Physico-chemical characteristics and biodegradability of contents of ventilated improved pit latrines (VIPs) in eThekwini Municipality

by

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Abstract

Lack of access to basic sanitation and water by rural and dense peri-urban communities in South Africa has necessitated the introduction of ventilated improved pit latrines (VIPs). However improper use and inadequate maintenance of these facilities could lead into system failure or to pits filling up faster than they are designed to do. VIPs present a problem once they are full. Pit emptying is an enormous challenge, particularly in communities where there is no space to dispose of pit contents on site. Pit latrine contents poses environmental and health risks since sludge may contain organic pollutant compounds and pathogenic micro-organisms. Contamination of surrounding ground- and surface water is also highly likely. Increased understanding of the biodegradative processes occurring in VIP pits will facilitate better management of pits during their lifespan and better handling of the pit contents upon emptying. These were the needs which drove the present study into the potential biodegradability of the contents of VIPs and the physico-chemical characterization of VIP contents and fresh faeces.

It was expected that user practices such as use of the pit for disposal of wastes other than faeces, and addition of chemicals alleged to reduce malodour from the pit would strongly affect the physical properties of contents from pit latrine. Therefore user practices related to pit functioning were investigated at the outset of the study, through an informal questionnaire survey.

The informal questionnaire survey provided a qualitative insight into VIP use. The different practices surveyed included dumping of household wastes, throwing grey water and also tap water into the pit, and the addition of chemical or commercial additives into the pit. These practices are considered as significant factors contributing to the variation in the studied properties of pit latrines contents, and to the overall function of VIPs.

In the laboratory-based component of the study, fresh faeces and samples of pit latrines contents from a number of locations within eThekwini Municipality were analysed for anaerobic biodegradability using a modified serum bottle test. All samples were also analysed for physico-chemical characteristics, including total and soluble COD, moisture, total solids, and organic solids.

Serum bottle test results for fresh faeces showed an average anaerobic biodegradability of COD of 70 % (n=5) at 95 % confidence. On the other hand, biodegradability tests on pit latrine contents produced less methane, relative to total COD and organic solids content of samples, which was understood to indicate an inhibition of anaerobic digestion. The biodegradability results for pit latrine material at showed that there was no significant difference (p<0.05) in anaerobic biodegradability at different depths, and that overall gas production was very low. This was unexpected and it was concluded that the serum bottle test was inappropriate for evaluating the biodegradability of VIP sludge samples. It is recommended that future studies use an aerobic biodegradability test to test the biodegradability of samples.

Despite the limitation of the poor results for biodegradability, it was possible to use results from physico-chemical analyses to develop a theoretical model of biological activity in the different layers in a pit latrine. In accordance with with model, faecal sludge in a pit can be divided into four layers, showing decreasing biological activity with decreasing depth. Biological activity in the top layer is considered to be aerobic, while that in lower layers is considered to be anaerobic This information can be used as a background to assess the feasibility of different management options for filling pits and different disposal possibilities for pit latrine contents. This is because feasibility of treatment depends on the inherent ability of the treatment processes to accept the load of solids and organic material in the VIP sludge, the residual biodegradability of the VIP sludge, and the health risks associated with handling the sludge.

Declaration 1- Plagiarism

The experimental work presented in this dissertation was carried out at the Pollution Research Group Biochemistry Laboratory School of Chemical Engineering for the School of Biological and Conservation Sciences, University of KwaZulu-Natal, Durban, South Africa, under the supervision of Dr Nicola Rodda, Katherine Foxon and Prof. C.A. Buckley.

This study represents original work by the author and has not otherwise been submitted in any form for any degree or diploma to any tertiary institution. Where use has been made of the work of others, it is duly acknowledged in the text.

Declaration 2-Publication and Presentation

The following publication forms part and / or include research presented in this Dissertation:

Publication 1-WRC report entitled "Scientific Support for the Design and Operation of Ventilated Improved Pit latrines (VIPs) and the Efficacy of Pit Latrine Additives". (2008). CA Buckley, KM Foxon, CJ Brouckaert, N Rodda, C Nwaneri, E Balboni, A Couderc and D Magagna.

Publication 2-Paper entitled "Biological degradation process within pit Latrines"CF Nwaneri, KM Foxon, BF Bakare and CA Buckley. Presented at the Water Institute of South Africa Biennual Conference, Sun City, South Africa, May, 2008.

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1 Introduction

Lack of basic sanitation and access to clean water supply is a major cause of diseases and infant mortality in developing countries (WHO, 2004). Worldwide, approximately 2.6 billion people lack improved sanitation and the largest part of these reside in Africa and Asia (WHO, 2004). Sanitation provisions that are connected to a public sewer, a septic system, a pour flush latrine and even simple or ventilated improved pit latrines (VIPs) are all considered by the World Health Organisation (WHO) as improved. But conventional pit latrines that are provided in most regions usually become sources of pollution to groundwater (Winblad and Simpson-Hébert, 2004). This indicates that the improvement of sanitation is an area that requires urgent attention. Consequently, around the world, there is a drive to ensure the provision of proper sanitation and access to clean water supply. In line with this, the Millennium Development Goals (MDGs) set a target to reduce the number of people with no access to basic sanitation by 50 %, by 2015 (Eales, 2005).

In South Africa, problems of sanitation and inaccessibility to potable water prevail. The Department of Water Affairs and Forestry (DWAF) is tasked by government to ensure that South African citizens gain access to clean water and safe sanitation while ensuring sustainable and efficient management of resources. Substantial advances have been made to ensure access to basic water and sanitation services for all citizens while ensuring environmental sustainability (Anon, 1994; 1998). The proportion of people with improved sanitation has increased by 20 % between 1994 and 2005 (Eales, 2005).

According to the Strategic Framework for Water Services (DWAF, 2003), a target has been set to ensure access to improved sanitation for all by the year 2010. This means that the goals will be achieved before the deadlines for the MDGs by 2015.

In South Africa, many Ventilated Improved Pit latrines (VIPs) have been implemented. EThekwini Municipality is centred on the South African coastal city of Durban, with approximately 3 million inhabitants (Eales, 2005). An estimated number of 100 000 Pit latrines are sited within the Municipality boundaries. VIPs are nationally recognized as the minimum level of acceptable sanitation in South Africa (Eales, 2005).

However, VIPs do not provide improved sanitation when they are full. Theoretically, the rate of stabilisation (biological conversion of large organic constituents to their simplest biologically inert form) or leaching occurring within the pit should equal the pit filling rate. But decreasing degradation rates as the pit fills results in pits filling up faster than degradation occurs. Degradation rates decrease for a range of reasons, including increased proportion of inert material, dilution of the biologically active fraction by the addition of water or household wastes, and introduction of biocidal matter into the pit. Understanding of conditions which affect the rates of biological degradation of pit contents remains limited. Full pits often present problems such as difficulties in relocating the superstructure and difficulty of access to pit, leading to a need for manual emptying. Manual emptying is difficult, hazardous and unpleasant. Once pits have been emptied, there remains the problem of disposing of the pit contents, which are typically unsuitable in unprocessed form for any of the usual domestic waste disposal routes (*e.g.* wastewater treatment, landfill).

Therefore the problem of providing improved sanitation is more complex than simply building new VIP latrines. Sanitation services must also consider means of emptying or replacing full pits, and this has been found to be an enormous challenge, particularly in communities where there is no space to construct new pits or to dispose of contents on site.

Many of the pits located within eThekwini Municipality are full and pose a threat to the health of the public, and to the surrounding ground and surface water (Eales, 2005). Consequently, as part of free basic sanitation services, the Municipality is developing a pit emptying service. The Municipality has estimated the cost of emptying of pit latrines at an average cost of R1 100 per pit, with none of the cost being borne by the householder (Eales, 2005). This cost to the Municipality is substantial, considering the number of pits that have to be emptied. Thus, to limit expenses, eThekwini Municipality, together with Water Research Commission, is interested in evaluating whether it is possible to reduce the rate of filling, and thereby to reduce the frequency of emptying and the associated cost.

As part of ongoing research into providing sustainable improved sanitation services, especially to households outside existing sewer networks, eThekwini Municipality together with Water Research Commission, supported research into the processes and nature of material found in pit latrines. This research endeavoured to provide information and decision support for managing pit latrines during their normal lifespan, and for managing pit emptying and associated sludge management (Buckley *et al.*, 2008)

Consequently, the Pollution Research Group (PRG) of the University of KwaZulu-Natal (UKZN) undertook to investigate VIPs around eThekwini Municipality. Part of this research aimed to address gaps in the understanding of conditions and biological processes occuring in VIP pits, particularly with regard to environmental conditions and user behaviours prevalent in areas served by eThekwini Municipality.

With background understanding of the processes occurring inside the pit, better methods can be devised to improve pit lifespan and to handle pit contents safely when pits are emptied. Both of these pit management problems relate to the residual biodegradability and organic load of the VIP sludge.

The major focus of this study was therefore to obtain information on the stabilisation processes of VIP wastes (pit latrine faecal sludge), as a source of baseline information for sludge management and disposal. The approach devised was to obtain scientific understanding of the biological conversion of the pit latrine faecal sludge by measuring physico-chemical properties and anaerobic degradability of pit contents at varying depths.. The objective was to relate these characteristics of the pit latrine contents to their extent of biological stabilisation.

The first set of VIP sludge samples investigated in this study were sampled from a VIP latrine located in the Tongaat area, north of Durban, serving as a pilot analysis. Subsequent in-depth laboratory analyses were carried out using pit latrine faecal sludge samples from 16 pit latrines from other locations that were within eThekwini Municipality. Part of the analyses was carried out in conjunction Mr Babatunde Bakare of the PRG at UKZN and will appear in his PhD thesis entitled "Scientific support for the design of ventilated improved pit latrines (VIPs)".

1.1 Overall project aims

The major aims of this study were to:

- Obtain baseline information on the overall function of VIPs and on the physicochemical and biological nature of VIP contents.
- Develop a theoretical description of the biological processes that occur in VIPs.

1.2 Specific objectives

Specific objectives were to:

- Conduct a household questionnaire survey to identify user practices that cause the failure of VIPs.
- Measure selected physico-chemical characteristics of samples of VIP sludge from varying depth in a number of VIPs
- As part of measurement of physico-chemical characterisation of VIP sludge, develop a methodology for measurement of soluble organics and determine the relationship between soluble organics and mositure at varying depths in a number of VIPs.
- Measure the anaerobic degradability of samples of VIP sludge from varying depths in a number of VIPs.
- Infer the biodegradation processes occurring in ventilated improved pit latrines, from the physico-chemical characteristics and biodegradability data.

Failure of a pit latrine indicates that the latrine is no longer able to serve the sanitation needs of the owners. This may be because the pit is completely full and therefore no more materials can be added; or for a partially filled pit, some other factor that renders the pit unusable. Human factors are the major contributor to VIP failure. A household questionnaire survey investigated how human factors contribute to pit latrine failure.

Physico-chemical characteristics of pit latrine sludge, measured at varying depths, were total COD, soluble COD, moisture, total solids, and organic solids. Anaerobic biodegradability was measured to describe the extent of biological stabilisation of samples.

The direction of water movement within the pit was initially considered important to the description of biological activity. Theoretically, micro-organisms break down the organic matter in a composite waste into biodegradable and non-biodegradable fractions. The readily biodegradable (RBCOD) materials, which normally are soluble, mostly seep away into the surrounding soil together with available water in the pit. The remaining RBCOD is utilized by micro-organisms for energy and cell multiplication. Therefore soluble CODwas measured at varying depths in pits and was correlated with the direction of water movement within the pit, measured as moisture content.

Finally, the biological processes occurring in VIPs were described in terms of the type of biological conversion of the biodegradable organic component of the pit latrine material that occurred from the time of addition of the material to the time of sampling from a particular layer or depth within the pit. These descriptions were synthesised into a theoretical model of biological activity in VIPs.

1.3 Hypothesis

To fulfill the objectives of this study, the following hypotheses were proposed:

- On-site failure of ventilated improved pit latrines is a result of user behaviourrelated practices.
- The layers in the pit latrine which show the greatest concentration of soluble organics describe the direction of water movement inside the pit.
- The serum bottle test technique can be used to measure the biodegradability property of both fresh faeces and pit latrine faecal sludge.
- Measurement of physico-chemical and biological characteristics of VIP sludge and fresh faeces permits the extent of biodegradation that has occurred in a pit latrine faecal sludge to be deduced.

