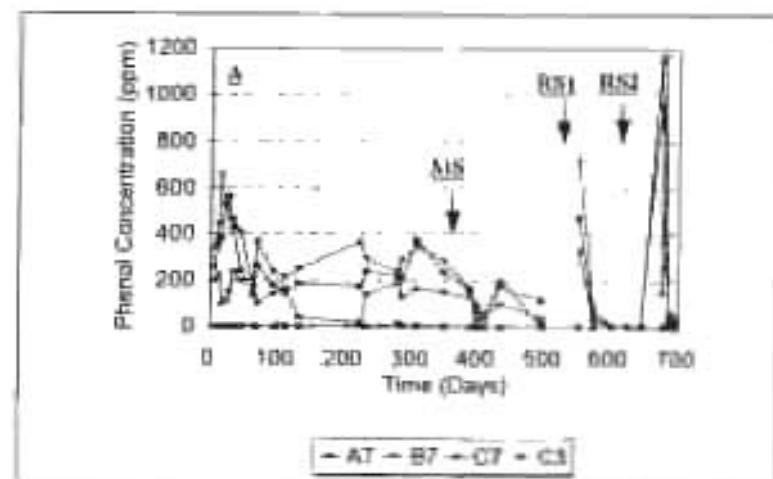
breakthroughs of the added molecule although subsequent rapid decreases were recorded. Further, following the phenol resupplementations all the columns, irrespective of the co-disposal regime, demonstrated phenol removal rates of the same order.

The refuse control column (C3) showed the characteristic decline in pH during the first 120 days (Figure 5.3 B) after which there was a steady increase to a neutral pH which was reached on day 229. As with the leachate recycle columns the sludge amended columns, and especially the "low load" column (B7), exhibited lower initial (<19 days) pH values than the equivalent sludge free columns. The phenol (C7) and sludge amended (A7 and B7) co-disposal columns were characterised by lowered pH values until the addition of the mineral salts. Subsequently, the pH values increased although Column A7 displayed a slower recovery to a neutral pH.

One possible explanation for the increases in the pH values, following the salts addition, of the co-disposal columns could have been the rapid decline in the 'total' VFAs concentrations (Figure 5.3 C). It is worth noting that as with the leachate recycle operated columns, the "high load" column (A7) recorded lower initial (<74 days) 'total' VFAs concentrations compared with the "low load" column (B7). However, during this early period, these two columns recorded higher concentrations than the equivalent phenol co-disposal and refuse columns. Subsequently, however, the sewage amended columns exhibited lower 'total' VFAs concentrations until day 227 when the refuse control column showed a dramatic decrease. No discernible accumulation of 'total' VFAs resulted following either of the phenol resupplementations.

From Figure 5.3 D it is clear that there were no distinct differences between the methane concentrations of all the columns until day 118 when the methane concentrations of the refuse control column sharply increased. Surprisingly, the "low load" column (B7) attained a methane concentration of 61% (v/v) on day 309 while the equivalent "high load" column recorded a concentration of 8.1% (v/v) and only increased to a maximum of 61%

(v/v) after the mineral salts addition. Following the mineral salts additions there were general increases in the methane concentrations of all the columns which were subsequently followed by concentration decreases in response to the phenol resupplementations. However, it is interesting that, with a few exceptions, the experimental columns, and especially the "low load" column, exhibited methane concentrations greater than the refuse control column.

From these results it appeared that the dual co-disposal of sludge and phenol did initially inhibit the fermentation of refuse as indicated by the protracted periods of low pH values and increased 'total' VFAs concentrations, as well as the extended lag phase to the onset of active methanogenesis. However, the inhibitory effect of the dual co-disposal was lower than that exerted by the phenol co-disposal column. Also, the dual co-disposal of sludge and phenol did not effect increases in the leachate phenol concentrations or refuse catabolism compared with the phenol co-disposal column. It was also apparent that the "low load" column exhibited higher pH values and methane concentrations than the "high load" column. The dramatic effects that the mineral salts additions had on the parameters discussed above were also evident in the transient accumulations of the electron acceptors nitrate and sulphate. For example, prior to the mineral salts additions, the "high load" column recorded nitrate and sulphate concentrations of 186 and 31.8 mg l^{-1}, respectively, while following the additions the concentrations decreased to below detection limits. Also of interest was the accumulation of nitrite (135 mg l^{-1}), of the "high load" column, before the mineral salts addition which, similarly, declined to an extremely low value following the addition. Unlike the refuse control column (C3), similar accumulations of nitrate, sulphate and nitrite were recorded for the "low load" (B7) and phenol co-disposal (C7) columns. These results seemed to indicate that the practice of either phenol co-disposal or the dual co-disposal of phenol and sewage sludge, operated in batch mode, negatively affected the interacting microbial associations particularly the nitrate and sulphate-reducing bacteria. According to Senior (1991), the practice of co-disposal is a superimposition on the refuse catabolic processes. Therefore, the dual co-disposal of sewage sludge and phenol with refuse could intensify the negative impacts of this practice. It is, thus, encouraging to note that these results indicated that the dual co-disposal did not effect increases in

microbial inhibition when compared to phenol co-disposal. In fact, the "low load" column showed that enhanced refuse catabolism was coincident with the dual co-disposal of phenol and sludge.

Simulated Rain Columns

The leachate residual phenol concentrations recorded for the rain simulated columns (Figure 5.4 A) showed rapid breakthroughs and subsequent declines to very low concentrations by day 75. Without radio-labelled phenol it was impossible to determine the degree of phenol catabolism compared with the amount of phenol leached from the refuse/sludge. In this instance, however, it is believed that the rapid declines in phenol concentrations were due to the washout effect of the moving liquid rather than phenol catabolism. For example, the start up procedures (Section 3.2) resulted in the addition of 0.7g phenol to the experimental columns, after which the columns were perfused at a rate of 180ml of distilled water per week. Taking into consideration the leachate phenol concentrations it was calculated, for Column A8, that 0.578g or 82.5% of the phenol had been leached during the first 43 days of operation. As a result of operational difficulties these columns were not resupplemented with phenol (1000 mg l^{-1}) as were the other columns. However, resupplementation with 2000 mg l^{-1} phenol (RS2) was followed by rapid declines in the leachate phenol concentrations after initial breakthroughs. These were also thought to be mainly due to leaching and desorption of phenol.

The pH values, with one exception, were, prior to the mineral salts additions, suppressed. Following the additions Column C4 showed transient pH increases (Figure 5.4B) Similarly, the 'total' VFAs concentrations, subsequent to day 7, were low and further concentration reductions followed the mineral salts additions (Figure 5.4 C).

The methane concentrations (Figure 5.4 D), in general, infered low methanogenic activity prior to the mineral salts additions following which brief increases in concentrations were recorded for Columns A8, B8 and C4. These increases could have been due to the addition of methanogenic precursors such as carbonate. The increases in the

Figure 5.4 Changes in Residual Phenol Concentration (A), pH (B), Total VFAs Concentration (C) and Methane Concentrations (D) for "High Load" (A8), "Low Load" (B8), Phenol Co-Disposal (C8) and Refuse Only Control (C4) Columns Operated with Simulated Rain During an Incubation Period of 695 Days

MS Designates Mineral Salts Addition
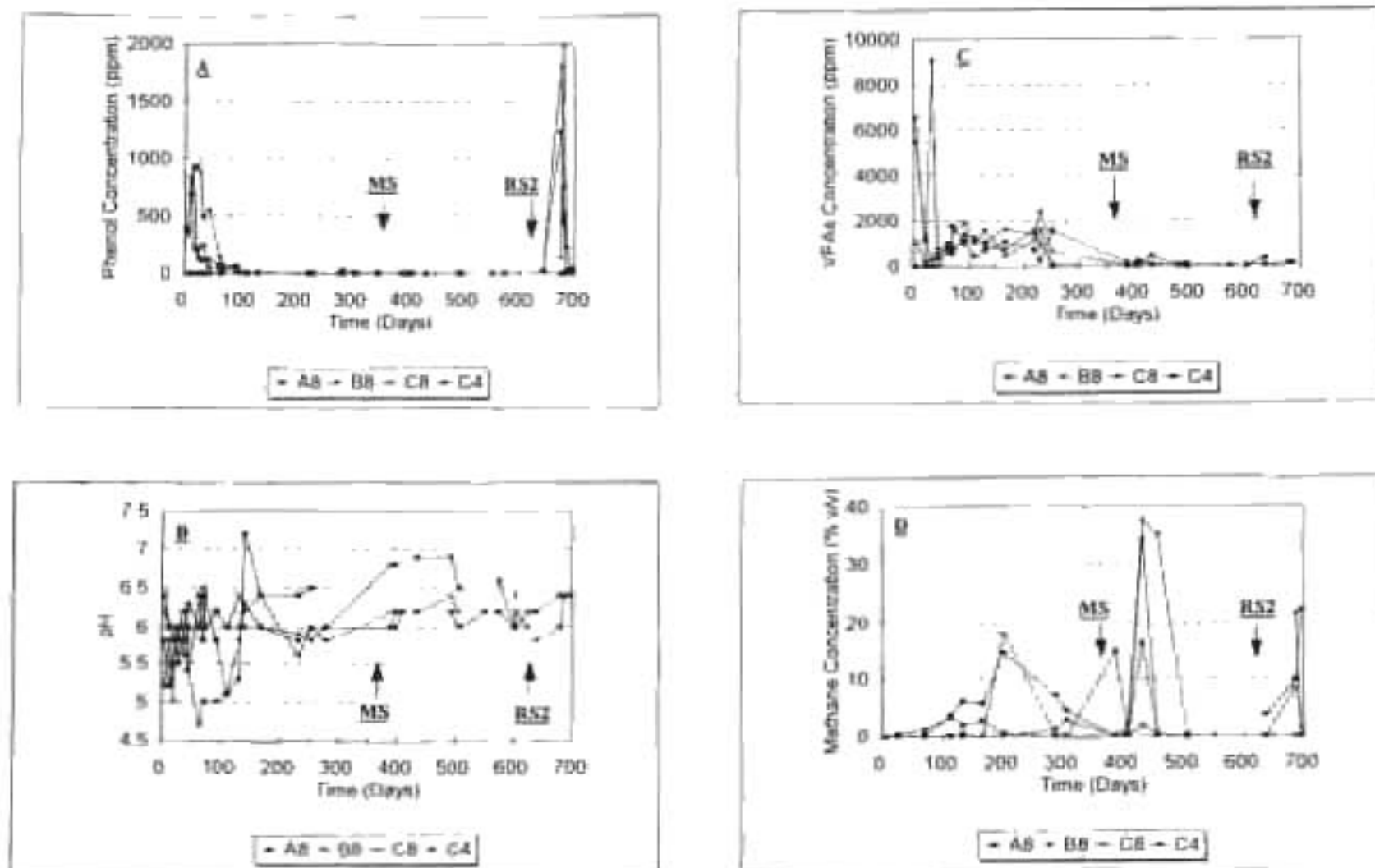
RS2 Designates Phenol Resupplementation (300ppm)

methane concentrations of the sludge amended columns following the second phenol resupplementations (RS2) were surprising. Why this should have had a stimulating effect on the methanogenic population was not clear although it was concomitant with slight increases in the pH values.

The recorded parameters such as the rapid washout of phenol, the low pH values, the consistently low 'total' VFAs concentrations and the inhibited methane generation seemed to indicate that very little refuse and phenol catabolism occurred and that desorption and subsequent washout of the phenol was the most likely outcome of this practice. This washout phenomenon was more pronounced in the rain simulated columns than the single elution columns which were continuously perfused with phenol. Also, the leachate recycle and batch operated columns were, in effect, closed systems and, therefore, restricted any washout.

5.2.3 Effects of Perfusion Strategy in the Dual Co-Disposal of Anaerobically Digested Sewage Sludge and 1000 mg l^{-1} Phenol with Refuse

"High Load" Columns

The effects that the operational regime had on the phenol concentrations, pH values, 'total' VFAs and methane concentrations, for the "high load" columns (A5-A8), are shown in Figures 5.5 A - 5.5 D.

Not surprisingly, the highest residual phenol concentrations (Figure 5.5 A) were recorded for the single elution column (A5) as this column was continuously perfused with 1000 mg l^{-1} phenol. The phenol breakthrough was relatively rapid with 897 mg l^{-1} phenol recorded on day 15. However, by day 110 the concentrations had stabilised between 400 and 600 mg l^{-1} until the addition of the mineral salts which resulted in temporary phenol concentration decreases. Surprisingly, Column A5 demonstrated similar leachate concentrations pre- and post the second (2000 mg l^{-1}) resupplementation. As discussed earlier (Section 3.3.2) both the organic loading rate and hydraulic loading are important

Figure 5.5 Changes in Residual Phenol Concentrations (A), pH (B), "Total" VFAs Concentrations (C) and Methane Concentrations (D) for the "High Load" Columns Operated with Single Elution (A5), Leachate Recycle (A6), Batch Mode (A7) and Simulated Rain (A8) During an Incubation Period of 695 Days
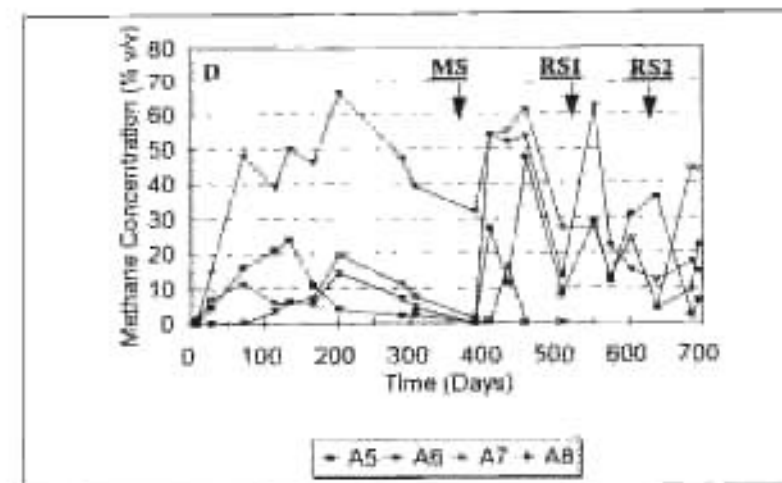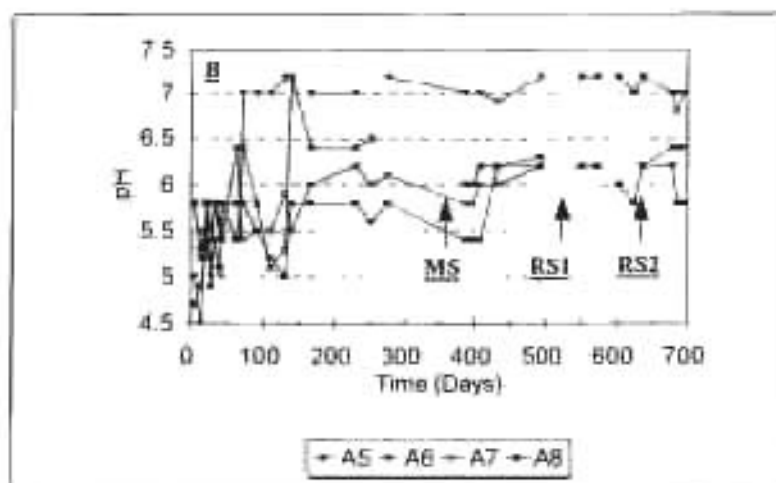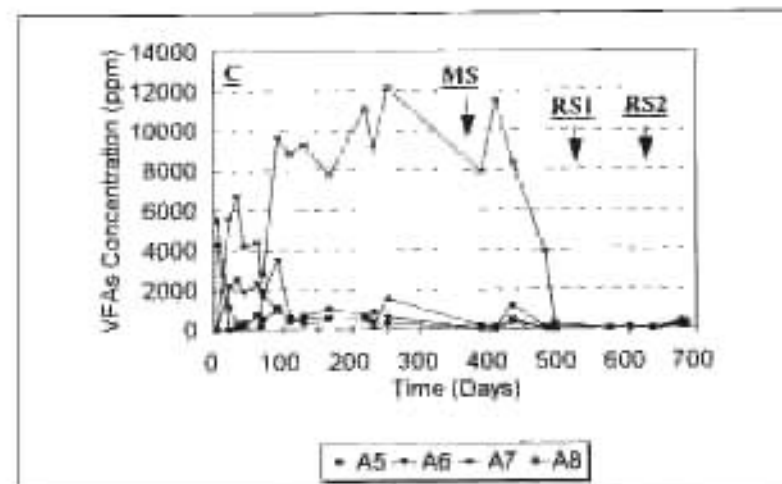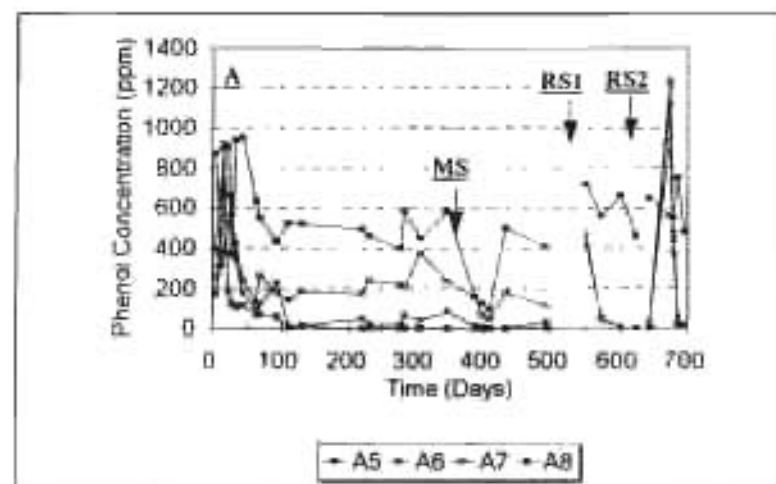
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (2000ppm)

RS2 Designates Phenol Resupplementation (4000ppm)

factors in phenol catabolism in refuse. For example, Watson-Craik (1987) demonstrated that phenol removal increased as the influent concentration increased up to approximately 1500 mg l^{-1}. This fact, together with possible microbial adaption and the elimination of possible elemental deficiencies could have accounted for the increased rate of phenol removal following the phenol resupplementations. For example, prior to the mineral salts addition Column A5 maintained an average phenol removal rate of 0.297g kg^{-1} d^{-1}, while, following the resupplementations (RS1 and RS2), the phenol removal rate increased to 0.73g kg^{-1} d^{-1}. Both the sludge additions and the increased influent phenol concentration, thus, appeared to effect mean removal rate increases, notwithstanding the initial breakthrough of phenol in the sludge amended columns which was, as discussed above, more rapid.