1.4 Methodology

The methodology of this study was to:

- Collect and synthesise literature from local and international sources to describe the processes occurring in pit latrines.
- Obtain information on the daily operation of pit latrines from users through an informal questionnaire survey.
- Determine the initial physico-chemical properties of fresh faeces, and pit latrine faecal sludge sampled from different pit depths.
- Carry out serum bottle tests on fresh faeces to determine its anaerobic biodegradability.
- Carry out serum bottle tests on pit latrine faecal sludge contents sampled from different pit depths to determine its anaerobic biodegradability.
- Compare susceptibility of fresh faeces and VIP sludge to anaerobic biodegradation through the analysis by mass balances of simple physical and chemical parameters before and after serum bottle tests..

In this study, the methodology for determining biodegradability described by Owen *et al.* (1979) and Remigi and Buckley, (2005), was applied to deduce the extent of anaerobic biological conversions that occurred in pit latrine faecal sludge sampled from

different layers within the pit. The efficiency of the serum bottle test in measuring the anaerobic biodegradability property of both fresh faeces and pit latrine faecal sludge was tested.

Also in this study, a COD fractionation technique was developed, and was used to quantify the COD of soluble and particulate organic material fractions in pit latrine faecal sludge from the different depths and fresh faeces. The soluble COD content of VIP sludge samples from the different depths within the pit were compared. For fresh faeces, the soluble COD measurement was used to estimate the fraction of fresh faeces that is readily biodegradable.

The characteristics of fresh faecal material were compared to those of VIP sludge. Through the interpretation of the physico-chemical analysis data, a theory describing the processes that occur in the pit latrine was developed.

Limitations of applying the serum bottle technique to the samples used in this study were highlighted. Aerobic biodegradability tests were recommended as an alternative method for measuring the biodegradability of the samples.

Finally, some conclusions were drawn from the results gathered regarding the nature of pit latrine contents and the significance of the results in design and operation of pit latrines and management of sludge from pit latrine emptying operations. Also recommendations were made where further research is necessary.

1.5 Dissertation outline

The outline of the Dissertation is as follows:

INTRODUCTION (CHAPTER ONE)

This chapter outlines the context of the study, including presentation of study aims and objectives. The hypotheses of this study are presented. Methodology used is briefly outlines and the structure of the dissertation is presented.

LITERATURE REVIEW (CHAPTER TWO)

The literature review is divided into four sections. Section 2.1 describes VIP construction. Section 2.2 presents a description of physical, chemical and biological nature of the waste contents that are found within a VIP pit, based on published literature. Section 2.3 and 2.4 presents background information on the theoretical concepts applied in this study.

MATERIALS AND METHODS (CHAPTER THREE)

This chapter presents the experimental plan used in this study. The different approaches used in this study are presented. This includes a description of the study area, (including the population that was studied), site visits (which includes questionnaire interviews, sampling procedure) and laboratory analysis (which includes the protocol for running experiments, the reasoning behind experiments and the procedure for the physico-chemical and biological characteristics of samples). The experimental protocol developed to quantify COD fractions and biologradability of the different samples is presented

RESULTS AND DISCUSSION (CHAPTER FOUR)

In this chapter, the results from the questionnaire survey, physico-chemical characterization and biodegradability assay of samples are firstly presented and afterwards discussed. To discuss the results, they are compared with results from past related research.

 DEVELOPMENT OF A MODEL DESCRIPTION OF BIOLOGICAL ACTIVITY IN THE DIFFERENT LAYERS WITHIN THE PIT (CHAPTER FIVE)

In this chapter, a description of biological activity in the different layers in the pit latrine sludge heap is developed through the interpretation of the physical, chemical and biodegradation data.

 GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS (CHAPTER SIX) In this chapter, some conclusions are made about the significance of the results gathered to the design and operation of pit latrines and to the management of sludge from pit latrine emptying operations. The limitations of this study were highlighted, and as well the generalisability of the findings of this study was evaluated Also recommendations for further research are highlighted.

2 Literature review

VIPs are acknowledged as the minimum level of acceptable sanitation in South Africa (Eales, 2005). Many VIPs have been installed countrywide, including in eThekwini Municipality, but without a formal scientific understanding of the operation of the system. For a pit latrine to be eligible to be a VIP, there are basic requirements that have to be met. These requirements distinguish VIPs from conventional pit latrines.

2.1 Description of a ventilated improved pit latrine

A VIP is an improved conventional pit latrine, constructed to remedy the problems associated with the conventional pit latrine such as offensive odour and breeding of flies. It is a type of waterless toilet system developed to resolve the problems of sanitation due to the unsuitability of the flush toilets in water scarce areas, and amongst low income communities in rural and dense peri-urban areas where insufficient water supply and total absence of sanitation prevails (Mara, 1984).

2.1.1 VIP construction

The VIP system differs from conventional pit latrines because it consists of a dignified enclosed brick structure, concrete cover slab and pedestal, door for privacy, light exclusion (to prevent flies), a pit with a cover, a ventilation pipe with fly screen leading from pit to above the level of the superstructure, and a hand washing facility.

The vent pipe eliminates odours, vents gas and prevent flies. As wind passes over the top of the vent pipe, it causes air to flow from the pedestal, into the pit and then up through the pipe to the atmosphere. Continual flow of air removes unpleasant odours and gas is vented through the vent pipe (Bester and Austin, 2000). The fly screen attached to the vent pipe prevents flies from leaving and entering the VIP. Flies that are attracted by the odours leaving the pits are prevented from coming in by the screened vent pipe. Flies already inside the pits are attracted to the light coming from the vent pipe and thus they become trapped by the screen (DWAF, 2003).

The brick superstructure gives privacy to the user, and also prevents flies from leaving the pit. The cover slab is mostly made of concrete. Wood can also be used, especially where a household cannot afford concrete. The cover slab supports the pedestal and superstructure, and acts as a barrier to prevent the users from coming into contact with the wastes. Human excreta are collected inside the pit. The liquid fraction seeps away into the surrounding soil, and the solid bulk is decomposed. Gaseous products of the decomposition process leave the pit through the vent pipe. Dissolved solids are likely to undergo oxidation or seep away from the pit along with the liquids (Mara, 1984).

2.1.2 Problems associated with the use of VIP latrines

Logistical constraints are encountered while using VIPs. These problems usually arise as a result of user practices, improper construction of VIPs, the terrain, soil structure and water table of the site where pit is located, or the leaching capacity of the pit base or wall. The soil structure and water table location affects hydraulic movement within the pit, and must be taken into consideration together with the pit depth when siting a VIP. Factors that affect hydraulic movement within the pit are presented in Section 2.3.2 of this chapter. The problems associated with the use of VIP latrines are presented in this section.

Ground water pollution

Ground water pollution is a major problem that is associated with the VIP toilet system. It may arise as a result of pit flooding. VIPs pose threats to surface water and groundwater quality especially during the rainy seasons. Polluted water is a transmission medium and breeding site for harmful organisms, and can cause disease outbreaks (Chaggu, 2004; Winblad and Simpson-Hébert, 2004). A high water table may result in pit flooding; if pits are not lined, water flows in and out of the system, making pit latrines a potential source of surface and ground water pollution (Chaggu, 2004). The depth of the pit should be such that it is not close to the water table (Chidavaenzi *et al.*, 1997).

Frequency of pit emptying

Pits may require frequent emptying if they fill up rapidly. This problem is associated with the leaching capacity of the pit wall and pit base. In the course of the decomposition processes that occur inside the pit, solids accumulate at the bottom of the pit. This results in blocking of the soil pores at the pit bottom with time. Eventually, permeation (infiltration) of water will occur, mainly through the sides of the pit. At the same time as sludge accumulation progresses, the remaining infiltration area will reduce and the pit will fill up faster (Chaggu, 2004).

Construction problems

The superstructure and substructure of a VIP should be firmly constructed to prevent collapse of the toilet. Any cracks and crevices between the blocks and slabs must also be firmly sealed. It is important that the vent pipe should be painted black and placed outside the superstructure to ensure heating. The vent pipe must be fitted with a fly screen at the top to prevent flies that may have entered into the pit from escaping (Buckley *et al.*, 2008). In addition the VIP should be constructed in such a way as to facilitate emptying when it is full (Buckley *et al.*, 2008).

Properly constructed VIPs may serve to protect the environment and humans by breaking the disease cycle and should function well (Buckley *et al.*, 2008). However, in most cases, these toilet systems are not properly constructed. Technology limitations are a vital contributor to system failure.

2.2 Description of ventilated improved pit latrine waste contents

Available literature on the nature of VIP waste is limited, but some information on the rate of pit filling is documented (Still, 2002). This section presents the literature description of the physical, chemical and biological characteristics of VIP waste.

If well operated, the pit contains faeces, urine, anal cleansing material and/or anal cleansing water (Buckley *et al.*, 2008). A pit should end up with stable sludge material, at the time when it is full and have virtually no volatile fatty acid (VFA) content at the time of emptying (Chaggu, 2004). However, disposal of multiple wastes into the pit results in non-homogenous properties of pit contents. Additives expected to reduce smell or enhance biological processes may also be added into the pit.

2.2.1 Fresh faeces

Faeces are the major feed material that goes into the pit. Studies by Lopez Zavala *et al.* (2002; 2004a; 2004b), characterising faeces and describing the biodegradability of organic matter present in faeces, showed that 80% of human faeces comprises slowly biodegradable organic matter (X_S) whereas only 20% is inert material (X_I). Readily biodegradable organic matter (S_S) was not regarded as a component of faeces (*i.e.* $S_S=0\%$). Furthermore, model results showed that only 15% of the slowly biodegradable matter was easily hydrolysable (X_{Se}) whereas 65% was slowly hydrolysable (X_{Ss}) (Lopez Zavala *et al.*, 2004a). Human faeces are high in organic matter, contributing about 44% of the COD load in domestic wastewater (Almeida *et al.*, 1999). The slowly

biodegradable portion cannot be utilised directly by micro-organisms and so has to be made accessible through extracellular hydrolytic (enzymatic) reactions (Lopez Zavala *et al.*, 2004a). This occurs when micro-organisms come into close contact with the slowly biodegradable organic substrates. Micro-organisms secrete hydrolytic enzymes to hydrolyse biological polymers to simpler molecules which can be absorbed and utilised in microbial metabolic processes. The kinetics and method of biodegradation of faeces can thus be related to its hydrolysability.

The composition and characteristics of human excreta (faeces and urine) are presented in Table 2.1. It is important to note that the composition and characteristics of faeces are influenced by the diet, health and age of individuals (Lopez Zavala *et al.*, 2002; Buckley *et al.*, 2008) and data are therefore dependent on the source population.

Parameter	Sources						
	Gaillard (2002	Lopez Zavala et al.,	Almeida <i>et al</i> ,				
	cited in Chaggu, 2004)	(2002)	(1999)				
Moisture	-	81.8%	79.2 %				
TS	-	18.2 %	20.8 %				
VS	-	84.4%	-				
Total COD	0.57 mg /mg	1.45 mg/mg	1.38 mg/mg				
Dissolved COD	0.09 mg /mg	-	-				
Suspended COD	0.46 mg /mg	-	-				
VFA	8.46 g COD/l	-	1.5 g COD/l				
T-N	17.82 mg N/g	60.1 mg N/g	-				
NH ₃ -N	-	3.4 mg/g	7.2 mg/g				
NO ₃ ⁻ -N	-	0.03 mg/g	0.14 mg/g				
рН	-	7.5	-				
SO4 ²⁻	-	1.1 mg/g	-				
Cl ⁻	-	4.2 mg/g -					

Table 2.1. Characteristics of human excreta

Where: TS= Total solids; VS=Volatile solids; VFA= Volatile fatty acids; T-N = Total Nitrogen; NH₃-N = Ammonia-Nitrogen; NO₃⁻-N = Nitrate-Nitrogen; SO₄²⁻= sulphate and Cl⁻ = Chloride;

2.2.2 Pit latrine wastes

The composition of faecal sludge collected from pit latrines and raw fresh faeces showed higher values for for faeces than for the pit latrine sludge for all parameters and characteristics (Table 2.2). This implies that faeces undergo a certain degree of decomposition while they are inside the pit. Seasonal shifts in temperature and humidity are expected to alter both the physical and biochemical characteristics of faecal matter that has been deposited into a VIP pit. Consequently, the properties of faeces inside the pit change with time as a result of change of factors such as moisture, temperature, carbon content and nutrient availability (Nordin, 2006).

Table 2.2 Typical values of physical and chemical characteristics of freshexcreta, and faecal sludge from a pit latrine. (Adapted fromSANDEC, 1997).

Pit Latrine sludge	Fresh excreta (Faeces and Urine)
8	45
90	110
5	10
0.15	0.20
20,000 - 50,000	-
5:1	-
	Pit Latrine sludge 8 90 5 0.15 20,000 – 50,000 5 : 1

Chaggu (2004), reporting on the faecal component from a urine diversion toilet, found that COD fractions at various points of faecal collection in the pit fluctuated all through the filling period. It may be assumed that similar effects would be observed in VIP sludge.

Studies done by Magagna (2006) to characterize VIP pit contents, presented in Buckley *et al.*, (2008), revealed significant differences in the physical, chemical and biological characteristics of pit latrine fecal sludge among pits when several pits were compared. This indicates that the properties of VIP pits vary considerably, both within a single pit and among pits.

The total organic load of VIP waste, measured in COD, could be increased by nonfaecal materials that are added to the pit. Anal cleansing material, such as toilet paper, will contribute to the total COD load of VIP pit. The pollution potential of the toilet paper is expressed in Table 2.3. Other sources of wastes include general household wastes and pit latrine additives. Although it is likely that most householders using VIP latrines may not be using toilet paper, other cleansing material (newspaper, leaves) may be used.

 Table 2.3 Pollution Load associated with toilet paper (mg per sheet).

 Source: (Almeida *et al.*, 1999).

COD _t	NH ₃ -N	NO ₃ -N	PO ₄ -P	TS	DS	TSS	VSS
706	0	0.06	0	578	32.5	546	526

2.3 Processes occurring inside the pit of Ventilated Improved Pit latrines

There is limited information on basic processes occurring in the VIP pit, and the factors that affect their rate of occurrence. Studies on the filling rate of VIP pits by Mara (1984) and Still (2002), and studies to provide practical guidance on how to select, design, construct and maintain appropriate excreta disposal systems to reduce feacal transmission risks and protect public health in emergency situations by Harvey *et al.* (2002), were identified during a thorough search of the relevant literature.

These results indicated that biodegradability property of the contents found within a pit influences the rate of filling (Mara, 1984), and that the rate of pit filling can vary, ranging range from 10 to 120l per person per year (Still, 2002). Harvey et al (2002) provided a guideline to design a pit latrine from an estimate latrine volume, and how to estimate the duration between operation and emptying of an existing pit latrine, by considering the number of users and the filling rate of the pit, soil infiltration rates, and the type of anal cleansing material that is used. The study acknowledged that spillage of excreta occurs during pit emptying and recommends that plans to site a sanitation system, must consider heath risks associated with human contacts with excreta, and put in place an organised system for emptying, haulage, disposal and treatment of the faecal sludge when the pit fills up.

Other studies that were found in literature aimed to identify and evaluate the appropriateness, advantages and disadvantages of different sanitation options (Tilley *et al.*, (2008) and how to improve the collection of faecal sludge and to manage feacal sludge after collection (Florian *et al.*, 2001; Klingel *et al.*, 2002; Straus and Motangero,

2002; GHK, 2005; Schaub-Jones, 2005; Scott and Reed, 2006; Strauss *et al.*, 2006; Harvey, 2007).

Therefore general information on design, operation and maintenance of waste treatment plants was used in this study to understand and describe proposed processes occurring inside the pit. However, it is worth mentioning that unlike wastewater treatment plants, the extent of control which can be exercised over VIP pit contents is minimal. This is a major contributor to system failure. Also VIPs are usually solid systems, probably with high ionic strength and therefore theory applied in wastewater treatment plants will apply only in part to VIP systems.

2.3.1 Conceptual theory of processes occurring within the pit

Major processes that occur inside the pit can be categorised into physical and biological (Buckley *et al.*, 2008). The physical processes involve the addition of materials and transport of solubilized materials and moisture. Biological processes comprise degradation of the organic content of the waste by a consortium of micro-organisms to soluble and gaseous compounds. The soluble part seeps away by infiltration to the surrounding soil while the gases leave the pit through the vent pipe. Any remaining biologically inert material remains in the pit.

2.3.2 Physical processes in VIPS

Filling rate

The rate at which the pit fills depends on the rate of accumulation of added material. Degradation of organic material causes the rate of pit filling to be lower than the rate at which material is added. The minimum filling rate depends on the amount of non-degradable material which is added to the pit. According to Still (2002), a 33 % decrease in the rate of filling of a VIP pit is possible if the pit is used for the disposal of human excreta only. This report showed that dumping of household solid waste into the pits increases the rate of sludge accumulation by as much as 10 to 20%. The number of users also affects the rate of pit filling (Still, 2002; Buckley *et al.*, 2008).

Hydraulic transport

The transport of soluble matter and water depends on the hydrogeological and topographical characteristics of the site where the pit is located (Buckley *et al.*, 2008) and on whether the pit is lined or sealed. Important determinants of how materials move through the soil are explained in the following sections.

Determinants of hydraulic transport within the pit

Soil characteristics

Characteristics of the surrounding soil are amongst the most important determinants of how materials move out of the pit. The rate of infiltration can be influenced by the soil permeability (Psarropoulos *et al.*, 2007). A pit surrounded by clay soil should retain more water than a pit surrounded by sandy soil. Sandy soils have large pore spaces between individual particles and the particles do not provide much surface area for adsorption or physical attachment of materials from the pit. In comparison, clay soils are made up of much smaller particles that slow down the movement of water and dissolved particles through the soil (Psarropoulos *et al.*, 2007).

Pit lining

The hydraulic transport also depends on whether the pit is lined or sealed. For a sealed pit, the liquid portion should consist mostly of urine and anal cleansing water only, and so hydraulic movement will be restricted. However in some cases, the pits are preexposed to rainwater and floods because of poor construction. Also, users may sometimes throw wash water into the pit (Buckley *et al.*, 2008).

If the pit is unlined, hydraulic movement is not restricted. Therefore there is flow of water in and out of the pit in a systematic style and leaching is likely to occur (Buckley *et al.*, 2008).

Water table location and subsurface geology

The subsurface geology and the position of the water table can affect ground water movement. In situations where there is a high water table, or supply of water above the pit bottom, water may flow into the pit together with soluble materials from the pit surroundings. Where the water table is lower than the pit contents, coupled with a surrounding soil that is permeable, water carrying disease-causing microbes, organic material and nutrients will seep out of the pit to the surrounding soil (Buckley *et al.*, 2008).

2.3.3 Organic Processes in VIPs

Because oxygen is present at the surface of pit contents, aerobic processes may be expected to dominate in the top layer of pit contents. However, below this surface conditions are expected to be anaerobic due to lack of oxygen diffusion into the pit contents and to the pit bottom (Buckley *et al.*, 2008).

2.3.4 Aerobic Digestion Processes

It is supposed that digestion processes at topmost layer of the pit are predominantly aerobic. This layer is exposed to oxygen flowing from the vent pipe, and from the pedestal. Previous studies have shown that VIP wastes contain insects and maggots, suggesting that there was enough oxygen present for their survival (Buckley *et al.*, 2008). For aerobic biodegradation to occur, the biomass (micro-organisms and waste substrate) absorbs oxygen to supply the energy requirements of the cells. The energy produced is used by the micro-organisms to produce new cells. Concurrently, the cell mass of the micro-organisms is chemically reduced by auto-oxidation (Taljaard *et al.*, 2003). The reaction is demonstrated by the following reaction;

 $organic waste + O_2 + nutrients \xrightarrow{micro-organisms} new \ cells + CO_2 + H_2O + Heat \\ + Non - biodegradable \ residue$

However the extent of aerobic conditions within the pit is not clear.

2.3.5 Anaerobic Digestion Process

Although aerobic processes may be possible at the topmost layer of the pit, degradation occurring below this layer is anaerobic. Anaerobic digestion is the conversion of high molecular weight polymers into low molecular weight compounds by micro-organisms in the absence of oxygen. Generally, gas is produced. This is principally methane and carbon dioxide although hydrogen can also be produced by different groups of micro-organisms. The entire anaerobic digestion process is depicted in Figure 2.1.



Figure 2.1. Conversion processes as presented in the Anaerobic Digestion Model No. 1 (ADM1). Abbreviations: LCFA: Long Chain Fatty acids, HPr:propionic acid, HBu: butyric acid, HVa: valeric acid, MS: monossacharides. Source: Batstone *et al*,(2002).

Biochemistry of anaerobic digestion processes

Different groups of bacteria catalyse the reactions taking place during anaerobic digestion (Anderson and Uyanik, 2003). These microorganisms co-exist in synergetic relations. They are classified as fermentative bacteria, hydrogen–producing acetogenic bacteria, hydrogen-consuming acetogenic bacteria, carbon-dioxide-reducing methanogens and acetoclastic methanogens. Anaerobic degradation of composite organic waste can be divided into four main steps (Seghezzo *et al.*, 1998), namely:

- Hydrolysis
- Acidogenesis
- Acetogenesis
- Methanogenesis

Hydrolysis

During the hydrolytic step, complex organic materials are converted into soluble substrates. The hydrolytic step can also be described as the disintegration or solubilisation step. Disintegration/solubilisation occurs through the action of enzymes secreted by micro-organisms (Batstone *et al.*, 2002). The products of hydrolysis are

amino acids derived from proteins and nucleic acids, sugars derived from carbohydrates, and long chain fatty acids and glycerol derived from lipids of the composite waste.

Acidogenesis

Through acidogenesis (fermentation, the products of hydrolysis, are converted to simple organic acids. Propionate and butyrate are intermediate products formed from acidogenesis and are further converted through acetogenesis to produce acetate. Acetate is the major intermediate in the bioconversion of organic matter to methane (methanogenesis) and carbon-dioxide (Seghezzo *et al.*, 1998). Two groups of micro-organisms are responsible for the fermentation process, namely acidogenic bacteria and the acetogenic bacteria (Anderson and Uyanik, 2003). The presence of hydrogen-utilising bacteria increases the production of acetate.

The stoichiometry for fermentation of the most common products of hydrolysis as presented in Anderson and Uyanik (2003), is as follows:

Glucose Fermentation:

Acidogenic bacteria ferment glucose as follows:

$$C_{6}H_{12}O_{6} + 2H_{2}O \rightarrow 2CH_{3}COOH + 2CO_{2}$$

$$C_{6}H_{12}O_{6} + 2H_{2} \rightarrow 2CH_{3}CH_{2}COOH + 2H_{2}$$

$$C_{6}H_{12}O_{6} + 2H_{2}O \rightarrow 2CH_{3}CH_{2}COOH + 2CO_{2} + 2H_{2}$$

$$C_{6}H_{12}O_{6} \rightarrow 2CH_{3}CHOHCOOH$$

Lactic Acid Fermentation:

$$\begin{array}{rcl} CH_{3}CHOHCOOH + H_{2} & \rightarrow & CH_{3}CH_{2}COOH + H_{2}O \\ \\ CH_{3}CHOHCOOH + H_{2}O & \rightarrow & CH_{3}COOH + 2CO_{2} + 2H_{2} \end{array}$$

The ratio of acetic acid to propionic acids produced from lactic acid fermentation is influenced by the hydrogen partial pressure.