The leachate recycle column (A6), similarly, showed high initial breakthrough whereas, unlike the single elution column, substantial decreases to low concentrations of 10 mg l^{-1} were recorded on day 110, after which the leachate concentrations remained low. Despite the attainment of these low residual phenol concentrations, for Column A6, the average phenol removal rate was 0.006 g kg^{-1} d^{-1}. The possible reasons for these lower removal rates, compared to the single elution column (A5), could have been the lower rates of liquid movement. For example, Column A5 was subjected to continuous movement of the influent while the equivalent leachate recycle column (A6) was subjected to a liquid flux of 50-75ml per week. It is now widely accepted that fermentation promotion by water addition involves two separate variables: the water content *per se* and its movement through the refuse mass (Klink and Ham, 1982). Further, in effect, the leachate recycle column had to contend with an ever decreasing concentration of phenol while the single elution column was challenged by a constant concentration. A similar conclusion, with refuse columns, was reported by Watson-Craik and Senior (1989a). The authors indicated that the lowest hydraulic loading rate (7.2 ml h^{-1}) was characterised by a mean phenol attenuation rate of 0.02mg kg^{-1} d^{-1}, while an eight-fold increase in the loading rate affected an attenuation increase to 0.202 mg kg^{-1} d^{-1}.

The lack of liquid movement through the batch operated column (A7) appeared to,

compared with the leachate recycle column, result in higher leachate phenol concentrations which fluctuated around 200 mg l^{-1} from day 43 until the addition of the mineral salts. Notwithstanding this prolonged period of high phenol concentrations, the phenol attenuation rate increased significantly following the phenol resupplementations. The reasons for the enhanced phenol removal rate could have been microbial adaption and/or the elimination of any nutrient limitation by mineral salts addition. Alternatively, the phenol resupplementation could have resulted in a certain degree of liquid movement, due to a limited amount of recycling to ensure that the influent had permeated throughout the column, although the impact of this was expected to be minimal.

As discussed above, the rapid decline in residual phenol concentrations for the simulated rain column (A8) was most likely due to leaching of the molecule which was facilitated by the speedy desorption of the added phenol from the refuse.

The pH values of the respective columns are shown in Figure 5.5 B from which it is clear that the different perfusion regimes directly influenced the pH values. During the early stages (<50 days) the columns were characterised by decreased pH values. However, notwithstanding these initial decreased pH values, the leachate recycle column attained a neutral pH by day 72 and remained around this value, despite the phenol resupplementations, until the termination of the study. The single elution (A5) column, on the other hand, never attained a pH value >5.7 until after the mineral salts addition which resulted in pH increases to a maximum of 6.7. Unlike, the leachate recycle column, the single elution column recorded temporary pH decreases following the phenol resupplementations (RS1 and RS2). The batch operated column (A7) also resulted in low pH values and the mineral salts addition did not appear to effect a sustained increase in the leachate pH values. Surprisingly, however, neutral pH values were recorded following the resupplementation with 1000 mg l^{-1}. The pH values for the rain simulated column (A8) were, in general, irregular and lowered although a neutral pH was recorded on day 138.

The consistently low pH values recorded for the single elution column (A5) were, according to Figure 5.5 C, not as a result of an accumulation of 'total' VFAs as the concentrations decreased from 4301 to 13 mg l^{-1} by day 22. A more likely explanation

was the leaching out of buffering compounds. The beneficial effect of leachate recycle was demonstrated by steady declines in the 'total' VFAs concentrations which coincided with increases in the pH values. It is also worth noting that, for the leachate recycle column, neither of the phenol resupplementations resulted in further accumulations of 'total' VFAs. The low pH values recorded for the batch operated column (A7) could have been due to the sustained high concentrations of 'total' VFAs. Furthermore, the addition of mineral salts did not appear to immediately effect rapid decreases although, by day 500, the 'total' VFAs concentrations had decreased significantly with concomitant increases in the pH values.

The washout or leaching of the fatty acids, which are the immediate precursors to methane production, as a result of a single elution regime, have been shown to significantly affect methane generation potential. For example, Leuschner (1989) calculated the volumes of leachate produced and stoichiometry, in 210 litre drums filled with refuse, and concluded that the practice of leachate discard resulted in the potential loss of 410 litres of methane. A similar effect could have been operative in Column A5 as progressive increases in methane concentrations (24% v/v) were recorded until day 129 after which the concentrations declined. These decreases could have resulted from labile substrate depletion. Despite the occurrence of phenol degradation, as discussed above, this was apparently insufficient to maintain the elevated methane concentrations. Notwithstanding the irregular nature of the methane generation trends, following the mineral salts addition, Column A5, in general, exhibited methane concentration increases with subsequent decreases recorded following the 2000 mg l^{-1} resupplementation. An enhanced methane concentration of 66% (v/v) was recorded for the leachate recycle column (A6) on day 201, after which the concentration declined until the addition of mineral salts. With one exception, the methane concentrations for this column appeared to be suppressed following the resupplementations (RS1 and RS2). However, this apparent inhibition was not concomitant with either an accumulation of 'total' VFAs or notable pH decreases. The batch operated column (A7) was characterised by a pool of 'total' VFAs, due to acidogenesis, although the simultaneous process of acidotrophy appeared to be suppressed until the addition of the mineral salts. Following the addition significant increases in the

methane concentrations were recorded which coincided with decreases in the 'total' VFAs concentrations. These higher methane concentrations were, however, not maintained and, subsequent to the second resupplementation (2000 mg l^{-1}), concentrations as low as 4.3% (v/v) were recorded. A further increase to 43% (v/v) could have been due to the ability of the microorganisms to adapt to the added phenol with the resulting mineralisation of the molecule to methane. The rain simulated column (A8), with one exception, did not appear to produce nor sustain elevated methane concentrations. This was apparent despite the mineral salts addition although slight increases were noted following the second resupplementation (RS2).

"Low Load" Columns

The effects that the operational regimes had on the residual phenol concentrations, pH values, 'total' VFAs and methane concentrations, of the "low load" columns (B5-B8), are shown in Figures 5.6 A - 5.6 D.

As recorded for the single elution "high load" column, the equivalent "low load" column operated with leachate discard (B5) exhibited the most rapid phenol removal rate (Table 5.1) which further increased following the resupplementations (RS2). Unlike the "high load" leachate recycle column (A6), which recorded a higher phenol removal rate compared with the batch operated column (A7), the equivalent "low load" leachate recycle and batch operated columns exhibited similar removal rates (Table 5.1). As discussed previously, the "low load" rain simulated column was characterised by the leaching of a large portion of the added phenol and correspondingly very low rates of phenol catabolism resulted.

The "low load" columns were characterised by suppressed pH values with none of the columns recording marked pH increases prior to the mineral salts additions. Following these additions, with a few exceptions, the leachate recycle and batch operated columns exhibited neutral pH values which were maintained despite the two phenol resupplementations. The low pH values of the leachate recycle and batch operated columns,

Figure 5.6 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for the "Low Load" Cols Operated with Single Elution (B5) Leachate Recycle (B6), Batch Mode (B7) and Simulated Rain (B8) During an Incubation Period of 695 Days.
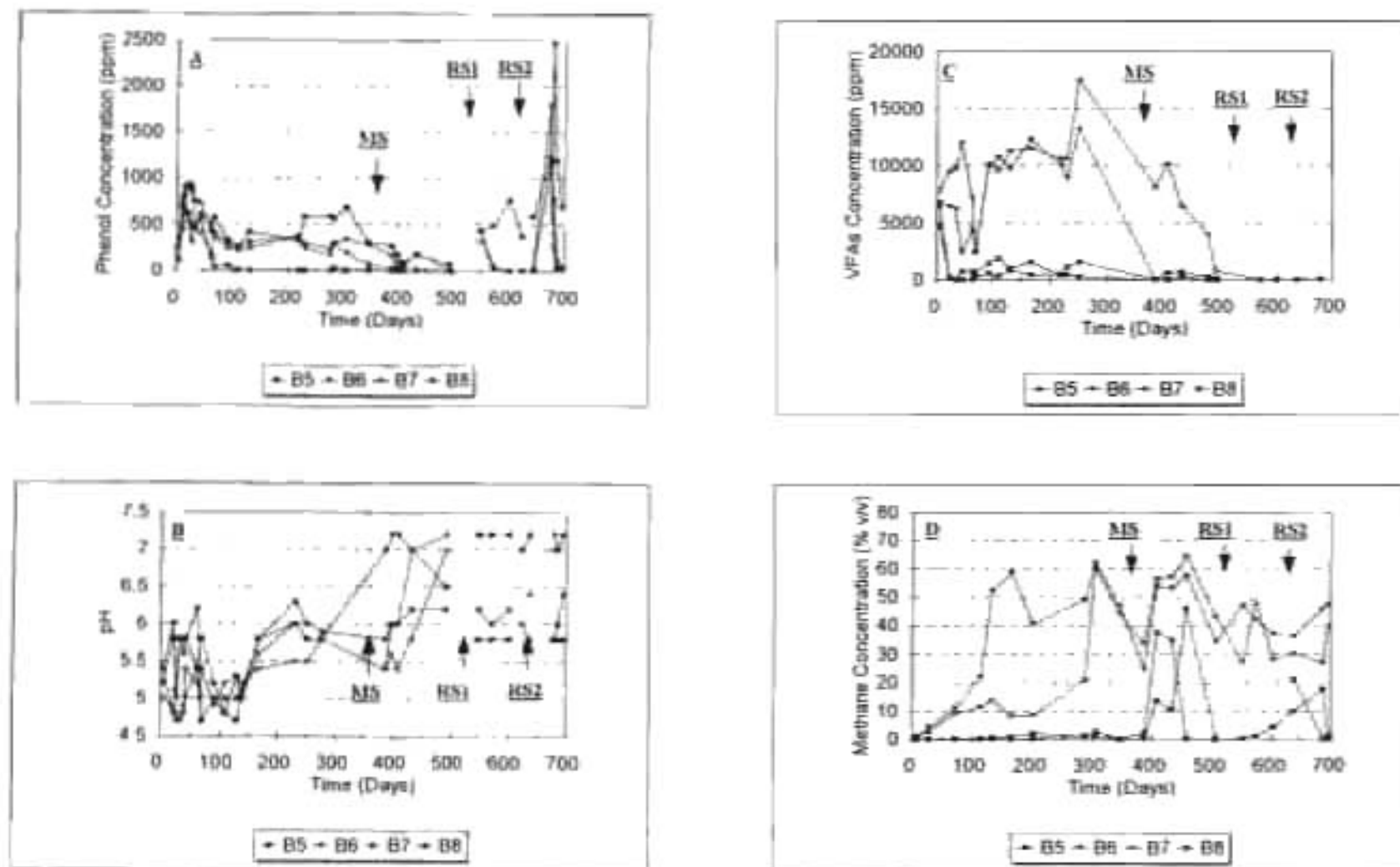
MS Designates Mineral Salt Addition

RS1 Designates Phenol Resupplementation (250ppm)

RS2 Designates Phenol Resupplementation (500ppm)

prior to the mineral salts additions, could have been due to the accumulation of 'total' VFAs (Figure 5.6 C) which subsequently declined. The single elution and rain simulated columns, on the other hand, which were subjected to relatively rapid liquid movement, exhibited low 'total' VFAs concentrations. The low pH values of these columns, in the presence of low 'total' VFAs concentrations, were most likely due to the leaching out of the buffering capacity. Finally, it is worth noting that the two resupplementations did not result in any detectable accumulations of 'total' VFAs.

The effects that the different operating regimes had on the methane concentrations, for the "low load" columns, are clear from Figure 5.6 D. Both the single elution and rain simulated columns exhibited very low methane concentrations prior to the mineral salts additions. The leachate recycle and batch operated columns, on the other hand, recorded concentrations >60% (v/v) on day 309. Further, of great interest were the methane concentrations which were, in general, >30% (v/v) following the two resupplementations.

As discussed earlier for the "high load" columns (Section 5.2.2), transient accumulations of nitrate and sulphate were again recorded, especially with the batch operated column, prior to the mineral salts additions. Subsequently, these accumulations decreased to below detection limits.

5.2.4 Conclusions

The mean phenol removal rates for the different operating regimes are given in Table 5.1. The residual phenol concentrations of both the "high load" and "low load" columns seemed to indicate that the mode of operation should be coupled to the leachate treatment requirements.

For example, if the leachate is to be discarded (for example to municipal sewer), without any further treatment, then the lowest possible concentration of phenol would be the preferred option.

Table 5.1 Mean Phenol Removal Rates for the Single Elution, Leachate Recycle and Batch Operated Phenol Co-Disposal (C5 - C6), "High Load" (A5-A7) and "Low Load" (B5-B7) Columns During an Incubation Period of 695 Days

Column No.	Mode of Operation	Mean Phenol Removal Rate Prior to Phenol Resupplementations ($g\ kg^{-1}\ d^{-1}$)	Mean Phenol Removal Rate Following Phenol Resupplementations ($g\ kg^{-1}\ d^{-1}$)
A5	Single Elution	0.297	0.73
B5	Single Elution	0.31	0.46
C5	Single Elution	0.27	0.59
A6	Leachate Recycle	0.006	0.033
B6	Leachate Recycle	0.0016	0.023
C6	Leachate Recycle	0.005	0.06
A7	Batch	0.0012	0.046
B7	Batch	0.0014	0.023

In this instance, leachate recycle should be implemented as this mode of operation facilitates the reduction of the co-disposed phenol concentrations. On the other hand, if the maximum removal of phenol is desired, then single elution, especially with an increased sludge:refuse ratio, would be the favoured method of operation. This method could, however, result in suppressed pH values which, in turn, could inhibit refuse degradation by, for example, 'souring' of the fermentation or increased heavy metal mobility. In general, the dual co-disposal of phenol and sewage sludge in batch mode could negatively influence not only the leachate quality (increased 'total' VFA's), but also methane generation. One possible reason for the decreased effectiveness of the batch mode of operation could have been the lack of moisture flux which, according to the U.K. Department of Environment (1994), is essential for successful co-disposal. It has been suggested that the system should be controlled so that the hydraulic retention time (HRT) within the refuse mass should be between 1 and 5 years (U.K. Department of Environment, 1994).

The HRT for the single elution columns was approximately two days while for saturated landfills it could be considerably longer, from several months to several years (Knox, 1989). In practice, in full-scale landfills the HRT can be limited by the lower permeability of the refuse which is a function of the refuse type and compaction. For example, plastic, which often constitutes a major portion of municipal solid waste, would tend to decrease the permeability and facilitate perched water tables (Daneel, 1993). The refuse used in this present study was hand sorted to remove all plastic and glass thus facilitating the flux of liquid. Increasing the flux of moisture through a landfill site and, therefore, utilising the enhancing effect of liquid movement, could be accomplished by decreasing the compaction ratio which, however, would also decrease the available airspace and longevity of the site.

From the results for the alternative electron acceptors, nitrate and sulphate, it appeared as if the interrelated processes of nitrate and sulphate reduction were inhibited and this is further discussed in Section 5.2.8

5.2.5 "High Load" and "Low Load" Anaerobically Digested Sewage Sludge and 2000 mg l^{-1} Phenol Dual Co-Disposal with Refuse

Single Elution Columns

The changes in residual phenol concentrations, pH values, 'total' VFAs and methane concentrations for the columns perfused with 2000 mg l^{-1} and operated with single elution are given in Figures 5.7 A - 5.7 D.

The phenomenon of more rapid phenol breakthrough in the presence of sewage sludge additions has been discussed earlier (Section 5.2.2). Initially (< 34 days), similar high leachate phenol concentrations, resulting from the addition of sewage sludge, were recorded recorded for the sludge amended columns (A9 and B9). Despite the increased concentrations due to rapid desorption in the sludge amended columns, these columns averaged significant phenol removal rates. For example, the "high load" (A9) and "low

Figure 5.7 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for 'High Load' (A9), 'Low Load' (B9), Phenol Co-Disposal (C9) and Refuse Only Control (C1) Columns Operated with Single Elution During an Incubation Period of 695 Days
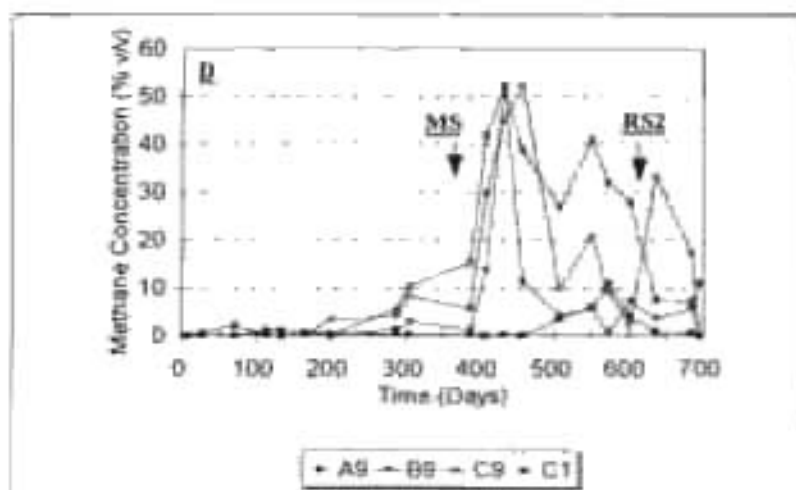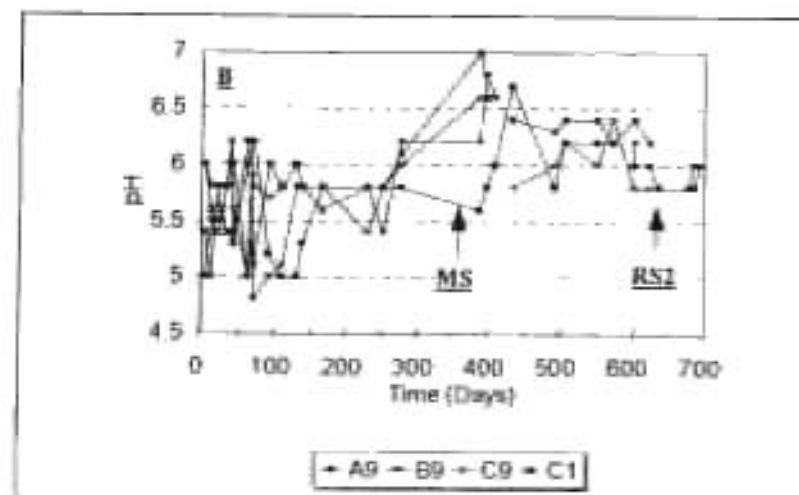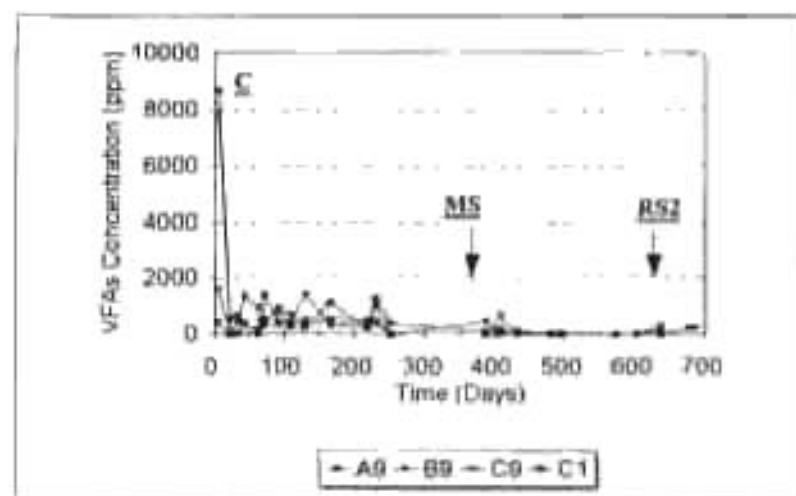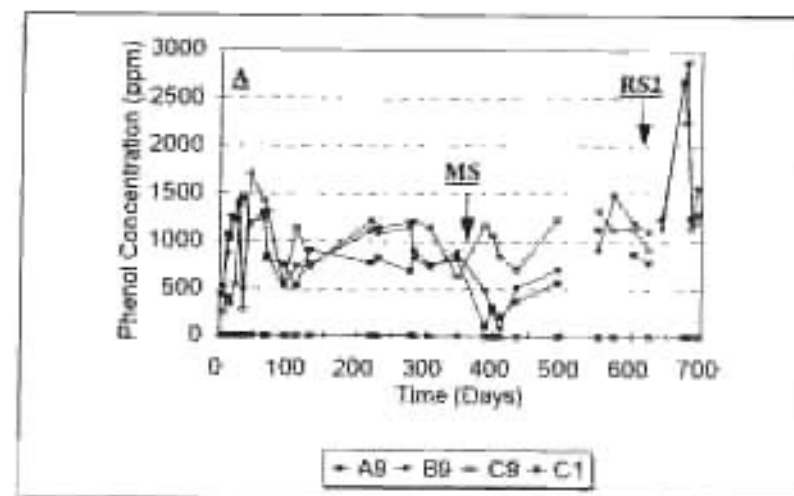
MS Designates Mineral Salts Addition