Amino Acid Fermentation

$$C_{5}H_{9}O_{3}N_{(amino\ acid)} + 3H_{2}O \rightarrow 2CH_{3}COOH + CO_{2} + 2H_{2} + NH_{3}$$

$$C_{5}H_{9}O_{3}N + 3H_{2}O \rightarrow CH_{3}CH_{2}COOH + 2CO_{2} + 3H_{2} + NH_{3}$$

$$C_{5}H_{9}O_{3}N + 1H_{2}O \rightarrow CH_{3}CH_{2}CH_{2}COOH + CO_{2} + NH_{3}$$

$$C_{5}H_{9}O_{3}N \rightarrow CH_{3}CHOHCOOH + 2CO_{2} + 4H_{2} + NH_{3}$$

Glycol Fermentation

$$1CH_2OHCHOHCH_2OH + 1H_2O \rightarrow 1CH_3COOH + 1CO_2 + 3H_2$$
$$2CH_2OHCHOHCH_2OH + NH_3 \rightarrow C_5H_7ON + 1CO_2 + H_2O + 4H_2$$

Anaerobic Oxidation of Long Chain Fatty Acids (LCFAs) (Anderson and Uyanik, 2003):

Oxidation of long chain fatty acids in an anaerobic digestion leads to the formation of acetic acid and hydrogen;

$$(-CH_2 - CH_2 -) + 2H_2O \rightarrow CH_3COOH + 2H_2$$

Acetogenesis

Through acetogenesis, the short-chain fatty acids (propionate and butyrate) are reduced to acetate, carbon dioxide and hydrogen (Anderson and Uyanik, 2003).

Conversion of propionate is as follows:

$$CH_{3}CH_{2}COOH + 2H_{2}O \rightarrow CH_{3}COOH + CO_{2} + 3H_{2}$$
$$CH_{3}CH_{2}COOH + 4NH_{3} \rightarrow 4C_{5}H_{7}O_{2}N + 2H_{2}O + 10H_{2}$$

Conversion of butyrate and other fatty acids is as follows:

$$CH_3CH_2COOH + 2H_2O \rightarrow 2CH_3COOH + 2H_2$$

$$5 CH_3 CH_2 CH_2 COOH + 4NH_3 \rightarrow 4 C_5 H_7 O_2 N + 2H_2 O + 10H_2$$

Homoacetogenesis

Carbon dioxide and hydrogen are converted to acetic acid by homoacetogenesis (McCarty and Mosey, 1991 as cited in; Batstone *et al.*, 2002).
$4H_2 + 2CO_2 \rightarrow CH_3COOH + 2H_2O$

Methanogenesis

The fourth stage of anaerobic digestion is methanogenesis. It is a crucial step in anaerobic digestion. Two different groups of bacteria are involved, namely acetoclastic methanogens and hydrogenotrophic (hydrogen-utilizing) methanogens. Both classes are strict anaerobes, forming methane as the end-product of their metabolism (Anderson and Uyanik, 2003). The acetoclastic methanogens produce up to 70% of methane from anaerobic digestion by degrading acetic acid. The other 30% of methane is produced by hydrogen-utilising (hydrogenotrophic) methanogenic bacteria. They produce methane by reducing carbon dioxide, formate and methanol, using the hydrogen produced during the fermentative step (Anderson and Uyanik, 2003).

Acetoclastic Methanogenesis

Acetoclastic methanogenesis involves the production of methane from acetic acid. This is generally represented in the following equation (Anderson and Uyanik, 2003)

 $CH_3COOH_{(sole \ substrate)} \rightarrow CH_4 + CO_2$

Hydrogenotrophic Methanogenesis (Anderson and Uyanik, 2003):

Carbon dioxide is reduced as follows:

$$4H_2 + CO_2 \rightarrow CH_4 + 2H_2O$$

Conversion of acetate to methane is often a rate-limiting step in anaerobic digestion. Hydrolysis is also a rate-limiting step, most especially as regards anaerobic digestion of pit latrine contents.

Overall, anaerobic digestion converts biodegradable organic matter, measured in terms of chemical oxygen demand (COD), to methane. Methane production can therefore be used to assess the anaerobic biodegradability of a material (Seghezzo *et al.*, 1998). The degree of conversion of biodegradable organic material to biomass and gases is referred to as the extent of stabilization (Buckley *et al.*, 2008). For a fully stabilized material the amount of residual biodegradable component is negligible.

Biomass is produced during anaerobic digestion according to the following general reaction;

2.3.6 Factors Affecting the Anaerobic Digestion Process

The micro-organisms that take part in anaerobic digestion have diverse requirements with respect to environmental conditions. Also, undissociated forms of intermediate catabolic products can be inhibitory to microorganisms at high concentrations. Under controlled concentrations, the bacteria may adapt themselves to a variety of substances (Fricke *et al.*, 2007). Thus operating parameters of the system such as pH, temperature, nutrient composition, mixing, toxicity and inhibition must be controlled to enhance microbial activity and increase digestion efficiency (Anderson and Uyanik, 2003). These parameters are addressed in this section.

pH

Anaerobic microorganisms, especially methanogens, are highly sensitive to changes in pH. Maintaining a constant and appropriate pH should be prioritised to ensure effective methanogenic digestion. Hydrogen ion concentration has significant effect on anaerobic microorganisms, buffering capacity, solubility and availability of dissolved ions, within a system. The optimal pH for anaerobic digestion ranges from 6.5 to 7.8. Four different reactions that can lead to change in reactor pH are: (Anderson and Uyanik, 2003)

- The consumption and release of ammonia.
- Production and consumption of volatile fatty acids.
- The release of sulphide due to the reduction of sulphate and sulphite.
- The conversion of neutral carbonaceous organic carbon to methane and carbon-dioxide.

Consumption of volatile fatty acids by methanogens reduces pH, depending on the equilibrium between methanogens and acidogens. The balance between methanogens and acidogens can be affected by changes in operational or environmental conditions (Anderson and Uyanik, 2003).

Temperature

Temperature is another major factor affecting anaerobic biodegradability of substrates in a system. Not only does temperature itself affect the activity of micro-organisms, but the pH of the system is affected by temperature fluctuations. Anaerobic processes exhibit higher sensitivity to temperature with alkalinity variations than aerobic processes. Depending on the temperature of the system, different groups of microorganisms will be active (Speece, 1996). Pit latrines operate at temperatures of 0 to 30° C. Therefore psychrophilic and probably mesophilic conditions are likely to predominate in the pit (Buckley *et al.*, 2008). Most importantly, micro-organisms responsible for the conversion of acetate to methane are affected by temperature fluctuations. Methane-forming micro-organisms are more temperature-dependant than acetate-forming biomass. Thus, at low temperature, the metabolic rate of the acetogens is less affected than that of the methanogens. As a result of this, there is a significant accumulation of volatile acids produced by catabolic action of the acetogens at low temperatures. This may exceed the buffering capacity of the system, causing a drop in pH. This suggests that a temperature decrease could have drastic effect on system capacity for any anaerobic digester type (Speece, 1996) and by inference for VIPs.

The temperature at any particular time can also affect the specific growth rate, decay rate, biomass yield, and reaction rate coefficient (K_s) of micro-organisms in the pit (Speece, 1996). Most micro-organisms exhibit a narrow optimum temperature range, and at this range most reaction rate co-efficients increase with increase in temperature. This can continue until a point is attained at which heat begins to inactivate the cells or thermophilic microbes outcompete them (Lopez Zavala *et al.*, 2004a).

Mixing

Mixing is not feasible in a VIP latrine, but it may be inferred that sub-optimal conditions due to temperature gradients and non-uniform distribution of substrates and nutrients may adversely affect anaerobic digestion in a VIP. Mixing in an anaerobic digester enhances contact between the micro-organisms and substrates, and ensures uniform distribution of nutrients. Dilution with water improves mixing characteristics during anaerobic digestion (Nordberg *et al.*, 2007). Furthermore, mixing destroys temperature gradients in the digester. However, mixing could be disadvantageous, if excessively carried out. It disrupts micro-organisms by mechanically damaging flocs or granules. Specific methanogenic activity of anaerobic biomass can be lost as a result of mixing (Brockman and Seyfried, 1996). Slow mixing is encouraged where it is necessary. Minimal mixing will enhance microbial consortia proximity to substrates.

Ability of micro-organisms to thrive

Anaerobic digestion depends on on co-operative action of diverse microbial consortia. For digestion to occur efficiently, the environment of the reactor must be suitable for these micro-organisms to thrive. Among the factors that determine the ability of a particular group of micro-organisms to flourish in an environment are the availability of suitable electron acceptors, substrates and temperature. Growth rate of micro-organisms and competition from other organisms for common substrates are also important (Henze *et al.*, 2002). Maintaining higher concentrations of micro-organisms will enhance anaerobic digestion efficiency, thereby reducing the time required for degradation to occur.

For a pit latrine, if the population of viable micro-organisms present in the waste heap is high and environmental conditions are suitable, a high rate of stabilization of feed materials will be achieved. Micro-organisms are usually introduced into the pit with faeces and other organic materials (*e.g.* anal cleansing material and leaves) that are thrown inside the pit. Studies indicate that the population of micro-organisms present in raw faeces is large enough for the faecal matter to undergo natural breakdown (Buckley *et al.*, 2008).

Nutrient availability

Nutrient imbalance and unavailability to anaerobes can inhibit biogas production or methanogenesis (Schanbacher *et al.*, 2005). This is because nutrients are essential requirements for cell growth, and for efficient synthesis of enzymes necessary for metabolism. Nutrients are classified either as macro- or micronutrients, based on the concentration at which they are beneficial. As with every biological process, nitrogen, phosphorus, sulphur and iron are necessary for growth. Other micronutrients are required in very small amounts (Henze *et al.*, 2002). Nitrogen in the form of ammonium is essential for the formation of new biomass during anaerobic digestion. The nutrient requirement for anaerobic micro-organisms is lower than that of their aerobic counterparts, because the mass of biomass formed is low (Fricke *et al.*, 2007). Fresh faecal material contains an adequate supply of macro– and micronutrients for anaerobic micro-organisms to carry out their activities.

Toxicity and inhibition

Substances that are potentially toxic or inhibitory at sufficiently high concentrations may include by-products of anaerobic metabolism which can slow down the rate of digestion or cause process failure (Anderson and Uyanik, 2003). Examples are ammonia, oxygen, volatile fatty acids, sulphide and alkali and alkali earth metals.

Oxygen

Strict anaerobes such as the methanogens are exceptionally sensitive to the presence of oxygen. However, methane production is still possible in the presence of oxygen, if facultative micro-organisms are present during the initial fermentative step. They will utilize the available oxygen. Only thereafter can the methanogens function (Seghezzo *et al.*, 1998).

Sulphides

Reduced forms of sulphur (sulphide ion and hydrogen sulphide) are strong inhibitors of anaerobic digestion. Sulphur inhibition can arise from competitive consumption of methanogenic substrates, acetate and hydrogen by sulphur-reducing bacteria which reduce sulphate to sulphide, thereby lowering methane production.

Ammonia

Anaerobic digestion of organic waste materials, which are rich in proteins, may release ammonia from the mineralization of organic nitrogen compounds. Depending on pH, either ammonia or ammonium may be produced. The release of ammonium during the anaerobic hydrolysis of organic nitrogen compounds is associated with an increase in pH. Ammonium is an important factor for the buffering capacity of an anaerobic reactor such as a VIP latrine. Ammonification can neutralize the reduction of the pH value associated with acidification step of anaerobic digestion (Fricke *et al.*, 2007). At increasing pH value (8.5 or more) ammonia is produced whereas at lower pH (less than 8.5) more ammonium is produced (Figure 2.2) (Fricke *et al.*, 2007). An optimal pH of between 6.4 and 7.2 is recommended for an anaerobic digestion process, and the ammonia/ammonium balance can help to maintain this.



Figure 2.2. Dissociation balance between ammonia/ammonium depending on pH and temperature. Source: Fricke *et al.* (2007)

Volatile Fatty Acids (VFAs)

Accumulation of VFAs occurs when the rate of hydrolysis is faster than the rate of onward conversion of the acids. As a result of this, the pH of the reactor decreases (acidic), to the detriment of methanogenesis. Methanogenes cannot function well at low pH.

2.4 COD fractionation occurring within the pit

Amongst the hypotheses tested in this study was that percolation (*i.e.* concentration) of soluble organics will describe the direction of water movement inside the pit (*i.e.* that an increase in soluble organics will occur in the same direction as increase in water). This is based on the assumption that mobile water molecules within the pit carry soluble (dissolved) organics as they pass through the different layers of the pit. Therefore it was hypothesised that the soluble COD concentration gradient through the pit layers can be related to the direction of water movement within the pit. The theoretical background underlying this hypothesis is given here.

Classification of soluble and particulate COD entails COD fractionation. In order to develop methods for COD fractionation, background knowledge of existing models defining biological treatment is required. This section presents available literature on the model of breakdown of influent organics in other biological systems.

Firstly, a hypothetical distribution of various fractions of COD that can possibly occur in VIP sludge is described by applying literature knowledge of different wastewater COD fractions. Then, because the concentration of the soluble COD within the different layers of the pit is of major interest to this study, literature on COD fractionation in mixed liquor is reviewed. It is assumed here that pit latrine faecal sludge can be related to a mixed liquor which comprises of waste contents and consortia of bacteria.

2.4.1 Theoretical description of COD distribution (fractionation) within the pit

COD fractionation entails classification of organics based on their rate of biodegradability (Orhon and Çokgör, 1997; Arslan and Ayberk, 2003).

Faeces, urine, anal cleansing material and anal cleansing water consist of water, organic and inorganic content. The breakdown of this organic and inorganic waste content takes place inside the pit. While a fraction of this will be biodegradable, a non-biodegradable portion will remain unaltered. Description of each portion at a particular time depends on the amount that has been added minus the amount which has been biologically degraded already, and the amount of moisture (water) present inside the pit. The remaining inorganic (ash) and non-biodegradable organic material, as a fraction of the total mass, will therefore increase with time (Buckley *et al.*, 2008).

Micro-organisms break down the organic matter in the composite waste into biodegradable and non-biodegradable fractions. Hypothetically, the readily biodegradable (RBCOD) materials, which are normally soluble, either seep away into the surrounding soil together with available water in the pit (including water from urine), or become utilized by micro-organisms for energy production and cell multiplication.

The slowly biodegradable counterpart (SBCOD) accumulates at the bottom of the pit and undergoes slow digestion. These materials remain at the bottom of the pit as long as it takes to digest them. For a full pit, particulate COD should reduce with time. The first step of the digestion of SBCOD is hydrolysis. Hydrolytic reactions are usually slow and could be reaction rate-limiting. SBCOD is hydrolyzed to readily biodegradable materials (RBCOD). The hydrolysis products are converted by fermentation to organic acids (long chain fatty acids, sugars and amino acids). The organic acids are assumed to be converted by acetogenesis to acetate and finally by the methanogens to produce methane gas.

The unbiodegradable portion is mostly from other waste components that are disposed of into the pit. A fraction of this is soluble (nbSCOD), while the other fraction is particulate. Neither can be metabolized by micro-organisms. They remain entangled in the accumulated sludge mass.

The inorganic components (nutrients) found in VIP waste are either soluble or particulate (Wentzel *et al.*, 1999). Microbes utilize a part of the soluble inorganics and transform them into gases and solids. The non-utilizable and soluble inorganics leave with the residual soluble organics through leaching.

It is hypothesised that the direction of movement of the solubles within the layers of the pit depends on the direction of infiltration. It is also hypothesised that the region of greater concentration of these solubles within the pit layers reflects their direction of movement in solution in the infiltrating water.

The COD fractionation of waste inside the pit is explained here according to the conventions for modelling activated sludge systems, as illustrated in Figure 2.3.



Figure 2.3 Complete subdivision of the influent organic material (measured as COD) showing 5 fractions required for dynamic modelling of the fully aerobic and anaerobic activated sludge systems. Source: Wentzel *et al*, (1999).

2.4.2 COD fractionation in a mixed liquor

Various kinetic models have been used to describe and /or quantify fractions of organic material (measured in terms of COD) in fully aerobic and anaerobic systems (Ekama and Marais, 1984; Wentzel *et al.*, 1990, 1992, 1999; Ekama *et al.*, 2007). These procedures are either physically or biologically based (bioassay tests), or a combination of both. Total COD (tCOD) of an organic substrate can be subdivided into two physical fractions: total soluble COD (SCOD) and total particulate COD (PCOD) (Rössle and Pretorius, 2001, as shown in Figure 2.4). Particulate COD is divided into slowly biodegradable COD (SBPCOD) and unbiodegradable particulate COD (UPCOD), while soluble COD is divided into readily biodegradable COD (RBSCOD) and unbiodegradable COD (UPCOD) and unbiodegradable COD (RBSCOD) and unbiodegradable COD (USCOD) (Dold and Marais, 1986).



Figure 2.4 COD fractions in mixed liquor a-.soluble COD components; b - particulate COD components. Source: Orhon and Çokgör (1997)

COD classification, as shown in Figure 2.4, is based on the theory that the observed differences in biokinetic response of microbial cells to RBCOD and SBCOD is a result of differences in molecule size. RBCOD is made up of smaller molecules which can be easily assimilated into microbial cells, whereas SBCOD consists of larger molecules which demand extracellular hydrolysis into smaller molecules before they can be assimilated into microbial cell (Dold and Marais, 1986; Wentzel *et al.*, 1999). Physical separation techniques have been used to estimate the two COD fractions since these are based on the molecular sizes of the two fractions. Filtration with different filter pore sizes is documented (Dold and Marais, 1986; Lesouef *et al.*, 1992; Mamais *et al.*, 1993; Torrijos *et al.*, 1994, as cited in Wentzel *et al.*, 1999). Depending on the pore size, part of the particulate material is likely to pass through the filter paper. Therefore accurate separation of the RBCOD is compromised. Furthermore, estimation of the COD of the residue (particulate COD) can be difficult.

Alternatively, physical centrifugation can be used, since it allows both the supernatants and pellets to be assayed (Naidoo, 1999; Melcer *et al.*, 2003). The limitation of this method is that the readily and slowly biodegradable COD fractions cannot be directly estimated. Bioassay tests may be used to monitor the reaction of microbial cells (biomass) to waste COD (Ekama *et al.*, 1986; Wentzel *et al.*, 1999) if the need arises.

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From the bioassay test, the unbiodegradable COD can be quantified. This is done by carrying out biodegradability assay on the supernatants. Subtracting the residual COD at the end of the assay from the initial COD of the supernatant gives the RBCOD.

The princples of COD fractionation in mixed liquor, as presented here, are applied in the next chapter to design a method for assessing soluble and particulate COD fractions of fresh faeces and of VIP sludge from different layers within a VIP pit latrine.

3 Materials and methods

A full list of different materials used for the laboratory analysis and site visits is presented in Appendix A. The preparation and compositions of all reagents and solutions used is reported in Appendix B.

3.1 Description of the study area

VIPs sampled in this study were located within Tongaat (La Mercy, Shayamoya) and Magwaveni), and the neighboring Umhlanga areas of eThekwini Municipality . These areas fall into the northern area of the eThekwini Municipality. EThekwini is a metropolitan municipality, and is one of the 11 districts of KwaZulu-Natal province of South Africa. The municipality is located on the east coast, and is inhabited serves approximately 3,090,000 people. The climate is subtropical with mild to cool winters, and warm summers with elevated humidity, but without frost. The area has an annual rainfall of 1,009 millimeters. The average annual temperature is 21°C, with daytime maxima from January to March peaking at approximately 28 °C, and a minimum of approximately 21°C. Daytime highs from June to August are approximately 23°C and the minimum approximately 11°C.

The Northern Area of eThekwini municipality is classified into in eight economic zones, namely Durban North, Inanda / KwaMashu, Mount Edgecombe, Phoenix, Tongaat, Umdloti, Umhlanga / La Lucia, and Verulam. La Mercy and Shayamoya are sub-areas in Tongaat and Magwaveni is a small informal settlement in Tongaat. The population of the Northern Areas represents 31.3% of the total eThekwini population (approximately 966 600). The total number of households in the Northern areas is 201 890 with 4.7 people per household. This represents 31.0% of the number of households in the municipality (eThekwini Northern Area Economic Analysis, 2005). The largest proportion of the population in the Northern Area live in the Inanda / KwaMashu economic zone (56.8%), followed by Phoenix (17.5%). Tongaat (8%) and Umhlanga are amongst the least populated (3%). The area's percentage input to the eThekwini GDP is comparatively small (15% to 17%).

Only the peri-urban communities in these areas were studied, and they have access to acceptable level of water supply. Water supply system consist mainly of standpipes,

which are unmetered but require water to be carried to individual households. The urban part of the northern areas of eThekwini has access to metered water supply.

In general, sanitation provision is a major challenge to eThekwini Municipality. After a local government election in 2001, the boundary of the eThekwini was extended. This led to the incorporation of more rural areas into the Municipality. These areas fell outside the reach of conventional waterborne sanitation and were often either unservices or depended mainly on traditional unimproved pits latrines. Since provision of sustainable sanitation is a priority of the Municipality, it aims to introduce sustainable on-site sanitation to all such areas within its control. The areas included in this study were all served by VIPs. They were chosen because they are located within the eThekwini municipal area and the pits were in the process of being emptied at the time of the study. The areas were similar in many respects. The main difference among them appeared to be proximity to the coast. Those closest to the coast were designated coastal; the remainder were designated inland. This type of classification was important because the nature of soil differed between the two locations. Soil characteristics are known to affect moisture content of pit contents. La Mercy, Tongaat (Magwaveni) and Umhlanga fell into the coastal category. Soil in these areas was sandy, *i.e.* soil was well drained. Tongaat (Shayamoya) was located farther from the coast. In this area, the soil had higher clay content than Magwaveni and therefore had the ability to retain more water.

3.2 Site visits

During site visits, informal questionnaire interviews were carried out alongside sampling. Personal observations of the state of the toilets and of the site were made.

3.2.1 Questionnaire-based survey

Household selection

The questionnaire survey was conducted in March 2007. Only households in Tongaat (Magwaveni) were included in the survey. It was assumed that user behaviours in Magwaveni were representative of those in all areas from which pit contents were samples, and indeed of VIP users throughout eThekwini Municipality.

There were approximately 600 households in Magwaveni, served by communal taps and communal VIP toilets, *i.e.* one VIP toilet served more than one household.

(Krakauer, 2004). An aerial photograph of Magwaveni informal settlement is given in appendix F.

Survey structure

A total of 10 informal questionnaire interviews were conducted. The ten respondents were from ten different households. There was no particular target group, as the questions were simple and could be answered by any member of the household between the ages of 18 and 70. Questionnaire-based interviews were carried out with an isiZulu-speaking facilitator. All questions were asked in isiZulu. Questions used are presented in Appendix C.

Interview questions

Questions used for the interview focused on user behaviour practices, user management routines and construction of VIPs. The user behaviour section addressed the ways the households use the toilets. For questions on user behaviour practices the most important information obtained focused on: (i) whether or not users throw water into the pit, and (ii) the type of water they throw into the pit.

Management-related questions were used to determine the measures carried out by users to ensure proper functioning of the pit and to reduce smell. For management measures, information focused on: (i) measures used to reduce malodour (*i.e.* chemical or additive; type and quantity of households that use each type) and (ii) measures used to enhance pit life.

Construction-related questions were used to describe the relationship between the malfunctioning of the VIP toilet and errors in construction. Failure of VIP in this context implies presence of flies as a result of broken or absence of the fly screen, a broken vent pipe or a broken door.

Personal observations

Colour and smell of pit latrine contents were observed, noted and related to the status of the pit. Observations were also made on the different materials that constituted pit latrine contents.

Data analysis and reporting

Information was collected and incorporated into an Excel spreadsheet for analysis. Data gathered from questionnaires were grouped into two sections, namely user behaviour practices and management measures. Results were represented in bar charts.

3.2.2 Sampling

Samples of pit latrine sludge were collected between March, and November, 2007, that is in between early autumn and the end of winter seasons. Procedures used for sampling fresh faeces and for sampling pit latrine sludge are presented in this section.

Fresh faeces

Fresh faecal samples were collected in plastic containers from members of the experimenter's household, immediately after defecation,, and were transported to the laboratory. Samples were stored in the cold room until use.