RS2 Designates Phenol Resupplementation (400ppm)

load" (B9) columns, prior to the phenol resupplementations, maintained phenol removal rates of 0.7 and 0.35g kg^{-1} d^{-1}, respectively while following the resupplementations the rates increased to 1.3 and 1.28g kg^{-1} d^{-1}, respectively. The "high load" column, therefore, exhibited enhanced phenol removal compared with the phenol co-disposal column which averaged pre- and post resupplementation phenol removals of 0.61 and 0.66g kg^{-1} d^{-1} (Table 5.2). It was evident that sludge additions, and particularly the high sludge loading, effected significant increases in phenol catabolism.

Table 5.2   Mean Phenol Removal Rates for the Single Elution (A9, B9, C9), Leachate Recycle (A10, B10, C10) and Batch Operated (A11, B11, C11) Columns During an Incubation Period of 695 Days

Column No.	Mode of Operation	Mean Phenol Removal Rate Prior to Phenol Resupplementations (g kg^{-1} d^{-1})	Mean Phenol Removal Rate Following Phenol Resupplementations (g kg^{-1} d^{-1})
A9	Single Elution	0.7	1.3
B9	Single Elution	0.35	1.28
C9	Single Elution	0.61	0.66
A10	Leachate Recycle	0.0013	0.05
B10	Leachate Recycle	0.0035	0.058
C10	Leachate Recycle	0.0023	0.043
A11	Batch	0.0032	0.055
B11	Batch	0.0031	0.062
C11	Batch	0.0029	0.039

One possible reason for the recorded phenol removal rate increases compared with the phenol co-disposal column, could have been the positive promotion recorded, for Columns A9 and B9, following the mineral salts additions.

From Figure 5.7 B it can be seen that the single elution regime suppressed the pH values with brief increases recorded following the mineral salts addition. Therefore, it appeared that the buffering capacity afforded by the sludge addition (Section 4.3.2) was insufficient to buffer the fermentation against pH decreases. More importantly, with regards to full-scale co-disposal operations, the phenol resupplementations, in general, appeared to further induce pH decreases.

During the first 20 days the 'total' VFAs concentrations were higher in the sludge amended columns (Figure 5.7 C) as discussed previously (Section 4.3.2). The subsequent rapid decreases were most likely due to the leaching effect of the operating regime. The negative effect that leachate discard had on methanogenesis was clear (Figure 5.7 D). Also, it was apparent that, apart from the brief period following the mineral salts additions, the sludge amended columns exhibited lower methane concentrations compared with the phenol co-disposal (C9) and refuse control columns (C1). The reasons for the suppressed methanogenic activity were not clear. However, one possibility could have been that, unlike the previously discussed columns, these columns did not, following the mineral salts additions, exhibit decreases in the concentrations of the alternative electron acceptors, nitrate and sulphate. Despite this apparent microbial inhibition phenol catabolism continued, as evidenced by the mean phenol removal rates which are shown in Table 5.2. This was unexpected as Senior and Balba (1987) reported that phenol catabolism required the effective processes of either sulphate reduction or methanogenesis to act as hydrogen sinks.

Leachate Recycle Columns

During the first 34 days the sludge amended columns (A10 and B10) were characterised by higher residual phenol concentrations (Figure 5.8 A) than the phenol column (C10). After approximately 110 days the sludge amended columns again, in general, exhibited higher leachate phenol concentrations than the equivalent phenol co-disposal column (C10). Following the additions of the mineral salts, however, all three columns recorded similar residual phenol concentrations. Despite these higher leachate

Figure 5.8 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for "High Load" (A10), "Low Load" (B10), Phenol Co-Disposal (C10) and Refuse Only Control (C2) Columns Operated with Leachate Recycle During an Incubation Period of 695 Days
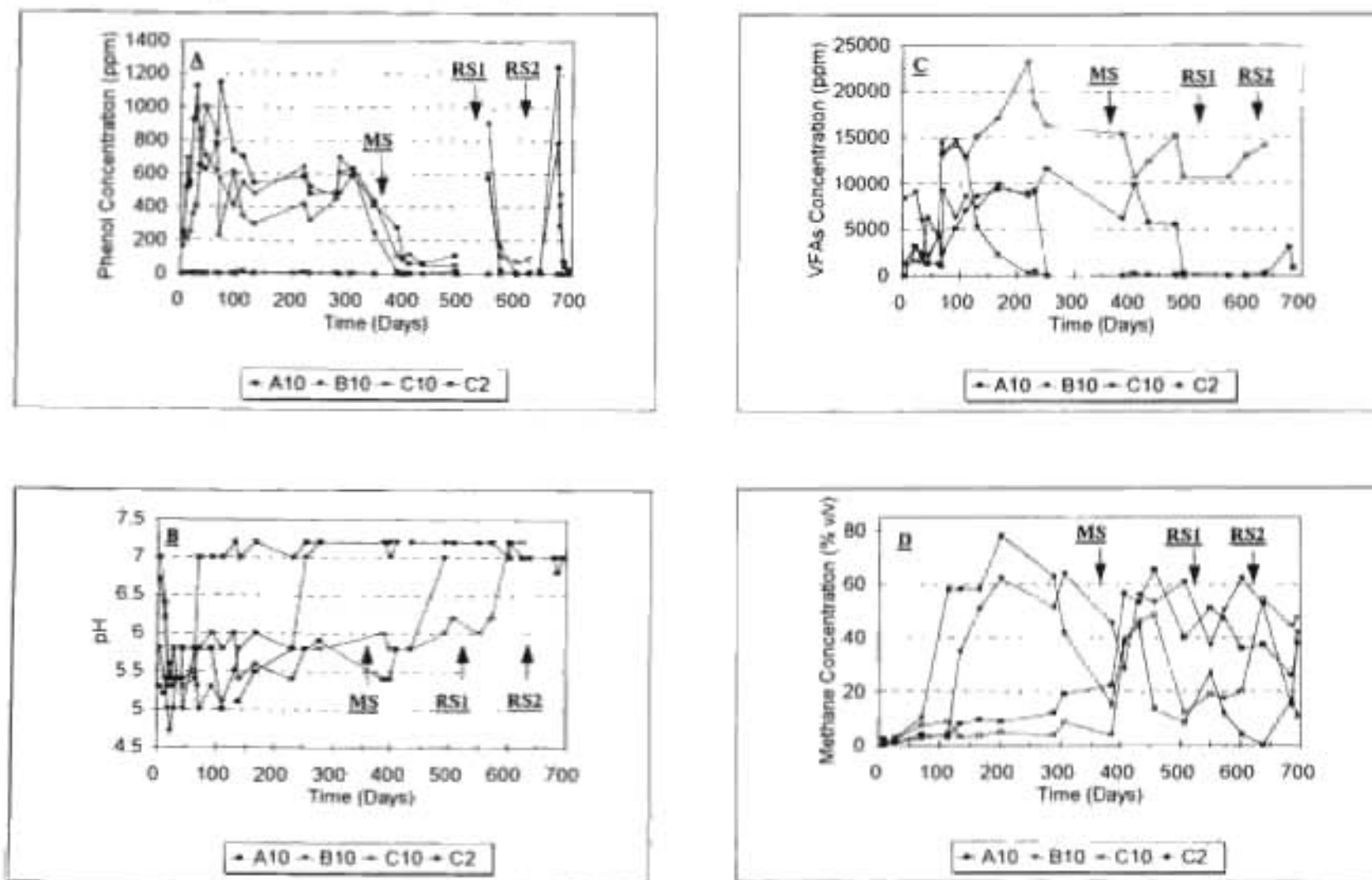
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (200ppm)

RS2 Designates Phenol Resupplementation (400ppm)

phenol concentrations, recorded for the sludge amended columns, the phenol removal rates approximated to or bettered the phenol co-disposal column. For example, the "high load" (A10), "low load" (B10) sludge amended and the phenol co-disposal columns exhibited mean removal rates of 0.0013, 0.0035 and 0.0023g kg^{-1} d^{-1}, respectively. Also evident from Figure 5.8 A was the positive effect that the mineral salts additions had on phenol catabolism. Subsequent to the phenol resupplementations (RS1 and RS2) the sludge amended and phenol co-disposal columns recorded phenol removal rate increases (Table 5.2) to comparable rates.

A possible reason for the increased phenol removal rate, recorded for the "low load" column, could have been the recovery of the pH (Figure 5.8 B) to attain a neutral value after 241 days. At the same time, the equivalent "high load" column exhibited a pH of 5.8. The importance of near neutral pH values in phenol catabolism has been discussed earlier (Section 3.3.2). Therefore, the co-disposal of 2000 mg l^{-1} phenol with the higher sludge:refuse ratio appeared to effect protracted periods of suppressed pH values which only recovered to attain neutral values following the mineral salts addition. However, the benefit of sludge additions, to the pH values, was evident as the phenol co-disposal column (C10) exhibited an even slower recovery.

The co-disposal of sewage sludge in refuse has previously been shown to enhance the release rate of 'total' VFAs (Section 4.3.2). For the present study, however, this was only evident during the first 41 days. The "low load" column (B10) recorded higher 'total' VFAs concentrations compared with the "high load" (A10), phenol co-disposal (C10) and refuse control (C2) columns (Figure 5.8 C). Notwithstanding these initial higher VFAs concentrations, both the sludge amended columns recorded lower mean VFAs concentrations until day 123, after which the refuse control concentrations decreased rapidly. The dual co-disposal of sludge and phenol with refuse, therefore, could be beneficial in negating VFA accumulations compared with the single co-disposal of phenol.

The negative effects of the 'total' VFAs accumulation and concomitant suppressed pH values on the "high load" column were reflected in the relatively low methane

concentrations. For example, a maximum methane concentration of 19% (v/v) was recorded prior to the mineral salts addition. Likewise, high 'total' VFAs concentrations and low pH values, of the phenol co-disposal column (C10), effected similar low methane concentrations. The positive effects of elevated pH values and declines in 'total' VFAs concentrations, on the other hand, were reflected in the enhanced methane concentrations recorded for Columns B10 and C2. The benefit of sludge additions was also apparent following the mineral salts additions and phenol resupplementations as, in general, the sludge amended columns recorded higher methane concentrations. It, therefore, appeared that the sludge loading as well as the phenol concentration could effect different fermentation balances. For example, the "high load" 1000 mg l^{-1} perfused column (Figure 5.2), operated with leachate recycle, exhibited enhanced phenol and refuse catabolism, whereas the equivalent 2000 mg l^{-1} challenged column exhibited a degree of microbial inhibition when pH, 'total' VFAs and methane concentrations were used as the analytical criteria.

Batch Operated Columns

The changes in residual phenol concentrations, pH values, 'total' VFAs and methane concentrations are shown in Figures 5.9 A - 5.9 D.

During the first, approximately, 131 days the "high load" column (A11) recorded the highest leachate phenol concentrations possibly due to the rapid desorption rates as a result of the sludge addition. The sludge amended columns (A11 and B11), as well as the phenol co-disposal column (C11), recorded decreased residual phenol concentrations after approximately 71 days with subsequent strong increases after 223 days. Possible reasons for the subsequent increases, especially for the sludge amended columns, were not apparent although physico-chemical phenomena such as adsorption/desorption could have been involved. Both the "high" and "low" load columns averaged similar phenol removal rates (Table 5.2) despite the, in general, higher leachate phenol concentrations of the "high load" column prior to the phenol resupplementations (RS1 and RS2). From the rates shown in Table 5.2 it can be seen that the sludge additions effected slight increases compared with

Figure 5.9 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for "High Load" (A11), "Low Load" (B11), Phenol Co-Disposal (C11) and Refuse Only Control (C3) Columns Operated in Batch Mode During an Incubation Period of 695 Days
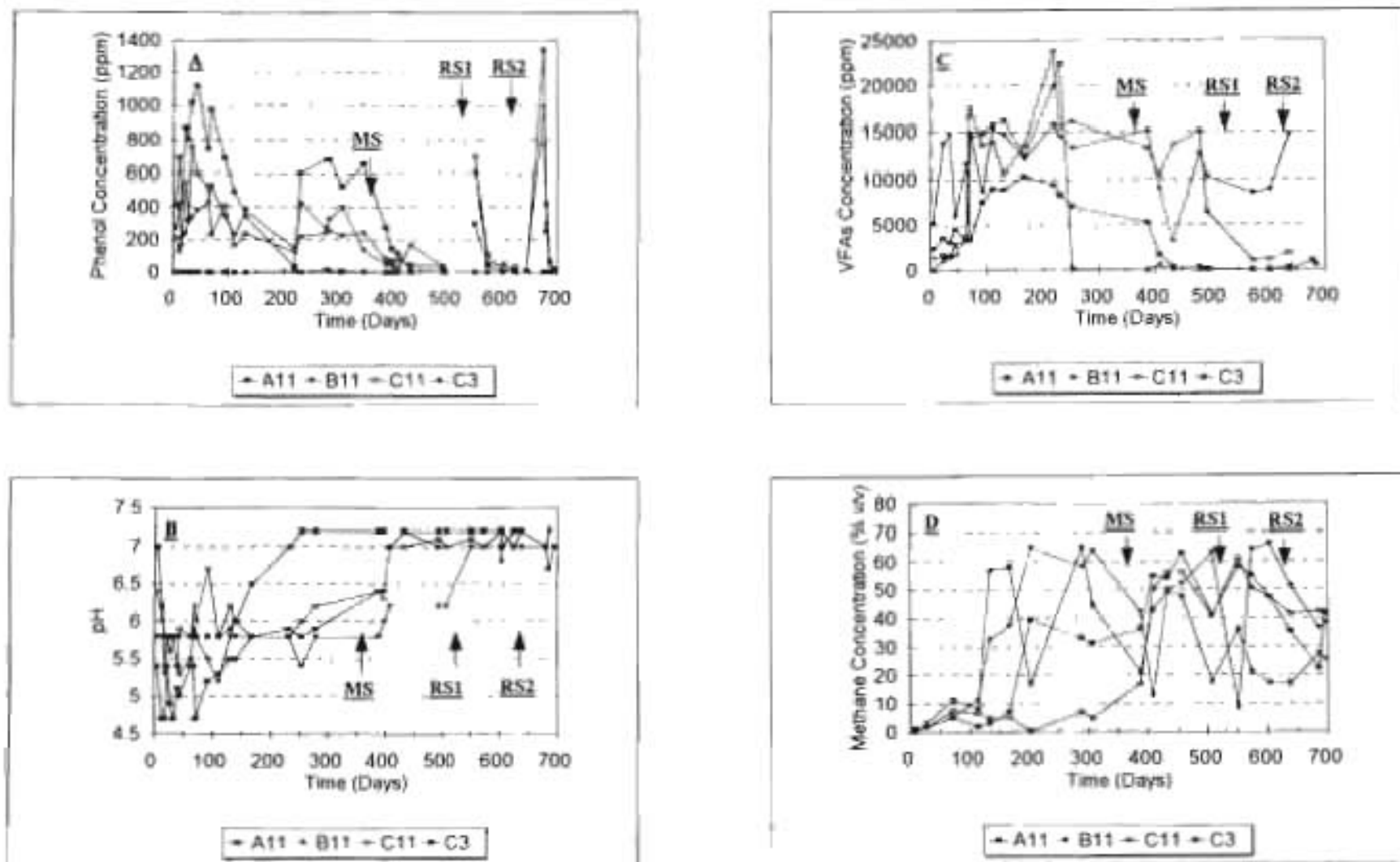
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (2000ppm)

RS2 Designates Phenol Resupplementation (4000ppm)

the phenol co-disposal column. Further, the phenol resupplementations effected increases in the phenol removal rates for all three columns although the highest removal rates were again recorded in the sludge amended columns. From Figure 5.9 A it is clear that the times required for the phenol concentrations to approach zero, after the initial breakthroughs, were significantly shortened following the phenol resupplementations and could have been due to the elimination of possible elemental deficiencies and/or microbial adaption. Alternatively, microbial activities could have been enhanced due to the establishment of more favourable physiological conditions. For example, the nitrate/nitrite concentrations appeared to accumulate prior to the mineral salts additions but subsequently declined.

From Figure 5.9 B it can be seen that during the first 19 days, the sludge amended columns, and especially the "high load" column, exhibited low pH values. The refuse control column (C3) and the phenol co-disposal column (C11) attained neutral pH values after approximately 220 and 414 days, respectively while the equivalent sludge amended columns, A11 and B11, required 391 and 551 days, respectively. For all three co-disposal columns increases in the pH values were recorded following the mineral salts additions.