Pit latrine sludge

Sampling of pit latrine sludge was carried out alongside the site visits in conjunction with pit latrine emptying campaigns by the Municipality in the different locations selected for inclusion in the study. Samples of volume approximately 300 mL were collected by shoveling samples from each layer into individual plastic buckets (Figure 3.1). To obtain samples from layers beneath the surface layer, the collector had to wait until the particular depth was reached by the pit emptier. This was repeated for each of the sampled layers. The buckets were wrapped in a black refuse bag before transporting them to the laboratory, to imitate the dark environment of the pit. This was done to ensure that the bioactivity of the samples was not altered. Samples were stored in the cold room at 4°C until use in the laboratory.



Figure 3.1 Photograph showing sampling technique for VIP faecal sludge. Student waiting, to collect contents from different depths within the pit, as emptied by the pit emptier, shown in orange overalls.

3.3 Analytical Methods

Two sets of experiments were undertaken. A pilot study was carried out in thich samples were collected from a single pit to ascertain if there was a significant variability in the physico-chemical characteristics of pit latrine contents located at different depths of the pit. These samples were obtained from the top, middle and bottom layer of the accumulated heap of a VIP toilet vault from one pit during an emptying exercise in Tongaat area. Each layer was separated by approximately 300 mm from the next. Analyses carried out on the Tongaat VIP sludge were:

- Physico-chemical characterisation of samples from the three different pit depths (Table 3.1).
- Serum bottle test for the determination of anaerobic biodegradability of samples.

The results of this set of experiments were presented as preliminary and indicated possible variation of characteristics of the pit latrine contents with depth.

A disadvantage associated with this initial analysis was that a quantitative measure of the depth at which samples were collected could not be scertained. Thus general conclusions, such as the extent of activity within the pit, could not be made. Also, the pilot study showed that sample size needed to be increased to ensure that results gathered were representative of the characteristics of pit latrine contents. Therefore, in subsequent full-scale sampling, samples were collected at four depths, namely surface layer, and depths of 0.5 m, 1 m and 1.5 m. Samples were collected from 16 pits in the full-scale experiment.

Analyses carried out on the VIP sludge from the 16 pits were:

- Physico-chemical characterisation of samples from the four different pit depths for all 16 pits (Table 3.1).
- Amongst the 16 pits, samples from Pit 9 were tested for biodegradability. This is because the sample appeared fresh and was likely to have more residual biodegradable COD than the other pits. This was supported by the highest organic solids content (82%/g wet sample) of its surface layer samples. Therefore it was expected that the samples would produce a measurable amount of methane gas to calculate biodegradability (low gas production during the anaerobic biodegradability test performed in the pilot study suggested that fresh sample with high residual biodegradable COD content was necessary to yield reliable results in this test). Conversely, samples from the remaining pits appeared already stabilised (dark in colour), and likely to contain less residual biodegradable COD.

In addition, COD fractionation was carried out on VIP sludge samples from all 16 pits and on fresh faeces. A technique for fractionating COD by centrifugation was developed. Centrifugation was used to separate total COD content of fresh faeces and VIP sludge, respectively, into soluble COD and particulate COD. Centrifugation is typically used to separate particulate materials suspended in a liquid medium. The success of the centrifugation method is strongly linked to the speed and the length of time of centrifugation. A longer spinning period ensures more effective the separation. This section describes the techniques used to determine the physico-chemical properties and biodegradability of fresh faeces and of pit latrine faecal sludge samples. The physico-chemical properties measured and the reasons for including each parameter are listed in Table 3.1.

Parameter	Reason for inclusion				
Total solids	As a step to determine moisture content, and				
	organic solids.				
	To eliminate variation in COD value, as a				
	result of dilution effect of different moisture				
	contents when quantifying COD of samples.				
Moisture	To quantify the moisture content of the				
	different samples and use it to relate to sample				
	biodegradability				
Organic solids (Volatile solids)	To quantify the organic material present in the				
	different samples				
Total Chemical Oxygen demand (tCOD)	To measure oxidisable organic matter in the				
	different samples				
COD fractionation by centrifugation	To isolate the soluble and particulate				
	components of COD in order to quantify				
	respectively, the dissolvable and particulate				
	oxidisable matter present in the different				
	samples.				

 Table 3.1 Parameters used to measure physico-chemical properties of fresh

 faeces and pit latrine faecal sludge

3.3.1 Sample preparation

Approximately 50 g of each sample was suspended in distilled water and made up to 1 L in a volumetric flask (Stock 1). To ensure homogeneity, the whole suspension, designated as Stock 1, was transferred into an electric blender and macerated for 1 minute. From Stock 1, 60 mL was withdrawn, transferred into another volumetric flask and made up to 500 mL using distilled water. This suspension was designated Stock 2. Serial dilution of Stock 2 was carried out to attain a COD concentration within the detection limits of the test (*i.e.* between 50 and 900 mg COD/L). Stock solution 2 was stored in the cold room at 4° C and used within 24 h of preparation.

For the COD fractionation test, 50 mL of Stock 2 for each sample was withdrawn and transferred into 50 mL centrifuge tubes (in triplicate). The triplicates were centrifuged for 1h at 4 000 rpm. The centrifuged solution was designated Stock 3.

3.3.2 Physico-chemical analysis.

Solids and moisture content determination

Solids were determined according to Standard Methods (A.P.H.A, 1998). Total solids (TS), volatile solids (VS), and moisture content of the samples were determined.

Total, soluble and particulate Chemical Oxygen Demand (COD)

The tCOD was measured using the open reflux method according to Standard Methods (APHA, 1998). Aliquots of 10 mL Stock 2 (for each sample) were withdrawn and placed into volumetric flasks in triplicate. For soluble COD, the supernatants from Stock 3 were decanted, and the COD (soluble COD) analysed according to Standard Methods (APHA, 1998). For particulate COD, the pellet in the tubes centrifuged as described in 3.3.1 was re-suspended in distilled water of volume equivalent with that of the corresponding supernatant. The resultant solution was analysed for particulate COD according to Standard Methods (APHA, 1998). (Figure 3.2). For inclusion of dilution factors in the calculation of COD, the following formula was used.

$$COD mg / g \ sample = \frac{COD mg / L}{Overall \ dilution \times 1000}$$

Where:

Overall dilution = $dilution_{stock 1} \times dilution_{stock 2}$

Estimation of particulate and soluble COD

The measured concentration of stock 2 was used to determine the total COD (tCOD) content of the sample (in mg COD/g sample) and the relative contribution of soluble (SCOD) and particulate (PCOD) fractions to the total COD. The volume of stock 2 is designated as $V_1(L)$.



Figure 3.2 Diagrammatic explanation of the separation between PCOD and SCOD

In a sample suspension of appropriately diluted VIP sludge of volume $V_1(L)$ there was:

XmgCOD(S)/L = COD of supernatant representing soluble COD

YmgCOD(P)/L = COD of pellet representing particulate COD

Then total COD $(S) = X \times V_1 [mgCOD (S)]$

and total COD $(P) = Y \times V_1 [mgCOD (P)]$

After centrifugation and decanting the supernatant:

All COD(P) was in the pellet and all COD(S) was in the supernatant

Then COD (S) in supernatant = $\frac{X \times V_1}{V_2} (mgCOD(S)/L)$

Where V_2 signify the volume of supernatant in L

The pellet was re-suspended in distilled water to make V_3 such that:

$$V_{3}(L) = V_{1}(L)$$

In that case *COD* (*P*) in re-suspended pellet = $\frac{Y \times V_1}{V_3}$ (*mgCOD* (*P*)/*L*)

COD in supernatant (X') was measured:

$$X' = \frac{X \times V_1}{V_2}$$

Therefore $X = \frac{X^{T} \times V_{2}}{V_{1}} (mgCOD(S)/L)$ of wastewater sample

Similarly, COD of the resuspended pellet (Y^{I}) was measured:

$$Y^{I} =: \frac{Y \times V_{1}}{V_{3}}$$

Therefore $Y = \frac{Y^{T} \times V_{3}}{V_{I}} (mgCOD(P)/L)$

Validation of COD fractions by mass balance

The sum of the COD of the pellet and the COD of the supernatant should equal the total COD:

Total COD of the sample suspension = total COD(S) + total COD(P) =

$$= X \times V_1 (mgCOD) + Y \times V_1 (mgCOD)$$
$$= \frac{X^{T}V_2}{V_1} \times V_1(mgCOD) + \frac{Y^{T}V_3}{V_1} \times V_1(mgCOD)$$

Substituting for X and Y in equation, where $V_1 = V_3$

$$Total \ COD = X^{I}V_{2}(mgCOD) + Y^{I}V_{3}(mgCOD)$$
$$= (X^{I} \times V_{2} + Y^{I} \times V_{1})mgCOD$$

which shows that sum of COD of supernatant and Pellet equals the total COD of the stock solution V_1 and therefore the procedure is correct.

Analysing and reporting data from physico-chemical analyses

Samples with higher moisture content tend to have decreased COD per gram sample value, due to the dilution effect by its moisture. Therefore results were presented on a per-dry-solids basis to eliminate variability related to different moisture content. Descriptive statistics data obtained for fresh faeces were tabulated for comparison with literature values.

Analysis of data from Tongaat VIP samples

Data for the Tongaat VIP faecal sludge were compared graphically by depth for all measured parameters. This was to assess whether there was a trend between measured physico-chemical characteristics of pit latrine faecal sludge samples and depth of sample in the pit. Error bars were used to indicate the upper and lower boundaries of the 95% confidence interval. This quantifies the confidence that the sample mean is within a certain range of the population mean, due to sampling error. Because the sample size was small (n=3), the standard error was multiplied by t value to calculate the confidence limits.

Analysis of data from the 16 different pits

Results of physico-chemical analyses of sludge from 16 pits were statistically tested for normality of distribution using the Kolmogorov Smirnov (KS) test. Data for fresh faeces were compared graphically to those of VIP pit latrine faecal sludge. Univariate analysis of variance (SPSS 15) was used to ascertain whether the differences between characteristics of fresh faeces and pit latrine faecal sludge of the 16 different pits were significant.

Data for VIP faecal sludge at different depths were compared graphically by plotting means at each depth and comparing these both visually and statistically. Data for individual pits were presented in Appendix E and reference to these was made as necessary. Correlation analysis (SPSS 15) was used to determine if there was a relationship between measured physico-chemical characteristics of pit latrine faecal sludge samples and depth in the pit. The correlation co-efficient, R was used to indicate the correlation (relationship) between a given parameter and depth in the pit.

Data for SCOD for the different layers amongst the pits were correlated with their respective moisture content. Thereafter, data for SCOD were also correlated with the corresponding tCOD content data. The R values obtained from the correlation analysis were compared. Higher R values indicated that either moisture content or tCOD content explained the variation in distribution of SCOD within the pit.

Univariate analysis of variance using the post-hoc Scheffe test was applied to determine if there were significant differences among physico-chemical characteristics of samples collected from different layers within the pit.

Finally, data were compared among the different pits examined, using multivariate analysis of variance with the post-hoc Scheffe test.

3.3.3 Biodegradability tests

A serum bottle test was used to investigate anaerobic biodegradability of fresh faeces and pit latrine sludge from the different layers (depth) of a VIP pit,. This test was intended to assess the extent of biological conversion that occurred at the different layers of the pit. The technique has previously been Owen *et al.* (1979), Remigi and Buckley (2005) and Angelidaki (2008). The test is based on the production of biogas pressure and methane formation (Remigi and Buckley, 2005; Angelidaki *et al.*, 2008). For this study, the serum bottle test technique was chosen over other techniques used for monitoring biodegradability of substrates under anaerobic condition because:

- The serum bottle test does not require sophisticated equipment other than a gas chromatograph.
- The technique requires comparatively less effort than other methods documented in literature for measuring anaerobic biodegradability of samples.
- It allows a large number of replicates to be tested under different experimental conditions, thereby providing comprehensive, reproducible and accurate results (Remigi and Buckley, 2005).

The specific objective for applying the serum bottle technique in this study was:

- to test whether the technique could be used to measure the anaerobic biodegradability of both fresh faeces and pit latrine faecal sludge,
- to test the biodegradability of the substrates.

The criteria used for accepting the serum bottle test were:

- Samples were able to produce sufficient volume of methane to calculate their biodegradability, and
- Results for gas production measurements did not have high variability (*i.e.* large standard deviation value).

Although the serum bottle test is described in the literature, it is not a widely used test at this stage and is therefore presented here in full.