In the early stages (<75 days) of the study the sludge amended columns, A11 and B11, were characterised by higher 'total' VFAs concentrations (Figure 5.9 C) compared with the refuse control (C3) and the phenol co-disposal (C11) columns. Subsequently, however, the "high load" column exhibited lower 'total' VFAs concentrations than the other columns until day 252 when the refuse control column showed a dramatic decrease. It is, thus, surprising that these lower VFAs concentrations, for the "high load" column, were, compared with the "low load" column, not reflected in pH increases prior to the mineral salts additions. The sludge additions to Columns A11 and B11 did not appear to effect increases in the phenol removal rates (Table 5.2) or the pH values compared with Column C11.

However, the benefit of sludge additions was clear with regards to the methane concentrations (Figure 5.9 D). For example, notwithstanding the protracted period of suppressed pH values of the "low load" column (B11), methane concentrations >60%

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(v/v) were recorded after 200 days. On the other hand, the equivalent phenol co-disposal column (C11) recorded very low concentrations of approximately 1.7% (v/v) over the same time period. Also of interest was the continued enhanced methane concentrations recorded for the co-disposal columns (A11, B11, C11). From Figure 5.9 D it is clear that following the mineral salts additions and the phenol resupplementations the headspace methane concentrations, with a few exceptions, were greater in the co-disposal columns than the refuse control (C3). The decrease in methane concentrations from an initial maximum was described in Section 3.3.2 where it was concluded that substrate limitation was the most likely cause. These protracted methane concentration increases, of Columns A11, B11 and C11, therefore, seemed to imply that the practice of co-disposal with either sludge or phenol additions could supplement the refuse with labile substrates which, in turn, could enhance methanogenesis. Alternatively, increased methane concentrations resulting from differential inhibition and re-directed electron flow could be implicated and has been discussed earlier (Section 3.3.4).

The increases in the phenol removal rates following the phenol resupplementations were ascribed above to the possible establishment of a favourable environment, particularly a low redox potential. However, the methane concentration results did not support this possibility as high concentrations, especially for the sludge amended columns A11 and B11, were recorded prior to the mineral salts additions and resupplementations. The heterogeneous nature of refuse could, however, have facilitated the formation of micro-niches where methanogenesis was enhanced despite, in general, unfavourable physiological conditions.

Simulated Rain Columns

The rapid leaching of the added phenol, described earlier, was also evident in these columns (Figure 5.10 A). For example, during the first 110 days approximately 73% of the added phenol was leached from the "low load" column. The practice of leachate discard has been associated with low pH values and attributed to the removal of the buffering capacity (Pohland, 1989b). The moisture movement through the rain simulated column appeared to

Figure 5. 10 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for "High Load" (A12), "Low Load" (B12), Phenol Co-Disposal (C12) and Refuse Control (C4) Columns Operated with Simulated Rain During an Incubation Period of 695 Days
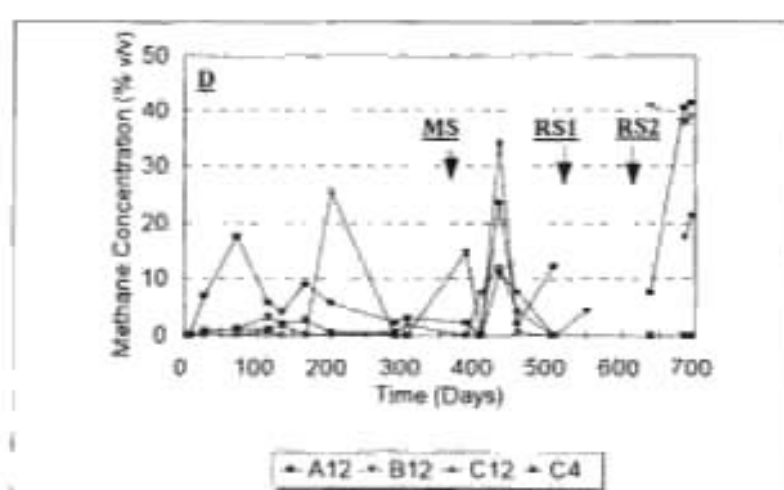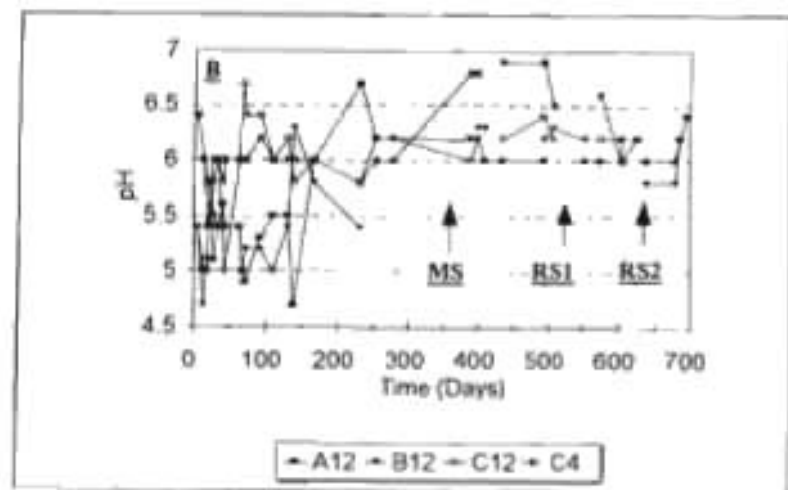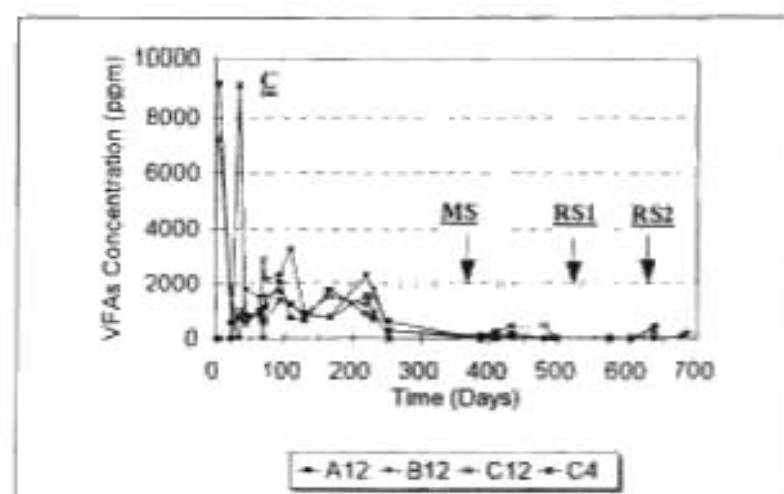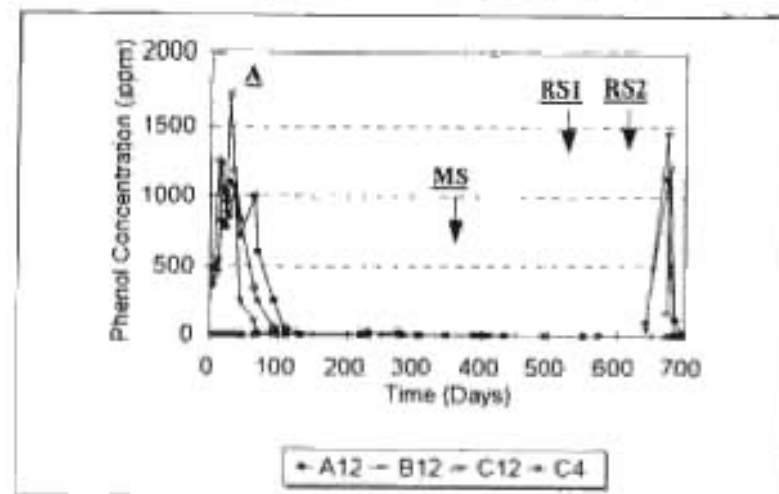
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (2000ppm)

RS2 Designates Phenol Resupplementation (4000ppm)

effect similar leaching characteristics which, in turn, resulted in lower pH values for the duration of the study (Figure 5.10 B). Similarly, the trends of the 'total' VFAs concentrations reflected the washout of these components (Figure 5.10 C). The inhibiting effect of this practice, on refuse degradation, was also evident in the generally low methane concentrations (Figure 5.10 D) recorded for the columns. However, it is worth noting that the phenol resupplementations effected methane concentration increases for the experimental columns.

5.2.6 Effects of Perfusion Strategy in the Dual Co-Disposal of Anaerobically Digested Sewage Sludge and 2000 mg l^{-1} Phenol with Refuse

"High Load" Columns

The changes in residual phenol concentrations, pH, 'total' VFAs concentrations and methane headspace concentrations are shown in Figures 5.11 A - 5.11 C.

Generally, the single elution operated column (A9) recorded higher leachate phenol concentrations than the other columns operated with leachate recycle (A10), batch mode (A11) or simulated rain (A12). The higher residual phenol concentrations of Column A9 were, however, prior to the mineral salts addition, concomitant with higher phenol removal rates (Table 5.2). The equivalent leachate recycle and batch operated columns (A10 and A11), in general, exhibited residual phenol concentrations >500 mg l^{-1} prior to the mineral salts additions with sharp declines following the additions. The simulated rain column (A12), on the other hand, was characterised by rapid leaching of the added phenol with 82% of the phenol washed out by day 92. Following the resupplementations (RS1 and RS2) phenol removal rate increases were recorded, especially for the single elution column, while the leachate recycle and batch operated columns demonstrated similar removal rates.

Prior to the mineral salts additions the columns, with a few exceptions, recorded relatively low pH values (<pH6). Surprisingly, however, the simulated rain column exhibited pH values > pH6 after 171 days. Following the mineral salts additions the single

Figure 5.11 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for the "High Load" Columns Operated with Single Elution (A9), Leachate Recycle (A10), Batch Mode (A11) and Simulated Rain (A12) During an Incubation Period of 695 Days
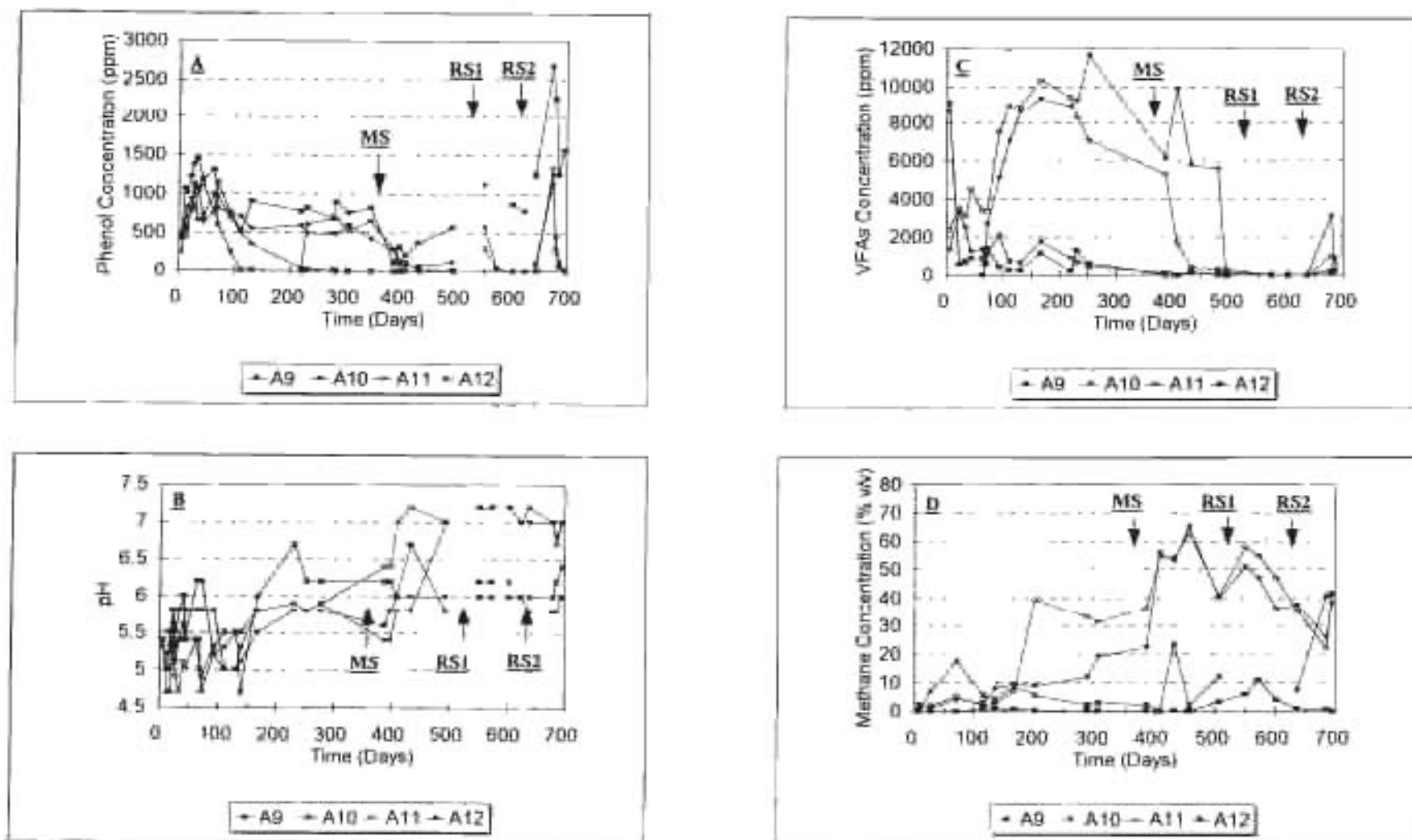
MS Designates Mineral Salts Addition

RS1 Designates Phenol Resupplementation (2000ppm)

RS2 Designates Phenol Resupplementation (4000ppm)

elution and simulated rain columns, with one exception, recorded pH values <6.5. The equivalent leachate recycle and batch operated columns, on the other hand, exhibited elevated pH values although slight decreases were recorded following the phenol resupplementations.

During the first 14 days the effects that the operating regime had on 'total' VFAs concentrations were clear (Figure 5.11 C). The higher 'total' VFAs concentrations recorded for the single elution and simulated rain columns could have been due to either increased acidogenesis or the mobilisation and leaching out of acids as a result of the liquid flux which, subsequently, resulted in continuously low concentrations. The equivalent leachate recycle and batch operated columns exhibited progressive increases in the 'total' VFAs concentrations which subsequently approached very low concentrations following the mineral salts additions. The slight pH decreases recorded for the leachate recycle and batch operated columns following the phenol resupplementations could have been due to the coincident slight increases in the 'total' VFAs concentrations.

During the first 100 days of the study the simulated rain column exhibited surprisingly high methane concentrations which were, however, not maintained. The leachate recycle and batch operated columns, prior to the mineral salts additions, exhibited methane concentrations of 31 and 19% (v/v), respectively. The positive effects of the additions, for Columns A10 and A11, were reflected in the sharp increases in the methane concentrations to >60% on day 458. From Figure 5.11 D it appeared that the phenol resupplementations negatively affected the methane concentrations of columns A10 and A11 although further increases were recorded towards the termination of the study. Also of interest were the increases in methane concentrations of the simulated rain column (A12) following the second resupplementation (RS2). It, therefore, appeared that the resupplementations enhanced methane production. These increases in methane concentrations could have been due to the addition of labile carbon (phenol).

"Low Load" Columns

Not unexpectedly, like the "high load" column, the single elution column (B9) with few exceptions, exhibited the highest leachate phenol concentrations (Figure 5.12 A) and the greatest phenol removal rate (Table 5.2) which again increased significantly following the phenol resupplementations (RS1 and RS2). Prior to the mineral salts addition the batch operated column (B11) exhibited lower residual phenol concentrations than the leachate recycle column (B10) although both averaged similar phenol removal rates (Table 5.2) pre- and post the phenol resupplementations (RS1 and RS2). The simulated rain column (B12) was characterised by rapid leaching of the added phenol.

Analogous to the "high load" columns, described above, the columns exhibited low pH values prior to the mineral salts additions following which the columns recorded pH increases. These elevated pH values were, however, not maintained for columns B9 and B12. The leachate recycle and batch columns, on the other hand, attained neutral pH values on days 392 and 600, respectively.

The relatively slower recovery in the pH values of the batch column (B11) compared with the leachate recycle column (B10) could have been due to the continuously higher 'total' VFAs concentrations in the former. Both the single elution and simulated rain columns were characterised by washout of the 'total' VFAs. The suppressed pH values for these two columns could thus not have been due to an accumulation of these acids and were more likely due to the leaching of the compounds responsible for the buffering capacity.