3.3.4 Test principle

- A measured volume of anaerobic sludge was incorporated into a gas tight vial. The sample was was supplemented with the organic substrate to be degraded and with a solution containing nutrients and minerals, sealed under anaerobic condition, and incubated at a controlled temperature (See appendices for nutrient media composition).
- The nutrient solution supported biomass metabolism
- Utilisable (biodegradable) substrates were degraded by anaerobic microorganisms.
- Biogas that was produced as a result of microbial activity accumulated in the headspace and caused a pressure build-up inside the vial.
- Gas production was measured by releasing headspace gas
- Substrate biodegradability was measured by monitoring the cumulative or total methane produced from a sample incubated in the defined medium under anaerobic conditions.

3.3.5 Methodology

Anaerobic sludge from Northern Works Waste Treatment Plant (NWWTP) was used. This came from an anaerobic digester that was fed with primary sludge containing faecal particulates. Consequently, it was expected that micro-organisms present in the anaerobic sludge would utilize any degradable material in faeces and VIP sludge.

Test Preparation

The total COD and volatile solids content of the anaerobic sludge used in the test was quantified before setting up the serum bottle experiment. The anaerobic sludge was preincubated until gas production became negligible. This was done to ensure that residual biodegradable organic materials in the sludge were completely used up by its biomass, so that gas produced from the bottles after substrate addition would be solely as a result of conversion of the substrates (samples). The pre-incubation was performed in a 125 mL bottle. Only nutrient medium was added to the bottles and no substrate was added. The pre-incubation conditions were similar to the test conditions.

Preparation of samples and standards for the serum bottle test as used in this study are represented in Table 3.2

Designation	VIP sludge from pit depth				Acetate	Fresh faeces	Anaerobic sludge	Nutrient solution	Number
					of				
	surface	0.5m	faeces	1.5m					replicates
Test units	Yes	No	No	No	No	No	Yes	Yes	3
	No	Yes	No	No	No	No	Yes	Yes	3
	No	No	Yes	No	No	No	Yes	Yes	3
	No	No	No	Yes	No	No	Yes	Yes	3
	No	No	No	No	No	Yes	Yes	Yes	3
Standards	No	No	No	No	Yes	No	Yes	Yes	3
Negative control	No	No	No	No	No	No	Yes	Yes	3
Total number of test units									21

Table 3.2 Preparation of samples and standards for serum bottle test

A volume of acetate equating approximately 1 g COD was added to a set of the bottles containing the anaerobic sludge and nutrient solution as standards. This was to done to ensure that the micro-organisms present in the sludge were active.

The test units were prepared in the same way as the standards. However, fresh faeces and VIP sludge samples were used as the substrates in place of acetate. In the first set of experiments (using Tongaat VIP sludge samples), sample volumes equating approximately 0.89 g COD of sample were used per bottle. (See Appendix D for calculation of the amount of sample in COD units inside the bottles). This concentration was based on results from a parallel study (Magagna, 2006) which indicated it to be sufficient for gas production. Also, with this concentration substrate inhibition was not expected, given that VIPs contain less residual biodegradable COD (Mara, 1984; Franceys *et al.*, 1992; Still, 2002). This suggests that the contents reduce in volume with time as a result of microbial decomposition within the pit (Puddifort, 1995 in Pitnet,

1996). For fresh faeces, this concentration was also expected to be adequate, since most of the biodegradable COD in fresh faeces is slowly biodegradable (Lopez Zavala *et al.*, 2002; 2004a; 2004b). Equal amounts of COD per sample were used in all bottles, in order to facilitate comparison between biodegradability measurement of fresh faeces and VIP sludge samples.

However, a second set of test unit were prepared using the same anaerobic sludge. In the second set of experiments using VIP pit 9, a lesser amount of 0.3g COD per bottle was used for all samples.

Also, it is important to note that VIP sludge used in the two separate experiments at the two different COD values (first 0.89 g, then 0.30 g), were sampled from two different pits. Therefore, the two experiments cannot be compared to one another because the VIP sludge used in the two separate set of experiment might have had different properties with respect to biodegradability.

In the second set of experiments, the same anaerobic sludge was used, but pit latrine faecal sludge originated from Pit 9 of the 16 VIP pits sampled. Based on results of the first experiments, a lesser amount of 0.3g COD of pit latrine sludge per bottle was used for all samples.

Negative controls (blanks) containing anaerobic sludge and nutrients only were prepared alongside the standards and test units. It was expected that the negative controls would produce less gas than the test units and standards.

Bottles were prepared in triplicates. Each of the bottles was flushed with a mixture of nitrogen and carbon dioxide gas (50% N_2 and 50% CO_2) to eliminate oxygen. The bottles were then sealed with rubber septa and aluminium crimps (Figure 3.3), and incubated in a temperature-controlled room (~38°C), in the dark. To equilibrate pressure, the septum of each vial was punctured two hours after sealing using a lubricated glass syringe. The volume of gas released was wasted.



Figure 3.3 Diagrammatic representation of a serum bottle containing a measured mass of sample, a measured volume of anaerobic sludge and nutrient solution, and leaving a headspace volume.

Gas volume and composition analysis

Headspace gas volumes were measured regularly using a lubricated gas syringe until the gas volume curve formed a plateau. Gas composition readings were taken alongside gas volume measurements, to measure methane content. Gas was collected using a gas-lock syringe and analysed using a GowMac gas chromatograph. The volume of gas produced reduced with the amounts of methane produced. Gas composition on days on which no gas was produced was typically approximately the same as that of the previous day. If no gas was produced from the bottles, but the methane composition measured was different from what was measured on the previous day, then it was inferred that the analysis was inaccurate and hence was repeated, or that CH_4 was produced as a result of CO_2 absorption.

Calculations

The methane content of the volume of gas produced from the serum bottles was assayed and used to calculate the biodegradability of the samples. The calculations were performed according to the method described by Owen *et al.* (1979) and Remigi and Buckley (2005). Biodegradability was calculated from produced gas volume and composition as follows:

Gas volume correction for residual pressure

The total volume of gas that was produced from a serum bottle with multiple insertions was defined by applying the method of Owen *et al.* (1979):

$$V_{T} = V_{s} \cdot \left(\alpha + \frac{\alpha V_{s} + V_{H}}{V_{H}} \cdot \sum_{J=1}^{N-1} \left[\left(\frac{V_{s} + V_{H}}{V_{H}} \right)^{N-(J+1)} \right] \right)$$

where:

 $V_{_{H}}$ = volume of headspace in the serum bottle

 V_s = maximum volume measured in the syringe

N =total number of insertions into the bottle

 α = fraction of the syringe filled during the last measurement and is calculated by:

 $=V_{last measurementt} / V_s$

Normalisation to Standard Temperature and Pressure (STP)

In order to make comparisons with literature data, gas volumes were normalised to standard temperature and pressure (STP), *i.e.* converted to 1 atm and 0°C (273.15K): After calculating V_{τ} it was normalised to standard temperature and pressure according to the formula by Remigi and Buckley (2005):

$$V_{_{T,N}} = V_{_{T}} \cdot \frac{101325kPa}{P_{_{atm}}} \cdot \frac{273.15K}{T_{_{meas}}}$$

Where:

1 atmosphere is assumed for P_{atm}

 T_{max} = the temperature of the bottles

Mass balance calculation to determine methane production

The COD degraded was determined as follows;

 $1gCOD = 0.350LCH_4$ at STP

Correction for endogenous respiration / residual COD conversion

To calculate the nett volume of methane produced solely from biodegradation of samples, the volume of methane produced during the incubation period by the control bottle was subtracted from the volume of methane produced by the sample:

$$V_{net} = V_{test} - V_{controls}$$

Calculation of biodegradability as methane produced per amount of COD added

COD degraded was used to express the biodegradability of the samples. This was done by quantifying the portion of the organic matter in the test material that underwent microbial degradation *i.e.*, that was converted to methane. Thus, by comparing the initial COD added to the serum bottle (COD₀) to the total amount of methane produced in the course of the test ($V_{CH,\infty}$), biodegradability was determined according to Remigi and Buckley (2005) by:

$$B \approx \frac{V_{CH,\infty}}{COD_0}$$

Analysing and reporting serum bottle test data

Data of daily gas volume measurements were checked for normal distribution using Kolmogorov-Smirnov test. Data were normally distributed. Univariate analysis of variance (SPSS 15) was used to ascertain whether there was significant variation in daily amount of gas produced from the bottles containing fresh faeces and the bottles containing VIP sludge. Univariate analysis of variance with the post-hoc Scheffe test was used to determine which samples showed significant variation from the others. Only samples with significant variability (p<0.05) are reported.

4 Results and discussions

4.1 Site survey

4.1.1 Results

This section reports the outcomes of interviews and observations conducted during initial visits to the sites included in the study.

4.1.1.1 Questionnaire-based interviews.

A total of 10 households out of about 600 households in Magwaveni Tongaat were interviewed, and thus is not statistically representative of the entire population of household in the area: The number of households surveyed in this study was not based on statistical considerations and is probably too small to allow firm conclusions to be drawn about user practices throughout the study area. The study was intended to give a qualitative indication of typical user behaviours which might impact on pit function, and should be considered in this light.

Information gathered regarding daily user behaviour is presented in Figures 4.1 and 4.2. The majority of respondents (8/10) reported throwing water into the pit, of which 7/10 added grey water only and 1/10 added both tap water and grey water. The remaining 2/10 of responding households did not add water to the pit (Figure 4.1). Grey water in this context includes water used for all types of domestic purposes such as washing dishes and laundry. Respondents were unable to provide reasons for this practice. It was concluded that the pit was used as a dumping site for grey water because there was not enough space around the households where they could dispose of their grey water. Those respondents that threw tap water into the pit assumed that adding tap water into the pit was a way of enhancing the performance of the pit. However, they were unable to explain this assumption further.



Figure 4.1: Questionnaire outcomes regarding addition of water to pits. Ten households were interviewed. Fractions on the bars indicate the fraction of households that added water to the pit, together with the types of water added, and the fraction that did not add water to the pit.

Addition of chemicals to the pit is shown in Figure 4.2. All the households visited used Jeyes fluid[®]. In addition, 2/10 of the households visited reported that Jik[®] and Domestos[®] were added to the pit as well as Jeyes fluid[®]. These chemicals were added into the pit to reduce unpleasant smell from the pit. The mode of preparation and application was similar in all cases. A small amount of any of the chemicals was diluted with water in a basin and poured into the pit. There was no specific amount that was used. The householders did not purchase the Jeyes fluid[®] in the original containers. Two different concentrations of the Jeyes fluid[®] were typically supplied in 250 mL glass bottles (nip bottles) to the householders by local vendors. Depending on the concentration, it was either regarded as "strong" or "mild". All the households used the Jeyes fluid[®] weekly. Although the Jeyes fluid[®] was used to reduce the smell, the householders reported that it temporarily reduced the smell, but did not eliminate the unpleasant smell from the pit completely. Another 2/10 of the households visited used pit latrine additive. However they were not able to recall the name of the additive used.

They explained that the additives were added to the pit once every year to reduce the heap of the contents in the pit. The additives were supplied to the householders by local vendors.





Another practice was revealed as a management or maintenance measure carried out to enhance pit performance / pit life. This is illustrated in Figure 4.3. It involved digging an adjoining pit at the side of the toilet pit. An opening was created between these two pits, and was used as an adjoining channel. The second pit was sealed so people could work over it. When the level of the waste built up to a particular height within the VIP pit, water was poured into the toilet pit and the pit contents were flushed into the adjoining pit. Although the explanation was not thorough because this activity was carried out by a hired labour, the idea was to extend the pit horizontally to prevent the original pit from filling up. Thirty percent of the households that were visited carried out this procedure as a method of pit de-sludging.



Figure 4.3 Schematic diagram of a method of pit de sludging carried out by some household using VIP toilet system

4.1.1.2 Description of the observed contents of the pit latrine

The observed characteristics of pit latrine contents differed among households. At the majority of households that were visited, there was solid refuse dumped into the pits. This included plastics, blankets and tins. In a few of the households, soil was the major constituent of pit contents. However there were also pits that contained mostly (~90 %) fresh faecal material together with toilet paper on the topmost layer, and less (~10 %) household rubbish.