The enhancing effects that the leachate recycling and batch regimes had on refuse fermentation were evident in the progressive increases in the methane concentrations to >60% (v/v) on day 200. Despite the irregular nature of the methane concentration results following the mineral salts additions, the leachate recycle and batch operated columns, in general, exhibited higher methane concentrations than both the simulated rain and single elution columns. Further, from Figure 5.12 D, it appeared that the phenol resupplementations resulted in additional methane concentration decreases. Similar to the

Figure 5.12 Changes in Residual Phenol Concentrations (A), pH (B), 'Total' VFAs Concentrations (C) and Methane Concentrations (D) for the "Low Load" Columns Operated with Single Elution (B9), Leachate Recycle (B10), Batch Mode (B11) and Simulated Rain (B12) During an Incubation Period of 695 Days
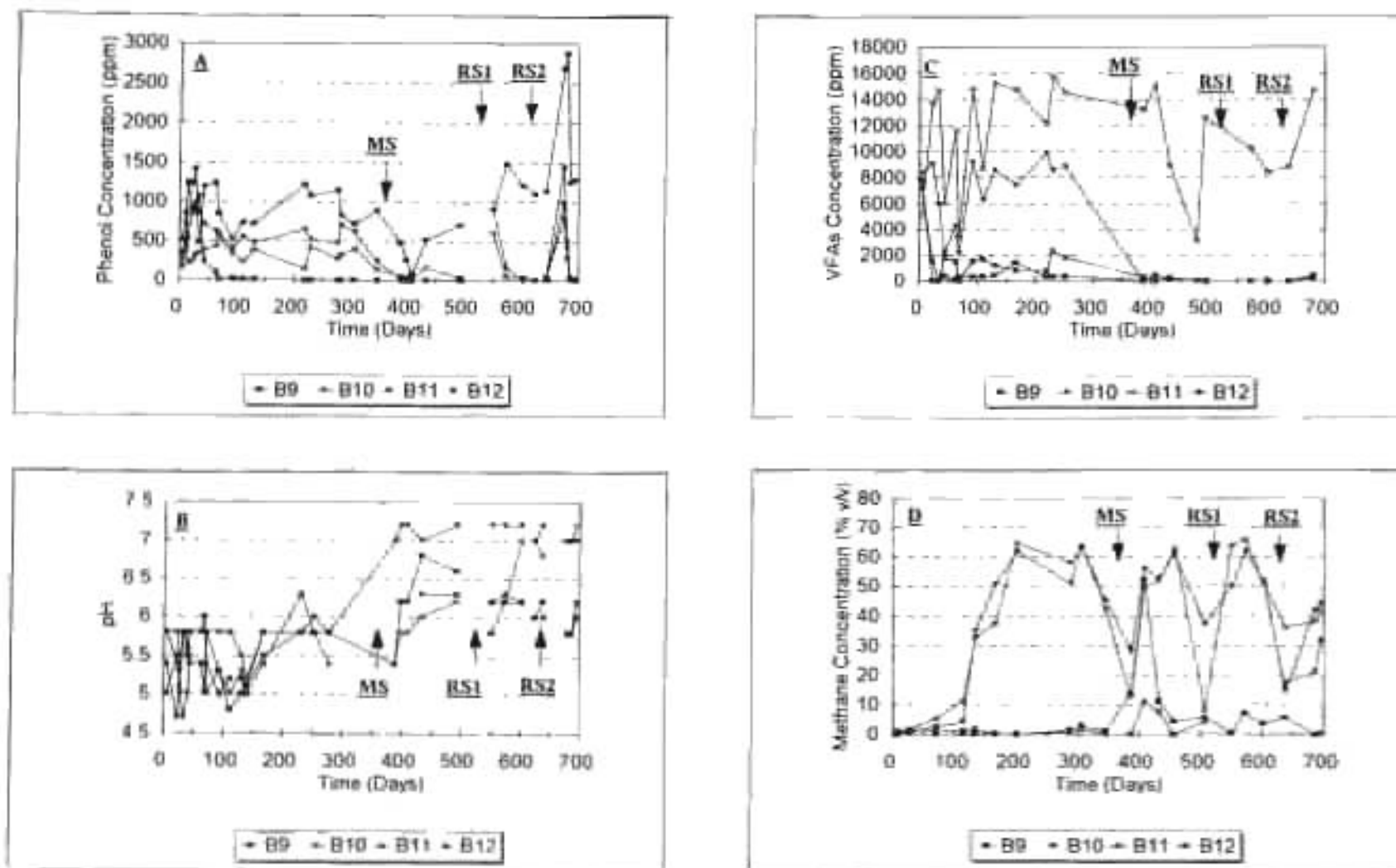
"high load" columns, the simulated rain exhibited an increase in methane concentrations following the second resupplementation to a terminal value of 32% (v/v). These, subsequent, methane concentration increases could have been due to one or a combination of the following: a. The addition of a labile carbon source (phenol); b. The enhancing effect of liquid movement; c. Microbial adaption to the added molecule; and d. The elimination of possible nutrient limitations. However, all the above factors would also have been operative in the single elution column which did not record similar increases in the methane concentrations.

5.2.7 Conclusions

From the results discussed above it can be seen that the addition of sludge in "high" loads slightly increased the phenol removal rates compared with the phenol only column. The reason for the lower removal rates recorded for the "low" load columns, prior to the phenol resupplementations, is not clear as the analytical parameters of pH values, 'total' VFAs and methane concentrations did not vary greatly from the other two co-disposal columns (A9 and C9). Although the single elution columns exhibited higher phenol removal rates compared with the other operating regimes this practice was also coincident with reduced pH values and rapid washout of the buffering capacity and methane precursors. However, it is worth noting that, following the phenol resupplementations, the single elution, sludge amended, columns were characterised by increased phenol removal rates, notwithstanding the continued low methane concentrations.

The significant influence of pH on refuse and phenol catabolism (Table 5.2) was evident from the results exhibited by the leachate recycle columns. The enhanced refuse and phenol catabolism exhibited by the low load" column (B10), compared with the equivalent "high load" column, could have, in part, been due to the more rapid attainment of a neutral pH. On the other hand the the "high load", batch operated column, exhibited enhanced refuse and phenol catabolism. It, therefore, seemed that, for these experimental columns, the optimum sludge loading was dependent on the operational regime employed.

Unlike the 1000 mg l^{-1} perfused columns (Figure 5.5), with one exception, none of the "high load" co-disposal columns challanged with 2000 mg l^{-1} (A9, A10, A11, A12), prior to the mineral salts additions exhibited a pH >6.4. Similar to the 1000 mg l^{-1} perfused columns, the single elution columnns with either "high" or "low" sludge loadings were, despite continuously low pH values and methane concentrations, characterised by the greatest phenol removal rates. Thus, irrespective of the sludge and/or phenol loading the single elution regime exhibited the greatest phenol removal rates. Further, similar to the 1000 mg l^{-1} perfused "high" and "low" sludge load columns with leachate recycle the equivalent 2000 mg l^{-1} perfused columns, A10 and B10, overall exhibited the most enhanced refuse degradation when pH, methane concentrations and 'total' VFAs were used as the analytical criteria.

CHAPTER 6

THE RELATIVE IMPORTANCE AND INFLUENCE OF ADSORPTION AND DESORPTION IN THE ATTENUATION OF PHENOL IN REFUSE

6.1 Introduction

According to McBride (1994) the fate and transport of contaminants and, therefore, the efficacy of co-disposal are largely controlled by two processes, adsorption, an abiotic process, and bioconversion or biodegradation. Effective co-disposal, therefore, is reliant upon the sorptive properties of the municipal waste, as well as the microbial degradation reactions which occur wherever putrescible material is buried.

The effect of adsorption as a retention mechanism is usually illustrated by means of an adsorption isotherm which is constructed with a wide range of aqueous concentrations of the adsorbate (co-disposed compound) each equilibrated with the adsorbent (ie. the refuse) (Weber, Best and Ganese, 1993). The shape of the resulting isotherm reflects the adsorption mechanism involved and, in particular, the interactions involved between the adsorbate and adsorbent (McBride, 1994).

Four main types of isotherms have been defined: 1) The S-type, or solvent affinity, isotherm occurs when the adsorbent has a lower affinity for the solute (adsorbate) than it does for the solvent at low concentrations of the adsorbate; 2) The C-type, or constant-partition, isotherm occurs when the adsorbent has an equal affinity for the adsorbate and the solvent, at all concentrations, while the availability of sites remains constant at all concentrations up to saturation point; 3) The L-type, or Langmuir, isotherm occurs when the adsorbent has a greater affinity for the adsorbate than it does for the solvent at low adsorbate concentrations. In this case, however, as the adsorbate concentration increases adsorption decreases due to the reduction in available adsorption sites; and 4) The H-type, or high affinity, isotherm occurs when the adsorbent has a high affinity for the adsorbate and hence removes all of the adsorbate from solution until its surfaces are completely covered by the solute at which time additional adsorbate remains in solution (Sposito, 1983; Weber and Miller, 1989; Weber et al., 1993; McBride, 1994).

The isotherm reflects the strength of the bonding mechanisms involved and it has been suggested that the strength increases in the order of H > L > C > S (Weber *et al.*, 1993). The strength of the adsorbate-adsorbent interaction determines how strongly the adsorbent will adsorb the adsorbate and, hence, in turn the reversibility of adsorption. The strength of these interactions is indicative of the form of adsorption involved. Strong interactions are indicative of chemical adsorption in which a covalent or electrostatic bond forms between the adsorbent and adsorbate. Weak adsorption is characteristic of physical adsorption in which the bonding interaction is not very energetic (McBride, 1994) and usually involves hydrogen bonding. In some instances adsorption involves a combination of the two and usually one type predominates depending, largely, on the nature of the adsorbate.

It is acknowledged that a large portion of this phase of the study was conducted by S. Costley, as part of a Bsc (Hons) project, whose assistance is greatly appreciated

6.2   Results and Discussion

6.2.1 Adsorption Isotherm

In the adsorption experiment, the mass of phenol which had been removed from the solution phase at the end of the study was assumed to be adsorbed on the refuse according to Kan, Fu and Tomson (1994):

$$q_{adsorbed} = (C_{initial} - C_{ads})V_w/Ws \qquad\qquad 6.1$$

where: $q_{adsorbed}$ = the mass of phenol adsorbed on the refuse (mg kg^{-1});

$C_{initial}$ = the initial phenol concentration (mg l^{-1});

C_{ads} = the amount of phenol remaining in solution at the end of the experiment (mg l^{-1});

V_w = the volume of the solution (l); and

W_s = the weight of the refuse sample (kg).

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To eliminate the possibility of microbial degradation of the phenol, which would influence the apparent amount of phenol adsorbed, sterile refuse microcosms were required. Sterilisation by autoclaving, for example, had previously been associated with changes in the soil matrix (C.A. du Plessis, personal communication) and hence it was decided that gamma-irradiation would have the least impact on the refuse matrix. Thus, any decline in the amount of phenol in solution was assumed to be due to adsorption. After incubation the amount of phenol adsorbed was determined (Kan *et al.*, 1994) and following this an adsorption isotherm was constructed (Figure 6.1).

Figure 6.1   Average Phenol Adsorbed on the Refuse ($q_{adsorbed}$) in Relation to the Solution Phase Phenol Concentration (C_{ads})
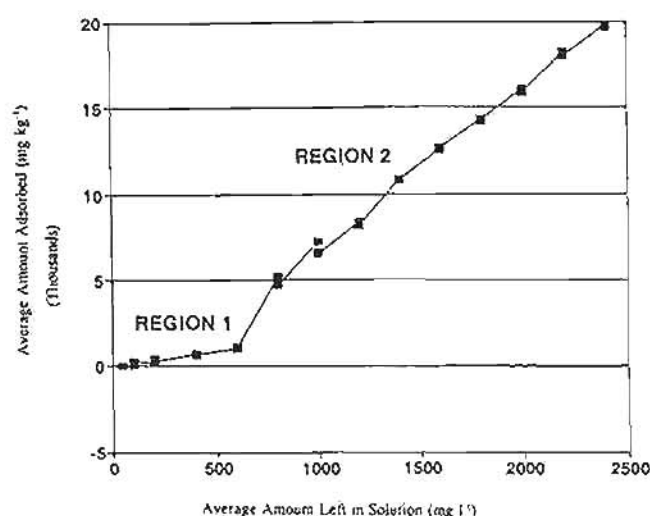
The isotherm clearly illustrates two regions of adsorption suggestive of an S-type isotherm or, alternatively, of multilayer adsorption. The plot shows an initial low rate of adsorption between phenol concentrations of 50 and 600 mg l^{-1} with an increased rate of adsorption with higher phenol concentrations. Determination of the percentage phenol adsorbed clearly indicates the different adsorption phenomena in the two regions. For example, below 600 mg l^{-1}, no more than 19% (w/v) of the phenol was adsorbed, whereas above 800 mg l^{-1}, on average, 75.3% (w/v) phenol adsorption occurred. These results were

unexpected and contrary to trends previously recorded where higher adsorption usually occured at lower concentrations and, characteristically, displayed a plateau indicative of saturation of the adsorptive sites (Knox *et al.*, 1993).

Previous studies have indicated isotherm data points which confirm strong linearity between adsorbed and solute concentrations (Karickhoff, Brown and Scott, 1979; Rogers, McFarland and Cross; 1980; Reinhart, Gould, Cross and Pohland, 1990). Therefore, it was expected that a linear isotherm would be obtained, ie. increased adsorption associated with increased initial phenol concentrations. Furthermore, phenols have been shown to have a higher affinity for organic materials than for water in adsorption studies made in soil media which also indicates that the phenol should exhibit a high rate of adsorption to the refuse (Bohn, McNeal and O'Connor, 1985).

However, this study failed to illustrate a linear isotherm. Phenol is moderately soluble in water (Londry and Fedorak, 1992). It has been shown that below the maximum water solubility of the compound adsorption is controlled by the adsorbent.

The low adsorption levels associated with the low phenol concentrations may have been due to the stronger affinity of the refuse matrix for the water molecules than the phenol molecules or, alternatively, affinity between adjacent phenol molecules may have been greater than the affinity of phenol for the refuse surfaces. Furthermore, organic molecules tend to be non-polar thus preferring an environment less polar than water. However, if another less polar phase, such as soil surfaces, is available the organic molecules are in fact forced out of the aqueous phase onto the less polar phase (Bohn *et al.*, 1985). Hence, the solubility of the refuse surface could largely have determined phenol adsorption.

These results emphasized the need for additional studies to focus on the surface properties of refuse and their affect on adsorption. The ability of phenol to form hydrogen bonds may stimulate its bonding with other phenol molecules or, alternatively, with water molecules, thereby reducing its apparent adsorption. Furthermore, water has a tendency to

compete effectively for adsorption sites and at low phenol concentrations, the water may effectively out-compete the phenol molecules for adsorption sites (McBride, 1994).

The second region, which occurred between 800 and 2400 mg l^{-1}, was associated with a rapid increase in the mean phenol adsorption concentrations. On average, 75% of the phenol was adsorbed indicating a high affinity of the phenol for the refuse which was further highlighted by the low aqueous phase concentrations. Unlike most isotherms, a plateau indicating saturation of adsorption sites was not reached with the phenol concentrations used in this study as the amount of phenol adsorbed continuously increased. Region two suggested higher affinity between the refuse and the phenol molecules which, in turn, allowed for increased adsorption.

The results obtained, therefore, suggested an S-type isotherm which is a non-linear isotherm and convex with respect to the abscissa (Knox et al., 1993) and illustrates an increase in the affinity of the adsorbent for the adsorbate after the initial adsorption. This has been attributed to strong intermolecular bonds (Haung, 1983) and the divergence from linearity is assumed to be due to multilayer adsorption (Kaufman, 1983; Bohn et al., 1985). If applied to the current results, region one appeared to reflect the initial low affinity between the adsorbent (refuse) and the adsorbate (phenol). During this period small amounts of phenol may be adsorbed to the refuse surface forming a monolayer of adsorption. Further increases in the concentration result in the build up of the monolayer into multilayers. It has been suggested that once a monolayer of adsorption has occurred, it may become thermodynamically more favourable for adsorbate molecules to begin 'stacking' on top of other adsorbate molecules. Hence, higher concentrations are associated with the adsorption of adsorbate molecules to other adsorbate molecules already adsorbed to the adsorbent. Furthermore, phenol molecules have been shown to form strong intermolecular bonds which could promote multilayer adsorption.

The occurrence of an S-type isotherm may also account for the lack of a plateau since if adsorption at higher concentrations was due to phenol molecules forming multilayers then the adsorptive capacity of the refuse, which normally determines the degree of saturation, would be of less importance. In this instance the degree of saturation would be determined by the continued ability of phenol molecules to adsorb to each other.

An alternative explanation for the nature of the isotherm, is the possibility that the adsorption sites exhibit different degrees of affinity for the adsorbate. In such cases, the sites with higher affinities would be filled first. At higher concentrations, the less favourable adsorption sites become occupied and hence may account for the increase in adsorption (Knox et al., 1993). However, such an isotherm would be limited by the adsorptive capacity which, once saturated, would prevent further adsorption.

Adsorption has also been suggested to occur in two stages, an initial fast step followed by a time dependent second adsorption step during which the compound diffuses deeper into the adsorbent (Reinhart et al., 1990). The presence of high concentrations of phenol could have allowed for faster diffusion of phenol into less accessible adsorption sites accounting for the apparent high adsorption rates at high capacities.

The nature of the adsorption isotherm can, further, be used to predict possible movement of solutions of different phenol concentrations through the refuse matrix. For example, it can be used to determine the retardation of phenol as well as its mobility. The low amount adsorbed in region one, implied a relatively high amount in solution which suggested a low rate of phenol retardation and an increased possibility for a high degree of leaching. However, when solutions containing higher phenol concentrations are applied, retardation by means of adsorption is enhanced which . likewise, suggests a reduced degree of dispersion. Hence, adsorption with high phenol concentrations appears to be more successful at retarding the movement of phenol in the refuse matrix.

6.2.2 Desorption of Phenol

Figure 6.2 illustrates the adsorption and desorption breakthrough isotherms from refuse challenged with a phenol concentration of 200 mg l^{-1}. Please note that C_o represents the initial phenol concentration while C represents the phenol concentration in solution. The breakthrough of phenol was very rapid as indicated by the slope of the curve with 50% (w/v) resulting within 2 pore volumes. Influent and effluent parity, was achieved after 9 pore volumes. This was in agreement with the previous adsorption isotherm. Subsequent desorption was also very rapid with 75% (w/v) resulting within 4 pore volumes.

Figure 6.2 Breakthrough Curve of a Refuse Column Challenged with 200 mg l^{-1} Phenol Influent
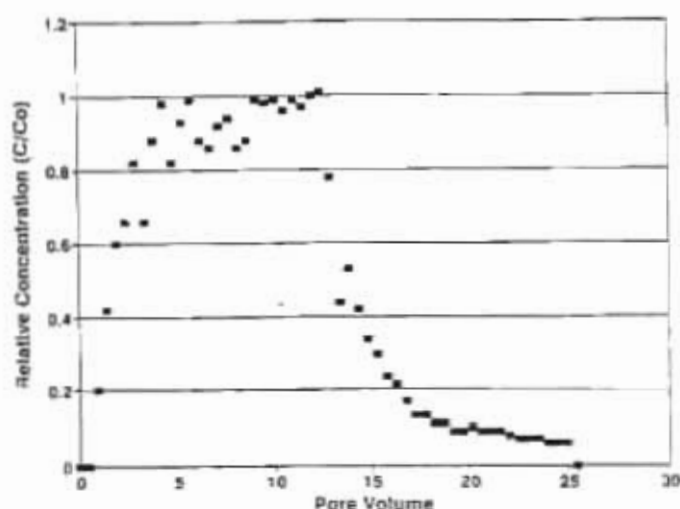
Figure 6.3, likewise, illustrates the breakthrough curve for the 2000 mg l^{-1} phenol challenged system which exhibited a similar rapid breakthrough; 50% (w/v) occurring within 2 pore volumes. Subsequent desorption was also very rapid; 90% (w/v) desorbing within 5 pore volumes. The remaining 10% (w/v) required approximately 18 pore volumes for complete desorption.

Figure 6.3 Breakthrough Curve of a Refuse Column Challenged with a 2000 mg l^{-1} Phenol Influent
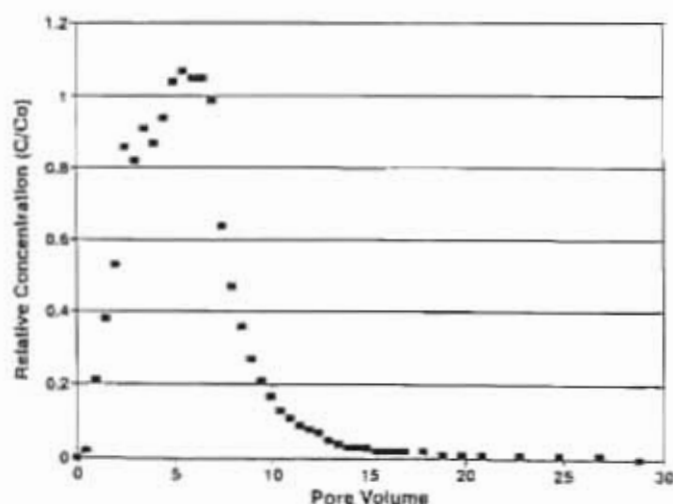
Figure 6.4 illustrates the similarity between adsorption of the 200 and 2000 mg l^{-1} phenol influent solutions. With both concentrations 50% (w/v) breakthrough resulted at 2 pore volumes. Similar, high rates, of phenol desorption were also apparent for both columns. Variations in the desorption rates were only apparent after approximately 75% (w/v) desorption has occurred as the 2000 mg l^{-1} phenol challenged series required almost twice as long to desorb than the 200 mg l^{-1} phenol solution which suggested a higher adsorptive affinity at 2000 mg l^{-1}.

Figure 6.4   Breakthrough Curves for Both the 200 and 2000 mg l^{-1} Phenol Influent
Concentrations
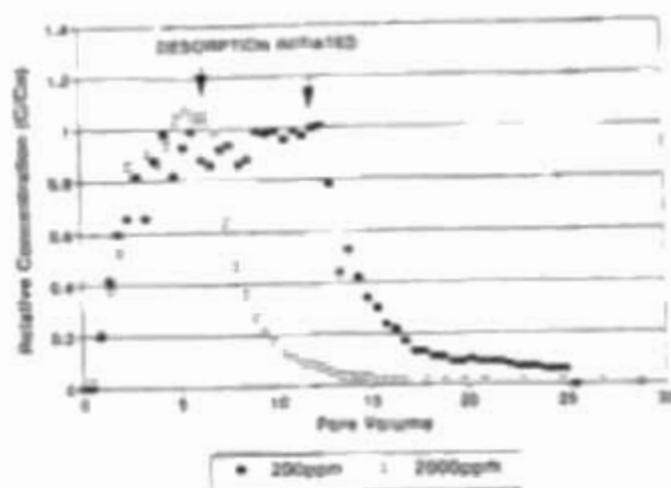
These experiments illustrated no delay in the breakthrough of phenol, with either of the two concentrations. Similar results were obtained by Knox (1989) who used column lysimeter studies and suggested that early breakthrough was possibly due to short-circuiting within the refuse, hence reducing the opportunity for adsorption. The channelling of liquids through preferential pathways may result in a rapid flow through of the compound and hence allow little time for adsorption (Watson-Craik and Senior, 1989b).

Higher concentrations in the adsorption isotherm were associated with higher adsorption. Hence the adsorption curve corresponding to a phenol concentration of 2000 mg l^{-1} was expected to exhibit a gradient less steep than the equivalent 200 mg l^{-1} curve. A shallow gradient would be associated with a high adsorptive capacity. However, the initial

gradient did not differ from that exhibited by the 200 mg l^{-1} adsorption curve as in both curves 50% (w/v) breakthrough of the phenol solution occurred within 2 pore volumes.

It was expected that desorption would be more rapid in the 200 mg l^{-1} concentration system and, therefore, would require less pore volume changes to desorb the phenol as a result of the lower adsorption affinities associated with lower phenol concentrations. However, comparisons of the two resulting desorption curves failed to highlight any significant difference in the slope of the two curves. This suggested that the phenol concentration and its related adsorption affinity had no affect on the rate of desorption. Variations between the two curves were visible only in the latter stages of desorption. For example, the 2000 mg l^{-1} solution required an additional 5 pore volumes compared to the 200 mg l^{-1} solution for complete desorption. This may have been due to the higher number of phenol molecules which were adsorbed with a greater affinity. The remaining fraction has often been referred to as the 'resistant fraction' in previous desorption studies. Other studies have also demonstrated the difficulty of desorbing this fraction (Kan *et al.*, 1994).

The shape of the breakthrough curves obtained with both concentrations was suggestive of relatively weak interactions between the refuse matrix and phenol molecules. This is a characteristic of physical adsorption which is furthermore characterised by multiple layers of adsorption (McBride, 1994), as suggested by the initial adsorption isotherm.

The fact that adsorbed phenol is desorbed relatively rapidly by water is disturbing since the nature of the landfill, with its many attenuation mechanisms, usually results in the downflow of a leachate composed of many different compounds. The presence of other compounds can effectively reduce the polarity of the leachate which encourages desorption of hydrophobic compounds. Sawhney and Kozloski (1984) found that despite phenol exhibiting extensive adsorption on clay, large amounts were still found to be present in the leachate. They concluded that the anaerobic conditions present in landfills may inhibit adsorption and consequently enhance the movement of phenol through the refuse mass. These results suggested that adsorption of phenol by the refuse cannot be relied on to retain the molecule indefinitely in the landfill and that leachate concentrations of previously

adsorbed compounds could increase significantly This may, however, result in greater availability of the compounds for biodegradation although elevated concentrations of the xenobiotic could be detrimental to the microorganisms.

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CHAPTER 7

THE IMPACT OF PHENOL CO-DISPOSAL ON INTERSPECIES INTERACTIONS IN A HEXANOATE OR BUTYRATE-CATABOLISING MICROBIAL ASSOCIATION BY USE OF A MULTI-STAGE CONTINUOUS CULTURE MODEL SYSTEM

7.1   Introduction

The methanogenic fermentation of both constitutive refuse polymers and co-disposed organic molecules necessitates the concerted metabolic activities of a range of physiologically distinct microbial groups (Senior and Balba, 1990). Further, to investigate the effects of phenol co-disposal on interspecies interactions of isolated associations requires the segregation of each physiological component of the association. Multi-stage chemostats were considered by Parkes and Senior (1988) to be ideal models for studying anaerobic transformations as for each association, the component physiological groups can be separated by careful manipulation of key parameters such as dilution rate and electron donor type and concentration thus facilitating sequential use of transient concentrations of electron acceptors in the presence of generating redox gradients (Senior and Balba, 1990). The result of which is the simultaneous separation of the 'habitat domains' of each physiological type with significant overlapping of the 'activity domains' (Wimpenny, Lovitt and Coombs, 1983). Thus, interspecies interactions and component species may be studied without violation of the constraints of the complete association.

In the study reported here, spatial separation of the component species of an anaerobic hexanoic acid-catabolising association was attempted in the presence of two exogenous electron acceptors, nitrate and sulphate, with the objective of examining the microbiology underpinning the dual co-disposal of phenol and anaerobically digested sewage sludge with refuse.

7.2   Experimental Procedure

Initially six, four-stage models were constructed, however, since fermentation balance results indicated that complete separation had not been affected, with wall growth implicated as a causal factor, washout experiments were initiated. Unfortunately, perturbation of the culture with LPG, accidentally supplied by the manufacturer in an OFN cylinder, changed the course of the study and gave extremely interesting results with major implications to the solid-state refuse methanogenic fermentation. Six models (Coutts *et al.*, 1987) with component vessel working volumes of 155,331,736 and 1732 ml were built. The first vessel of each array was inoculated (1:4) with the hexanoic acid-catabolising microbial association and maintained under batch culture conditions at 30°C for 3 days. The models were then operated under open culture conditions and influent medium was introduced into the first vessels by means of a Watson-Marlow 202v flow inducer to give discrete dilution rates in the four sequential vessels of 0.03, 0.015, 0.007 and 0.003 h^{-1}. Due to the lack of sufficient spatial separation of the component physiological groups of the interacting microbial association the above discrete dilution rates were increased first to 0.039, 0.018, 0.08 and 0.04 h^{-1}, and then 0.048, 0.022, 0.010 and 0.004 h^{-1}.

7.3   Results and Discussion

7.3.1   Four-stage Chemostat

Figures 7.1 and 7.2 show the results of one of the arrays (Model 2). For the initial dilution rate regime nitrate and nitrite reduction were effected in the first vessel of the array. In contrast, only limited sulphate reduction resulted while methanogenesis appeared to first increase down the array before decreasing in the final vessel (Figure 7.1).

Perhaps surprisingly, a stepwise increase in dilution rate (Regime 2) had a positive effect on sulphate reduction, with a corresponding increase in methanogenesis also recorded (Figure 7.2).

Figure 7.1   Changes in Methane (% v/v) and Residual Sulphate Concentrations (mg l^{-1}) in the Component Vessels of a Four-stage Chemostat Model Subjected to Discrete Dilution Rates of: 0.03, V1; 0.015, V2; 0.007, V3; 0.003h^{-1}, V4
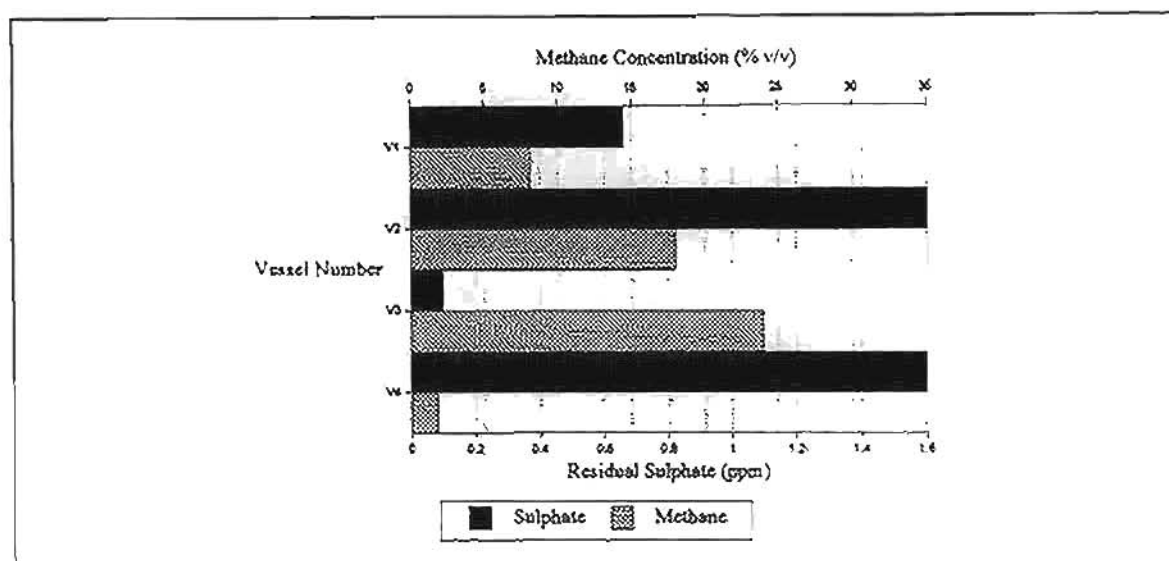
Figure 7.2   Changes in Methane (% v/v) and Residual Sulphate Concentrations (mg l^{-1}) in the Component Vessels of a Four-stage Chemostat Model Subjected to Discrete Dilution Rates of: 0.039, V1; 0.018, V2; 0.008, V3; 0.004h^{-1}, V4
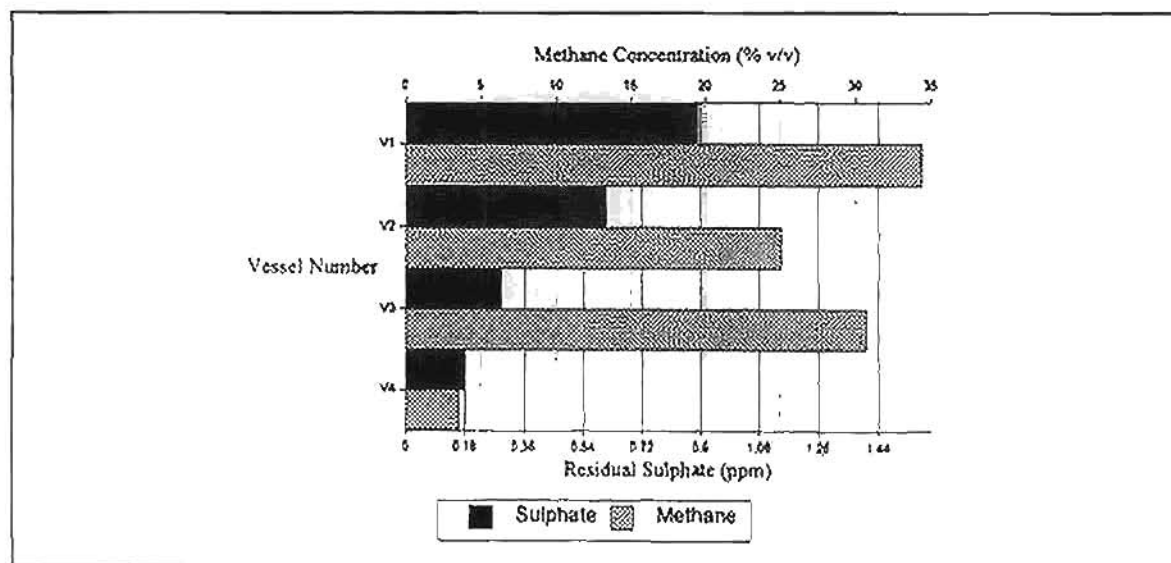
Although the dilution rate regime applied did not effect the planned spatial separation of nitrate reduction, sulphate reduction and methanogenesis further selection

pressure changes could not be examined as an unforseen problem overtook the study. The erroneous supply, by the manufacturer, of LPG in an OFN gas cylinder inflicted a major perturbation to the culture which was not detected for 24 hours. Unfortunately, a major fermentation balance change resulted. Although nitrate reduction proceeded as before, the subsequent reduction of nitrite ceased and concentrations as high as $(2.2 \times 10^{-3} \text{ mol } l^{-1})$ were recorded. Sulphate reduction continued but was consolidated in Vessels 1 and 2 while methanogenesis was suppressed (Figure 7.3).

Although inhibition of sulphate-reducing bacteria, cultured in multi-stage continuous cultures, in response to an added perturbant $(2 \times 10^{-3} \text{ mol } l^{-1}$ phenol) has been recorded (Watson-Craik and Senior 1989d), this is the first instance of a similar fermentation balance change effected by an exogenous gaseous perturbant.

Figure 7.3   Changes in Methane (% v/v) and Residual Sulphate Concentrations (mg l^{-1}) of the Component Vessels of the Four-stage Chemostat Following Perturbation with LPG.
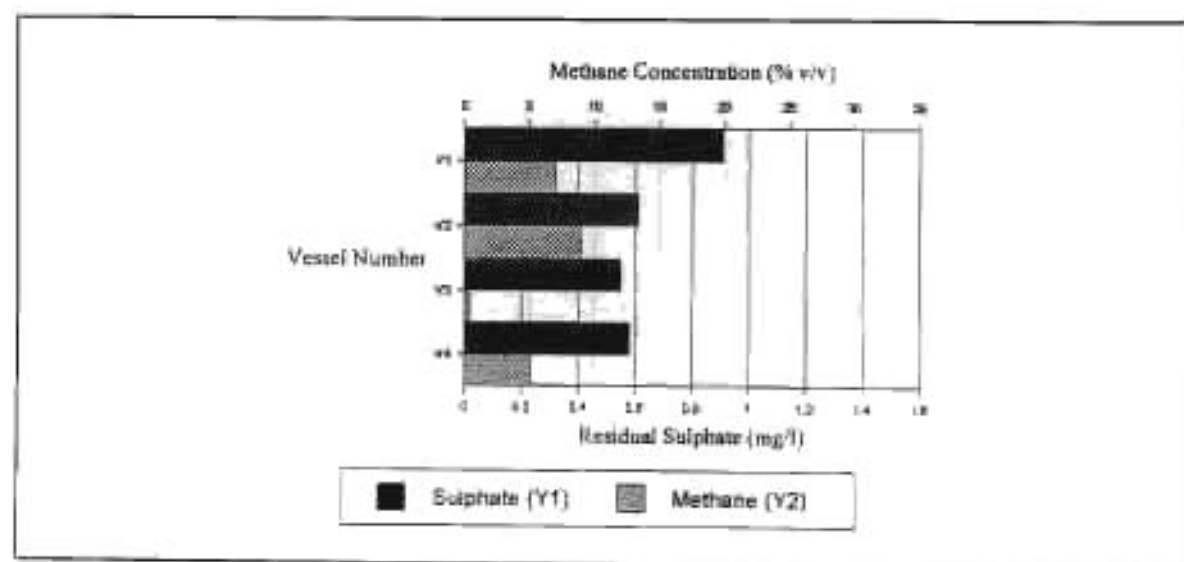
It is, however, known that gaseous products and concentrations of these can mediate changes in the solid-state refuse methanogenic fermentation. For example, Kasali *et al.*, (1990b), examined the effects of fermentation gases (H_2, CO_2, CH_4) on refuse acidogenesis and methanogenesis and showed that increased concentrations of hydrogen mediated a

fermentation balance change such that the concentrations of reduced and oxidised fermentation products increased and decreased, respectively.

In anoxic ecosystems hydrogen plays a pivotal role in regulating fermentation balances (Large 1983; Aragno 1988). The gas is generated via the oxidation of reduced pyridine nucleotides (Buvet 1980; Kasali 1986):

$$NADH + H^- \rightleftharpoons H_2 + NAD^+ \qquad \Delta G^0 = +18 \text{ kJmol}^{-1} \qquad 7.1.$$

Since the equilibrium constant (K_{eq}) for this reaction strongly favours the direction of NADH + H$^+$, rather than hydrogen formation (Large 1983), active populations of hydrogen sink bacteria (nitrate reducers, sulphate reducers and methanogens) are required to maintain a low partial pressure of hydrogen (Kasali 1986; Senior and Balba 1987). Clearly then, if the partial pressure of hydrogen increases due to, for example, inhibition of hydrogen sink bacteria, the flow of electrons (from NADH) shifts from hydrogen production to the formation of fatty acids (Senior and Balba 1984; Aragno 1988).

In the study reported here, no fatty acid accumulations resulted. Thus, in the absence of increased biomass production, but concomitant depressed methanogenesis, hexanoic acid catabolism must have been accounted for by carbon dioxide evolution.

At this stage it was impossible to identify the specific bacteriostatic/bactericidal component(s) of the LPG which contains a range of molecules in different concentrations (Table 7.1). However, one possibility is the relatively high concentration ($\leq 2\%$ v/v) of acetylene. Acetylene (C$_2$H$_2$), like sulphide (HS$^-$), inhibits nitrous oxide (N$_2$O) reductase and, therefore, the reduction of N$_2$O to N$_2$. This inhibition has been shown to be reversible with acetylene concentrations <2% (v/v) while at higher concentrations the blockage is irreversible. Acetylene does not inhibit the onset and progress of denitrification prior to N$_2$O blockage (Paul and Clark 1989). In our study the generation and possible accumulation of nitrous oxide were not monitored although it appeared that any accumulation of N$_2$O inhibited nitrite reduction.

Table 7.1   Typical Composition of LPG, as Determined from the Analysis of 72 Different Batches, with the Concentrations of the Same Molecules Detected in Lansdfill Gas from Six Landfill Sites

Component	LPG (% v/v)[1]	LFG (% v/v)[2]
Methane	< 0.1	-
Ethane	2.3	-
Ethene	< 0.1-1.0	-
Ethyne	< 0.1	-
Propane	48.3	< 0.0051
Propene	3.2 - 45	-
*Iso*butane	7.4	-
Butane	39.6	< 0.003
Butenes	0.8	< 0.0036
Pentanes	< 0.1	< 0.0033
1-3-butadiene	< 0.1	-
Total Acetylenes	< 0.1 - 2	-
Total Dienes	< 0.1 - 10	-
Total Sulphur[3]	60 - 200	1 -195

[1] G.E. Bleimschein (Personal Communication)

[2] U.K. Department of the Environment (1988)

[3] Total sulphur concentrations are recorded as mg Kg^{-1}

Furthermore, although not demonstrated in Furthermore, although not demonstrated in refuse, Reis, Almeida, Lemos and Carrondo, (1992) showed that hydrogen sulphide had a direct and reversible toxic effect on sulphate-reducing bacteria. Therefore, more detailed investigations will have to be made to establish the relative inhibitory effects of acetylene and gaseous sulphur-containing compounds.

The need for an Anaerobic Bioassay Test to assess the impacts of specific perturbants has been highlighted (Blum and Speece 1991; U.K. Department of the

Environment 1994). Current tests based on rates and product concentrations of acidogenesis and methanogenesis (Battersby and Wilson 1989; Azhar and Stuchey 1994) are somewhat limited. Use of methanogens has been justified on the basis of their sensitivities (Jarrell and Saulnier 1987). Increasing evidence, however, shows that other key members of interacting associations may be more vulnerable to specific perturbants than methanogens.

Blum and Speece (1991), for example, examined the toxicity of >50 chemicals to aerobic heterotrophs, *Nitrosomonas* sp. and methanogens. The results showed that the aerobic heterotrophs and methanogens were equally sensitive to non-reactive toxicants while *Nitrosomonas* sp. was much more susceptible. Watson-Craik and Senior (1989d) similarly demonstrated the lower susceptibility of methanogens to phenol than sulphate-reducing bacteria. Further evidence of differential susceptibility was demonstrated in our study with nitrate reducers seemingly unaffected while the nitrite reducers were vulnerable to the bacteriostatic/bactericidal component(s) of the perturbant LPG.

Although the direction of this study dramatically changed as a consequence of the accidental perturbation, two important messages resulted. Firstly, fermentation balance changes may be effected by the components of landfill gas. As a consequence, the components of landfill leachate may change thus altering its environmental impact potential. Secondly, there is still a need for a structured Anaerobic Bioassay Test which is able to differentiate the effects of perturbants on specific physiological groups in interacting microbial associations.

7.3.2 Three-Stage Chemostat

Following the premature termination of the experiment and as a result of the lack of sufficient physiological group separation the model was re-configured as a three-stage chemostat. Three models (Coutts *et al.*, 1987) with component vessel working volumes of 155,331 and 1732 m*l* were built. The first vessel of each array was inoculated and operated as discussed previously (Section 7.2) to give discrete dilution rates in the three sequential vessels of 0.03, 0.015 and 0.003 h^{-1}. The carbon source was also changed and was replaced with butyrate as this electron donor has been shown to facilitate separation (I.A. Watson-

Craik, pers. comm.) To ensure the accurate determination of the methane concentrations, in the respective vessels, the overgassing of the vessels with oxygen free nitrogen was stopped approximately 24 hours prior to analysis.

In the control array, nitrate reduction was exclusively localised in the first vessels with no nitrate or the intermediate nitrite detected in either B_1 or C_3. The residual sulphate concentrations, pH, methane concentrations and VFAs concentrations are given in Table 7.2. From the results it was clear that the processes of sulphate reduction and, surprisingly, methanogenesis occurred mainly in the second vessel (B_1). The degradation of butyrate to methane is only possible in the presence of syntrophic associations of methanogens with hydrogen-producing acetogenic bacteria, since the former do not directly use organic acids higher than acetate (Widdel, 1988). There are two pathways of butyrate dissimilation: β-oxidation to two molecules of acetate; and decarboxylation to propionate and iso-butyrate (Sulisti, 1994). Since, acetate was the only VFA recorded, this suggested that β-oxidation was the operating catabolic pathway as reported by Coutts (1987) and Watson-Craik (1987). Therefore, it seemed probable that, as reported by Watson-Craik (1987) and Sulisti (1994), the following groups of bacteria were responsible for the complete fermentation: a. Acido- and Acetogens; b. Sulphate-reducing bacteria (SRB); and c. Methanogens. The concentrations of VFAs decreased down the array and appeared to be concomitant with pH increases.

Table 7.2   Ranges of Residual Sulphate Concentrations, Methane Concentrations and VFAs Concentrations in the Influent Reservoir and Vessels A_3, B_3 and C_3 of the Three-Stage Chemostat Control.

Vessel	Residual [SO_4] (mg l^{-1})	CH_4 (% v/v)	pH	[Acetate] (mg l^{-1})	[Residual Butyrate] (mg l^{-1})
Influent	109 - 129	—	6.1 - 6.7	0.9 - 1.3	270 - 320
A_3	99 - 116	2.63 - 4.5	6.2 - 7.1	1.7 - 3.7	0.1 - 8.7
B_3	1.9 - 4.6	21.1 - 45.3	7.1 - 7.9	0.002 - 2.1	0.001 - 0.41
C_3	2.1 - 3.7	0.55 - 2.5	7.2 - 7.8	0 - 0.001	0

Subjecting the system to increased, and decreased, dilution rates in addition to various concentrations of the exogenous electron acceptors, nitrate and sulphate, did not effect complete separation of the component physiological groups as evidenced by the majority of methane continuously produced in the second vessel. This was unexpected as Coutts et al., (1987) effected significant separation of the component physiological groups, of a hexanoate-catabolising microbial association, either by elevated sulphate concentrations (0.960mg l^{-1}) or by the imposition of a non-constant dilution rate regime. Further, a similar consolidation of methanogenic activity in the second vessel of a four-stage continuous culture model was recorded by Watson-Craik et al., (1993) although an increase in the dilution rate resulted in significant separation. In the present study, however, a sufficiently high dilution rate, to effect a similar separation of the physiological groups on the basis of their μ_{max} values, was not obtainable. The reason for this was not clear although "pressure locks", which inhibited the flow of medium between the vessels of the model, could have been implicated.

Significant rates of sulphate reduction and methanogenesis, in the same vessel (Vessel B_2), was also unexpected as competition for common substrates, hydrogen and acetate, should, thermodynamically, favour sulphate reduction (Widdel, 1988: Senior, 1990). However, in practice this is not always the case. For example, van Esch, Williams, Jones, Cross and Pohland (1989) reported that there was a five-fold increase in acetate conversion rate when aceticlastic methanogens were cultured in the presence of hydrogenotrophic SRB. Despite the majority of methane being recorded in vessel B_1, the discrete concentrations were, compared to normal anaerobic digestion criteria, relatively low and possibly reflected the competition between the SRB and methanogens. In this present study the relationship between SRB and methanogens could have been non-competitive. Coutts et al., (1987) and Sulisti (1994) reached a similar conclusion that the SRB tended to preferentially utilise hydrogen while the methanogens utilised acetate.

In contrast to the SRB-methanogenic bacteria interaction(s) separation was achieved between the nitrate reducing bacteria, which were mainly localised in the first vessel, and the sulphate-reducing bacteria, which were localised in the second.

Phenol Perfusion

Phenol (10 to 300 mg l^{-1}) was introduced into the experimental vessel (B_2) via the influent tube at the bottom of the vessel. According to Watson-Craik and Senior (1989d) and Sulisti (1994) throughout the duration of their studies, to examine the effects of phenol and *o*-cresol on the microbial interactions by use of continuous cultures, no phenol or *o*-cresol dissimilation occurred. Due to operational problems with the model system similar conclusions could, however, not be made for the present study. Although, according to Coutts *et al.*, (1987), the multi-stage model approximates to a plug flow system, the lack of any mixing in the present study resulted in higher phenol concentrations than the planned maxima being recorded.

Although, in Vessel B_2, no significant inhibition of butyrate catabolism to acetate was recorded with phenol concentrations ≤ 160 mg l^{-1} further increases in the phenol concentrations affected VFAs, and especially acetate, production. For example, following perturbation with 170 and 300 mg l^{-1} phenol the acetic acid concentrations were 1.6 and 22.2 mg l^{-1}, respectively. Methanogenesis, on the other hand, demonstrated enhanced concentrations with phenol perturbations ≤ 50 mg l^{-1} and increasing inhibition with phenol concentrations ≥ 80 mg l^{-1} (Figure 7.4).

Figure 7.4   Changes in Headspace Methane Concentrations (% v/v) in Experimental Vessel B_2 of a Three-Stage Chemostat Model
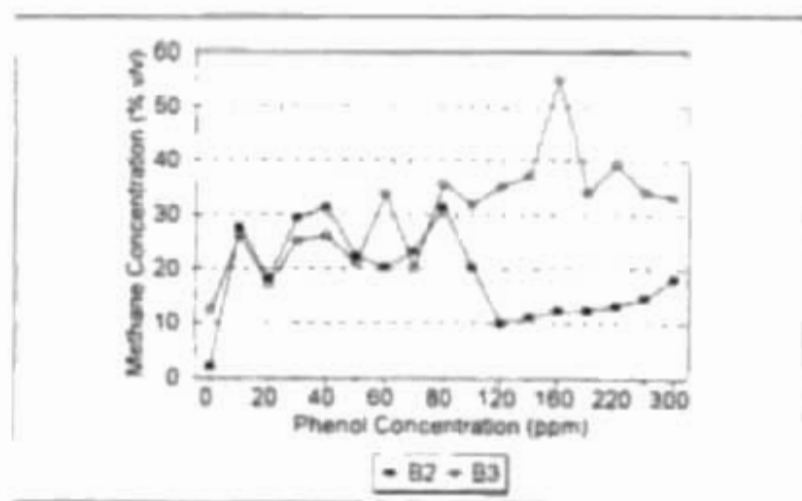
However, increasing the phenol concentrations > 120 mg / l did not appear to effect further inhibition of methanogenic activity. This incomplete inhibition was, however, not surprising as Sulisti (1994) reported that, in a similar multi-stage system, partial methanogenesis inhibition resulted from 10 mM (960 mg / l) o-cresol perturbation while complete inhibition was recorded with a concentration increase to 20 mM (1920 mg / l). The low acetic acid concentration increases, discussed above, could, therefore, have been due to either a decrease in sulphate-reducing bacterial activity or methanogenic bacterial activity, as both groups are acetotrophic (Widdel, 1988).

Another effect of the increasing phenol concentration was a decline and, subsequent, shift of SR bacterial activity. From Figure 7.5 it can be seen that with phenol concentrations ≤ 120 mg / l no significant decrease in SR bacterial activity was recorded in Vessel B_2, which furthermore, exhibited similar residual sulphate concentrations as the control, Vessel B_3. However, with concentrations > 120 mg / l sulphate reduction steadily declined and the residual sulphate concentrations increased, particularly, following the perturbation of Vessel B_2 with phenol concentrations > 160 mg / l.

Figure 7.5 Changes in Residual Sulphate Concentrations (mg / l) in Experimental (B_2) and Control (B_3) Vessels of Three-Stage Chemostat Models Following Phenol Perturbation
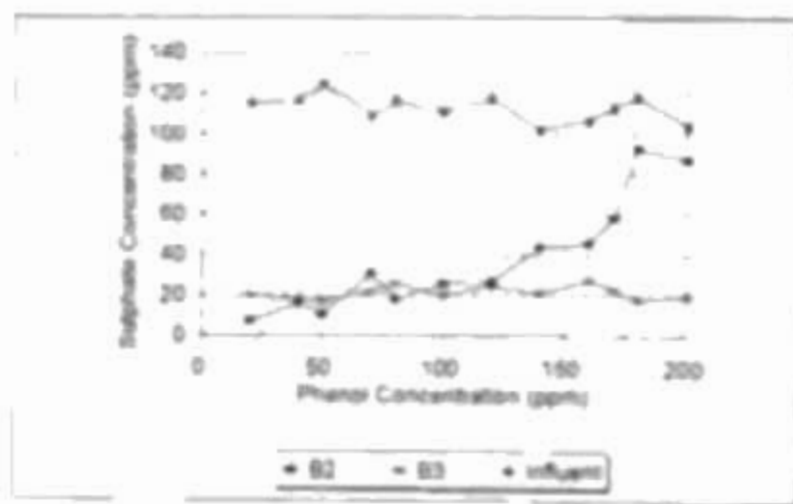
Further, the residual sulphate concentrations in the first experimental vessel (A_2), which was not subjected to phenol perturbation, declined slightly. Thus, prior to, and post, perturbations of > 160 mg l^{-1} phenol, the residual sulphate concentrations were 83 and 107 mg l^{-1}, respectively. Thus, low sulphate reducing activity was now operative in the first vessel. This perurbation (B2) resulted in concentration increases of VFAs. For example, the butyrate concentration increased from 1.8 to 14.1 mg l^{-1} following the shift.

In multi-stage studies Watson-Craik (1987) and Sulisti (1994) showed that the SRB were more sensitive, than the methanogens, to the added xenobiotic (phenol or o-cresol). From the results of the present study it can be concluded that the methanogenic bacteria were partially inhibited by phenol concentrations >80 mg l^{-1} with complete SRB inhibition resulting in the presence of > 160 mg l^{-1}. Possible reasons for the maintainance of methanogenic activity with phenol concentrations > 120 mg l^{-1} could have been due to one or more of the following:

a.   Microbial adaption of the methanogenic microorganisms;

b.   The model relied on dilution of the added, low volume high concentration, phenol. Therefore, in the absence of mixing concentration gradients could have resulted; and

d.   The methanogenic population was not mono-specific and, therefore, the acetoclastic methanogens could have been inhibited while the hydrogenotrophic methanogens were relatively un-inhibited. A similar conclusion was also reported by Watson-Craik (1987) and Sulisti (1994).

CHAPTER 8

8.1   Synopsis and Management Summary

Refuse disposed in landfills passes through sequential stages of degradation before all the labile matter has been catabolised. These stages have been described by Aragno (1988), Barlaz, Schaefer and Ham (1989a) and Senior and Balba (1990) and will not be further discussed here.

The effective anaerobic digestion of refuse depends on the active participation of the component species of the interacting microbial associations. For example, according to Senior (1986), under aerobic conditions mineralisation of organic compounds to CO_2 is often achieved by the intervention of a single microbial species. In the absence of oxygen, however, complete dissimilation, characteristically, requires the co-operative metabolism of a microbial association, each member of which contributes a partial oxidation, until CO_2 and/or CH_4 results. Further, the associations are still strongly influenced, both positively and negatively, by chemical gradients of electron donors and acceptors and hydrogen concentrations.

The component species of anaerobic interacting microbial associations are able to use a variety of inorganic acceptors, such as nitrate, sulphate and carbon dioxide, often in sequence according to the energy liberated from a common electron donor (Barlaz, Ham and Schaefer, 1990; Westlake, 1990). Thermodynamic constraints dictate that the bacteria using the more oxidised acceptors gain a distinct advantage which is usually manifested as a kinetic advantage (Large, 1983; Archer, 1988). Thus, for example, although sulphate-reducing bacteria and carbon dioxide-using methanogens are able to grow under similar ecological conditions, if they are competing for the same limiting substrate(s) (hydrogen and/or acetate) then, theoretically, the former species should displace the latter due to their higher affinities for both substrates (Senior, 1986).

According to Senior (1986), the key to the actual products generated during anaerobic microbial dissimilation of organic molecules is the interactions between different metabolic groups of organisms although examination of the terminal catabolisms only (Figure 8.1) exemplifies the complexity of the situation.

A complete understanding of these interactions is, therefore, an essential pre-requisite to the identification of rate-limiting steps and, thus, optimisation of the process of anaerobic digestion. Therefore, one of the aims of this study was to determine the effects of phenol and/or sewage sludge co-disposal on the component species of the interacting microbial associations.

Figure 8.1   Microbial Interactions in Terminal Anaerobic Catabolisms (Senior, 1986)
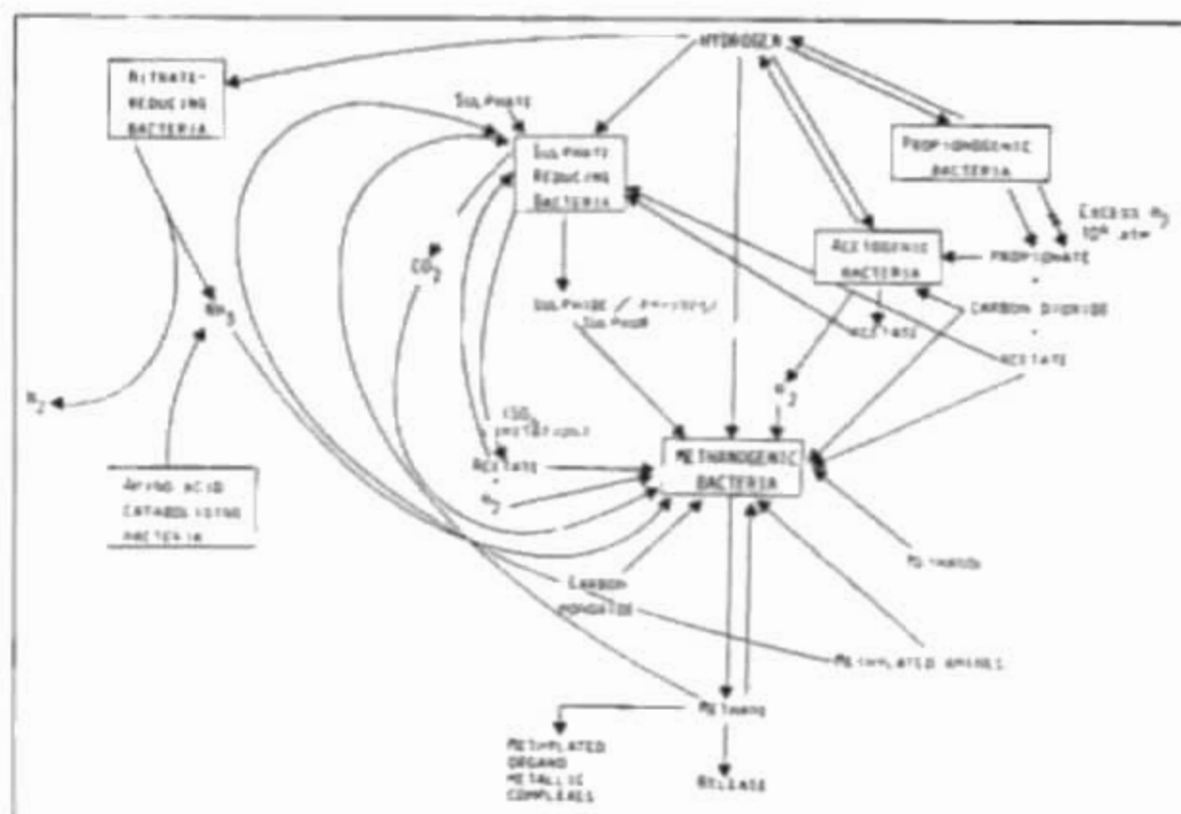
In general, irrespective of the phenol or sludge loading, the experimental refuse columns responded positively to mineral salts additions. The often recorded accumulations, prior to the mineral salts additions, of nitrate/nitrite and sulphate indicated that the nitrate-

and sulphate-reducing microorganisms could have been inhibited by either the added xenobiotic or nutrient limitations *per se*. Inhibition of the nitrate- and, more specifically, nitrite-reducing microorganisms could result in nitrite toxicity (Section 7.3.1). Inhibition of the sulphate-reducing bacteria could also result in an increase of the hydrogen partial pressure which has been shown to inhibit anaerobic digestion (Schink, 1993). Inhibition of these microorganisms could also suppress the lowering of the redox potential which is necessary for strict anaerobes and, thus, efficient anaerobic digestion (Large, 1983). Also, the accumulations of 'total' VFAs, which were often concomitant with low methane concentrations, seemed to suggest that the acidogens as well as the acetogens were less affected by the inhibitory effects of the added phenol and/or nutrient limitations than the methanogens. Alternatively, the methanogens could have been inhibited simply by the lowered pH values and/or 'total' VFAs accumulations. Thus, the phenol may not have had any direct inhibitory effect on the methanogens. This explanation is, however, thought less likely as significant methane concentrations were often recorded concomitant with low pH and/or VFAs accumulations.

Although landfill sites are not usually characterised by nutrient limitations (Senior and Balba, 1990), this study indicated that the addition of mineral salts significantly enhanced microbial activity. It is thought unlikely that this limitation was as a result of macro-nutrient (nitrogen or phosphate) limitations. However, micro-nutrients (trace elements) such as copper and nickel, which are often required for enzyme production, could have been implicated (Schlegel, 1992). In practice, it is extremely difficult to determine the requirements/limitations for/of these trace elements as they are required in micromolar concentrations.

Together with the above factors, the enrichment *per se* of a phenol-degrading population or the selection of phenol-tolerant microorganisms could also have been implicated. Further studies should, therefore, be made to determine the specific enhancing effect(s) of the mineral salts addition and to answer the following questions:

a.   Did the mineral salts addition eliminate a nutrient limitation *per se*?

b. What was the relative buffering effect of the added carbonate on the pH and, thus, the overall functioning of the interacting microbial associations?

c. Did the addition result in the enhancement of nitrate- and sulphate-reducing bacteria which, subsequently, lowered the redox potential sufficiently for effective anaerobic digestion?

d. Could the enrichment of a phenol degrading population and/or the selection of phenol tolerant microorganisms have been operative?

The advantages and disadvantages of co-disposing phenol (1000 and 2000ppm) and/or anaerobically digested sewage sludge with refuse in the presence of different operating regimes are summarised in Tables 8.1 - 8.6.

From the results discussed in this study and summarised in Tables 8.1-8.6 it was apparent that the operating regime employed was a key factor in refuse and phenol degradation although with time, and especially following the phenol resupplementations, the operating conditions played a less significant role.

Despite the higher phenol removal rates exhibited by the single elution microcosms the overall favoured operating method, taking into consideration pH, 'total' VFAs concentrations and methane generation, was the leachate recycle regime. Phenol co-disposal on a batch basis could inhibit refuse and phenol degradation while rain affected refuse could result in rapid washout of the co-disposed compounds.

The sewage sludge co-disposal experiments, likewise, demonstrated that, depending on the sludge:refuse ratio, the operating regime was extremely important in optimising the refuse degradation processes. For refuse subjected to single elution regimes, "high loading" could result in low pH values and inhibition of the refuse degradation while "low load" co-disposal operations demonstrated no significant inhibition. For refuse subjected to leachate recycle the opposite was true with distinct advantages recorded for the "high load" columns. Co-disposal operations under both batch and rain conditions could result in inhibited refuse catabolism.

Table 8.1 Advantages and Disadvantages of 1000ppm Phenol Co-Disposal with Refuse in the Presence of Different Operating Regimes

Operating Regime	Advantages	Disadvantages
Single Elution	High phenol removal rates. Enhanced methane concentrations. Moisture flux through the refuse mass.	Continuously depressed pH values. Rapid washout of the buffering capacity and methane precursors. Continuously elevated residual phenol concentrations in the leachate.
Leachate Recycle	Rapid decrease of leachate residual phenol concentrations. Attainment of neutral pH values. Moisture flux through the refuse mass.	Low phenol removal rates. Logistical and equipment requirements for leachate recycle.
Batch	Rapid decrease of leachate residual phenol concentrations. Elevated methane concentrations.	Low phenol removal rates. Low pH values coincident with increased 'total' VFAs concentrations. No or little moisture flux through the refuse mass.
Simulated Rain	Low leachate residual phenol concentrations. Moisture flux through the refuse	Extremely rapid washout of the buffering capacity, methane precursors and nutrients. Very low phenol removal rates. Continuously suppressed pH values. Low rates of methanogenesis.

Table 8.2 Advantages and Disadvantages of 2000ppm Phenol Co-Disposal with Refuse in the Presence of Different Operating Regimes

Operating Regime	Advantages	Disadvantages
Single Elution	High phenol removal rates. Moisture flux through the refuse mass.	Continuously depressed pH values. Low methane concentrations. Rapid washout of the buffering capacity and methane precursors. Continuously elevated residual phenol concentrations in the leachate.
Leachate Recycle	Decrease in the leachate residual phenol concentrations. Moisture flux through the refuse mass.	Depressed pH values and increased 'total' VFAs concentrations. Low methanogenic activity. Relatively low phenol removal rates.
Batch	Decrease in the leachate residual phenol concentrations.	Depressed pH values and increased 'total' VFAs concentrations. No or little moisture flux through the refuse mass. Low phenol removal rates.
Simulated Rain	Low leachate residual phenol concentrations.	Extremely rapid washout of the buffering capacity, methane precursors and nutrients. Very low phenol removal rates. Continuously depressed pH values. Low rates of methanogenesis.

Table 8.3 Advantages and Disadvantages of "Low Load" Sewage Sludge Co-Disposal with Refuse in the Presence of Different Operating Regimes

Operating Regime	Advantages	Disadvantages
Single Elution	Low 'total' ammonia concentrations. Moisture flux through the refuse mass.	Low pH values and methane concentrations. Increased 'total' VFAs concentrations. Possibility of high numbers of pathogens in the leachate due to washout. Increased volumes of leachate.
Leachate Recycle	Enhanced refuse stabilisation rates. Moisture flux through the refuse mass. Low 'total' ammonia concentrations.	Low pH values and increased 'total' VFAs concentrations. Low methane concentrations and inoculum potential. Logistical and equipment requirements for leachate recycle.
Batch	Increased pH values. Low 'total' VFAs concentrations. Higher methane concentrations and a shorter lag phase.	No or slow moisture flux through the refuse. Brief period of active methanogenesis.
Simulated Rain	Moisture flux through the refuse mass.	Low pH values. Rapid washout of the buffering capacity and nutrient content. Low methanogenic activity.

Table 8.4   Advantages and Disadvantages of "High Load" Sewage Sludge Co-Disposal with Refuse in the Presence of Different Operating Regimes

Operating Regime	Advantages	Disadvantages
Single Elution	Enhanced methane generation and shortened lag phase. Moisture flux through the refuse mass Low 'total' ammonia concentrations.	Low pH values. Increased 'total' VFAs concentrations. Brief period of active methanogenesis. Possible generation of increased volumes of leachate.
Leachate Recycle	Elevated pH values. Higher methane concentrations and shortened lag phase. Low 'total' VFAs concentrations and increased rates of refuse stabilisation. Low 'total' ammonia concentrations.	Loading rates could compromise site working conditions. Logistical and equipment requirements for leachate recycle. Generation of greater volumes of leachate.
Batch	Enhanced methane concentrations and shortened lag phase. Decreased 'total' VFAs concentrations Longer periods of active methanogenesis.	None or slow moisture flux through the refuse mass. Low pH values.
Simulated Rain	Moisture flux through the refuse.	Low pH values. Rapid washout of the buffering capacity and potential nutrients. Low methane concentrations.

Table 8.5   Advantages and Disadvantages of "High Load" and "Low Load" Sewage Sludge and 1000ppm Phenol Dual Co-Disposal with Refuse in the Presence of Different Operating Regimes

Operating Regime	Advantages	Disadvantages
Single Elution	High phenol removal rate. Moisture flux through the refuse mass.	Constantly high leachate residual phenol concentrations. Increased phenol desorption rates. Low pH values and methane concentrations.
Leachate Recycle	Low leachate residual phenol concentrations. Moisture flux through the refuse mass. Increased pH values and methane concentrations. Low 'total' VFAs concentrations.	Low phenol removal rates. Increased rates of phenol desorption.
Batch	Low leachate residual phenol concentrations. Initially low 'total' VFAs concentrations.	Increased rates of phenol desorption. Low pH values and methane concentrations with extended lag phases. Low phenol removal rates. Increased 'total' VFAs concentrations.
Simulated Rain	Moisture flux through the refuse mass. Low leachate phenol concentrations.	Rapid washout of the buffering capacity and essential nutrients. Very low phenol catabolism. Low pH values. Low methane and 'total' VFAs concentrations.

Table 8.6 Advantages and Disadvantages of "High Load" and "Low Load" Sewage Sludge and 2000ppm Phenol Dual Co-Disposal with Refuse in the Presence of Different Operating Regimes

Operating Regime	Advantages	Disadvantages
Single Elution	High phenol removal rates. Moisture flux through the refuse.	Increased rates of phenol desorption. Low pH and methane concentrations. Initially increased 'total' VFAs concentrations. Continuously high residual phenol concentrations.
Leachate Recycle	Increased pH and methane concentrations, especially for the "Low Load" regime. Low leachate phenol concentrations. Decreased 'total' VFAs concentrations. Moisture flux through the refuse mass.	Increased rates of phenol desorption. Low phenol removal rates. Low pH and methane concentrations for the "High Load" regime.
Batch	Low leachate phenol concentrations. Low 'total' VFAs concentrations and enhanced methanogenesis for the "High Load" regime.	No or little moisture flux through the refuse mass. Low pH values. No reduction in the lag phase.
Simulated Rain	Low leachate phenol concentrations. Moisture flux through the refuse mass. Enhanced methanogenesis and elevated pH values for the "High Load" regime.	Rapid washout of the buffering capacity and nutrients. Low pH values. Initially higher 'total' VFAs concentrations. Low methane concentrations.

Irrespective of the sludge and/or phenol loading, for the dual co-disposal of phenol and sewage sludge, the single elution regime exhibited the greatest phenol removal rates but these were, however, coincident with lowered pH values. Further, for both the 1000ppm and 2000ppm phenol perfused "high" and "low" sludge load columns, leachate recycle overall exhibited the most enhanced refuse degradation when pH, methane concentrations and 'total' VFAs were used as the analytical criteria.

The importance of liquid flux has been stressed by the U.K. Department of the Environment (1994) which stated that flushing bioreactor conditions are needed for successful co-disposal. Therefore, in general, despite the phenol and/or sewage sludge loading, compared with the other operating regimes, leachate recycle resulted in higher phenol removal and refuse degradation rates, but did not simultaneously compromise the final leachate quality.

8.2 Code of Practice to Determine the Effects of Co-Disposed Molecules on Microorganisms in Refuse.

The U.K. Department of the Environment has identified a need for an Anaerobic Bioassay Test (ABT) which can be used to assess the fate, and impact, of individual compounds or wastes when co-disposed. This would be particularly important for organic compounds which have not prevously been investigated before. A draft test protocol was, therefore, developed by the U.K. Public Health Laboratory Service and this is illustrated in Figure 8.1. The protocol consists of serum bottles (in triplicate) containing acclimatised sewage sludge which are challenged with different concentrations of chemical wastes. The volume of gas, and the percentage headspace methane concentration, produced by each culture is then monitored on a weekly basis for 28 days. Unfortunately, there are a few problems with this method. For example, sewage sludge from different areas could react differently to the added compound(s). Also, the method does not differentiate between different microbial species. The lack of surfaces could also account for apparent microbial inhibition. It has been demonstrated, for example, that, unlike the free-living microorganisms, attached microbial associations are often able to degrade increased

Figure 8.1 Flow Diagram of the Suggested Liquid Anaerobic Bioassay Test Protocol (U K Department of the Environment, 1994)

Acclimation bottle	fresh filtered sewage sludge
	+ 50 50 (w/v)
	medium (autoclaved & reduced)

Incubate 2-3 days (37°C)

Test bottles	80 ml medium (sterilised)
	-
	0 8 ml "reduced vitamins" solution
	+
	20 ml acclimated inoculum
	(leave 3 bottles without sludge as chemical - medium controls)

Incubate 7 days (37°C)

Gas volume & methane analysis

(replace any unsuitable bottles)

Add chemicals/waste to test bottles (in triplicate) after flushing each bottle headspace with anaerobic gas mixture

A	B	C	D	E	F
control sludge only 1/250	medium + waste	reference compound (If req)	waste dilution	waste dilution eg 1/500	waste dilution eg 1/400 e g

Incubate 37°C

Weekly volume & methane analysis

Day 28 plot data & assess results

Longer-term monitoring for acclimatization as required

concentrations of xenobiotics without visible signs of inhibition (Bettman and Rehm, 1984). Further, the question should be asked is methanogenesis inhibition important as long as the xenobiotic is degraded and there is no significant negative impact on the generated leachate? An increased rate of methanogenesis is not necessarily indicative of catabolism of the co-disposed molecule and could be simply due to electron direction away from inhibited sulphate reduction.

In an endeavour to overcome some of these problems a draft protocol for a solid-phase landfill system has also been suggested and a flow diagram for the schedule is illustrated in Figure 8.2.

Although the solid-phase model configuration is more representative of a landfill environment it also has limitations. For example, despite the use of refuse this experimental protocol does not consider the different co-disposal practices operative at sites. This present study clearly exemplifies that, together with hydrualic and organic loading rates, the methods of operation are extremely important. Therefore, the subtle differences determined by monitoring the generated gases only might be overshadowed by variations due to different operating regimes. Also, the possible inhibition of microorganisms other than the methanogens is not considered. The protocol calls for the incubation of the reactors for four weeks prior to perturbation with the chemical/waste. Therefore, the relative effects on the catabolic events which precede methanogenesis, such as acidogenesis, may be minimal.

As a result of the complex processes in a refuse mass (Section 1.8.4), and the bi-directional interactions between the microorganisms and the co-disposed molecule (Section 8.1), it is recommended that a full co-disposal sudy should be undertaken. Initially, however, the procedures described in Figures 8.1 and 8.2 could be carried out, to gain first approximations of the microbial effects resulting from the co-disposal of the compound in question. Following this, the results obtained can be consolidated by further in depth study. This encompasses, firstly, refuse microcosms challenged with different concentrations of the compound as well as simultaneously subjecting the columns to various operating regimes which are likely to be encountered in a landfill site.

Figure 8.2   Flow Diagram of the Suggested Solid-Phase Model Landfill System

Collect landfill material from cores drilled into sites.
(Store under nitrogen, in cold and dark)

Sort and cut-up landfill material
(under nitrogen)

Seal reactor containing 2 kg methanogenic
landfill refuse plus one litre water (under
nitrogen

Incubate 37°C

Gas volume and methane analysis

(Weekly readings)

Select actively gassing reactors
(After approx 4 weeks incubation)

Add chemicals/waste to reactors
(preferably with replicates)

A	B	C	D
control	reference	waste	waste
reactor	compound	dilution	dilution
	(If req)	eg 1/100	eg 1/500

Incubate: 37°C

1   Weekly volume and methane analysis;
2   Leachate sampling, according to when chemical
     additions are made, for pH, VFAs, BOD and parent
     compound; and
3   Plot data and assess results.

Monitor for a minimum of 8 weeks after
additions made

Long-term monitoring for acclimatization as

This phase of the study is esential to determine not only the effective organic and hydraulic loading rates but also the most efficient method of operation. Further, the relative effects of this practice on different refuse types/compositions and ages can be determined. The possible need for additional inocula or extended acclimation periods as well as the effects of nutrient limitations and/or supplementations can be ascertained. Apart from attaining all the above data, refuse microcosms are also able to determine the effects of numerous applications of either single or mixed compound(s) in the long term. Therefore, the data obtained from this phase of the study can be used to evaluate the effectiveness of co-disposing molecules singly which, in practice, may identify the need for discrete areas of a landfill site being reserved for the co-disposal of certain molecules.

Secondly, the relative effects of co-disposal practices (ie. organic and/or hydraulic loading rates) on the different component species of the interacting microbial associations should be determined by use of multi-stage models. Multi-stage models have, following microbial population stabilisation, numerous advantages such as simplicity of operation and rapid attainment of results. Further, provided sufficient stages have been included, the model can be simultaneously challenged with different concentrations of the xenobiotic. Clearly, the system also lends itself to long-term research as once the microbial association has established the effects of many different compounds can be determined over time.

The Anaerobic Bioassay Test, as decribed above, was designed for single target species which is clearly not the situation in a co-disposal landfill site (Section 1.6). The multi-stage system, on the other hand, can be used to determine the combined effects of two or more xenobiotics on the growth rate-dependent microbial population of a landfill site.

It is also recommended that when a wastewater, rather than a single xenobiotic, is co-disposed, at least one complete GCMS analysis should be made to indentify the key microbicidal/microbiostatic molecule(s). Finally, due consideration should be given to increased stress on any landfill gas extraction system resulting from possible enhanced methane production as a result of co-disposal practices.

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