An oily smell was perceived from the contents being emptied from pits that had been standing for a long while, full and not in use. This was suggested to be due to anaerobic digestion, which was supported by the dark colour of the sludge.

Some pit contents were observed to be wetter than the others. Contents from pits that were located close to a stand pipe were found to be very wet. Contents from pits located closer to the coast were drier than contents from pits located in areas further inland. Larvae were observed in contents from toilets that had broken a vent pipe when investigated from the back plate. This was probably due to passage of flies in and out of the pit, but also indicates that oxygen entered the pit, providing an aerobic environment in which flies could breed.

4.1.2 Discussions

The household survey has revealed that householders add different kinds of water, and chemicals and additives to the pit. These types of behaviours have been reported in the literature (Buckley *et al.*, 2008), and are confirmed by the questionnaire interviews reported here. Another type of behaviour of concern is the dumping of different kinds of household waste into the pit. Observations of this practice from this study were consistent with literature (Taljaard *et al.*, 2003). This confirms that onsite failure of ventilated improved pit latrines could be as a result of user behaviour-related practices. Therefore the third hypothesis as outlined in (section 1.3), *viz.* that onsite failure of ventilated improved pit latrines is a result of user behaviour-related practices, should not be rejected.

4.2 **Results obtained from laboratory analyses**

This section is subdivided into two sections. In the first subsection (4.2.1), results of physico- chemical analysis of fresh faeces and pit latrine faecal sludge are presented and discussed for all pits sampled. In the second subsection (4.2.2), results of biodegradability assays of fresh faeces and pit latrine faecal sludge (Pit 9 only) are presented and discussed.

4.2.1 Physico-chemical properties of fresh faeces and VIP sludge

4.2.1.1 Results

For the purpose of comparison, data pertaining to fresh faeces are presented before those pertaining to VIP sludge.

Fresh faeces

The average results of physico-chemical analyses of fresh faeces, together with the 95 % confidence interval, are presented in Table 4.1. COD was calculated per gram dry solids for all samples analysed in this study so that comparisons could be drawn among samples on a moisture-free basis.

The high ratio of particulate COD to soluble COD (2:1) suggests that most of the biodegradable COD which was present in the faecel samples was slowly biodegradable.

In addition, variability among faecal samples was low (*i.e.* low C of V values), presumably because samples came from individuals of same household with the same diets.

Average moisture content value shows that 78% of fresh faecal matter was water, whereas total solids value constituted 22% of fresh faeces.

The percentage organic solid (84%) of fresh faeces indicates that faeces were high in organic matter that can be biodegraded, and contained little inorganic material (16%). Inorganic material remains non-transformed in the event of biodegradation.

					95% confidence					
Parameter	Units	Ν	Mean	Std.	interval		Min	Max	C of V	
				Dev.	for mean				(%)	
					Lower	Upper				
					bound	bound				
Total COD	mgCOD/mg dry	9	1.11	0.07	0.99	1.24	1.07	1.17	8	
	sample									
Soluble	mgCOD/mg dry	6	0.37	0.05	0.32	0.42	0.32	0.43	12.8	
COD	sample									
Particulate	mgCOD/mg dry	6	0.67	0.06	0.61	0.72	0.59	0.75	8.5	
COD	sample									
Moisture	%/gwet sample	6	78	1.90	76	80	75	80	2.43	
Total Solids	%/gwet sample	6	22	1.90	20	24	20	25	8.7	
Organic	%/gdry sample	6	84	5.20	79	90	79	89	6.1	
Inorganic	%/gdry sample	6	16	5.20	11	21	11	21	-	
solids										

 Table 4.1: Physico-chemical characterisation of fresh faeces

Where N = total number of samples, Min =minimum value measured, Max= maximum value measured, C of V= coefficient of variation, indicating the extent of variability between samples Std. Dev=Standard deviation

These values are consistent with literature values. Average moisture content and organic solids obtained for fresh faeces by Lopez Zavala *et al.* (2002) were 81.8 % and 84.4 % respectively, whereas average moisture content obtained for fresh faeces by Almeida *et al.* (1999) was 79.2 %. The high ratio of particulate COD to soluble COD (2:1) suggests

that most of the biodegradable COD present in faeces is slowly biodegradable, according to Wentzel *et al.* (1999) and Orhon and Çokgör (1997). However, COD values of 1.11 mg/mg measured in this study were slightly lower than the 1.45 mg/mg that was reported in the literature (Lopez Zavala *et al.*, 2002).

Pit latrine sludge

For clarity, VIP sludge as defined in this study represent waste from the pit, but do not include domestic waste such as plastics, bottles and blankets as are usually found in these pits (Taljaard *et al.*, 2003).

Results of preliminary physico-chemical analysis of pit latrine sludge from the Tongaat area are presented in Figures 4.4 to 4.6.

Sample COD results (per mg sample) were presented on a wet and dry solids basis because there appeared to be no variation in COD load on a wet basis for the three layers sa mpled (Figure 4.4). This is due to the dilution effect by sample moisture, as explained previously. In order to show change in COD content amongst the different layers more clearly, the COD (per mg sample) values were also presented on a per-dry-solids basis. This eliminates the dilution effect by sample moisture content. On a dry basis, it can be observed that COD content was lowest in the bottom layer (0.50 mg COD/mg dry sample) and similar in the top (0.74 mg COD/mg dry sample) and middle (0.73 mg COD/mg dry sample) layers. Almost 32% COD reduction occurred between the top and the bottom layer.


Figure 4.4 Average COD of contents from three different layers within pit latrine from the Tongaat area. Error bars indicate the upper and lower boundaries of the 95 % confidence level calculated from the three replicates of the experiment. Allowance was made for the small sample size (n=3), by reading off the probability from the t-table, which was used to calculate the confidence limit.

Figure 4.5 shows average moisture content of the three different layers, whereas Figure 4.6 shows average organic solids for the three different layers. Contrary to the general trend of COD with depth in Figure 4.4, the moisture (Figure 4.5) and volatile solids (Figure 4.6) contents decreased evenly from the top layer of the pit to the bottom layer of the pit.



Figure 4.5 Average moisture value in percentage per g wet sample of contents from three different layers within pit latrine from the Tongaat area. Error bars indicate the upper and lower boundaries of the 95 % confidence level calculated from the three replicates of the experiment. Allowance was made for the small sample size (n=3), by reading off the probability from the t-table, which was used to calculate the confidence limit.



Figure 4.6 Average organic solids in percentage of contents from three different layers within pit latrine from the Tongaat area (VS: volatile solids; TS total solids). Error bars indicate the upper and lower boundaries of the 95 % confidence level calculated from the three replicates of the experiment. Allowance was made for the small sample size (n=3), by reading off the probability from the t-table, which was used to calculate the confidence limit.

For subsequent experiments, physico-chemical characteristics of pit latrine sludge were compared to those of fresh faeces. Data collected from 16 different pits were used in making the comparison. COD results and results for solids and moisture analysis for the four different layers of the 16 pits are presented.

Comparison of fresh faeces with pit latrine sludge from 16 pits

Figure 4.7 shows average COD and inorganic solids of fresh faeces and pit latrine sludge from four different layers in the pits. The mean total COD (0.54 mg COD/mg dry sample) of the top layer was significantly less than that measured for fresh faeces (1.13 mg COD/mg dry sample) (p<0.05). This is contrary from what might be expected, namely that the top layer would be similar to fresh faeces. Mean COD decreased from the topmost layer to the bottom layer, implying that most stable material was found at the bottom of the pit. Assuming that the fresh faeces analysed in this study were similar to those entering the pit, almost half of the biodegradable COD in fresh material was lost at the surface layer. This suggests that the contents of the top layer had already undergone some degree of stabilisation relative to fresh faeces. The three layers beyond the surface layer then underwent even greater stabilization since they had less COD per g dry sample compared to the topmost layer. This is reasonable considering the length of storage period of deeper layers. However, the variability between the third and bottom layer was less marked. This suggests that there was an increase in stabilisation with increase in depth.

The same trend was observed with organic solids, as shown with the points in Figure 4.7. The average percentage organic solids measured for the surface layer (58 %) was less than that measured for fresh faeces (84 %). However, this value was higher than that measured for the other layers below this point. This is as expected, since fresher material is expected at the top layer. This means that much of the biodegradable organic matter present in fresh faeces degraded naturally after defecation, but there was some partially degraded material remaining at the surface layer. For this reason, a reduction in organic solid value at the surface layer was observed relative to fresh faeces. The other layers had lost more of the biodegradable organics with time than the surface layer. The bottom layer contained the least organic matter content.



Figure 4.7 Mean total COD and organic solids composition of fresh faeces and faecal sludge sampled from four different layers (depth) within the pit of 16 VIP latrines (N=16). Error bars indicate the upper and lower boundaries of the 95 % confidence interval.

With the large sample size, (N=16), 95 % confidence limits were calculated by multiplying the standard error (SE) by 1.96. Therefore, the confidence that the sample mean is within a certain range of the population mean, due to possible sampling error, was quantified. This was done to account for the anticipated increase in sampling error that results from increase in sample size.

The plot of mean moisture content (%) of fresh faeces and the four different layers within the pit (Figure 4.8), shows that the difference in moisture content of fresh faeces and pit latrine sludge from the topmost layer was less marked (which differs from Figure 4.6 and Figure 4.8). There was marked drop in moisture content from the top layer to the 0.5 m layer and then again to the 1 m layer. By contrast, there was little to no further change from 1 m to 1.5 m.



Sample source

Figure 4.8 Moisture content (%) of fresh faeces and faecal sludge sampled from four different layers (depth) within the pit of 16 different VIP latrines. The error bars indicate the upper and lower boundaries of the 95 % confidence level.

Comparison of fresh faeces and pit latrine sludge by univariate analysis of variance (SPSS 15) showed that differences between characteristics of fresh faeces and pit latrine faecal sludge of the 16 different pits were significant (p<0.05).

Variations in physico-chemical characteristics among 16 pits Total COD

Mean total COD values were observed to decrease from the surface layer to the bottom layer (Figure 4.7). This is validated by the high R value of 0.9288, obtained by correlating mean total COD values from the four different layers with the depths. R signifies the correlation coefficient (refer to section 3.32). Analysis of variance indicates that significant differences (p<0.05) exist among all measured physico-chemical characteristics across all pits.

However, analysis of variance indicated that for all the pits, COD in the surface layer was significantly higher (p<0.05) than in the bottom layer of the pits.

Soluble COD

Mean soluble COD (SCOD) content of sludge decreased from the surface layer to the bottom layer (Figure 4.9). However, the decrease was not statistically significant (p>0.05). Soluble COD showed a similar trend to total COD (Figure 4.9). This explanation is supported by a lower R value (R=0.323) obtained for correlation analysis between soluble COD and moisture content *i.e.* compared to an R value of 0.476 obtained by correlating soluble COD content to total COD content.



Figure 4.9 Mean soluble COD of faecal sludge sampled from different layers in 16

pits. Error bars show upper and lower limits of 95 % confidence interval.

Organic solids

There was a general trend towards decreasing percentage organic solids with increase in depth (Figure 4.7). Positive correlation existed between pit depth and organic solids content (R=0.8934). Analysis of variance indicated that the surface layer was significantly higher in organic solids (p<0.05) when compared to the bottom layer of the 16 different pits. This implies that more stabilized material was located in the lower layers of the pit.

Moisture content

There is a general trend towards decreasing moisture content with increase in depth (Figure 4.8). Positive correlation existed between pit depth and moisture content

(R=0.857). However, significant variability existed and the trend was not consistent for the individual pits: Analysis of variance indicated that moisture content differed significantly (p<0.05) among the different layers across independent pits. The irregular distribution of moisture within the layers of the pit could be because moisture content depends on a range of factors, and results here suggest that pit depth is one of these. Another factor which is likely to be important is the role of ingress water.

4.2.1.2 Discussions

Comparison of physico-chemical properties of fresh faeces with pit latrine sludge Available literature is not sufficient to make comparisons to results obtained in this study for pit latrine faecal sludge. However, certain suggestions can be made based on data collected for fresh faeces data in this study, compared to available literature on the physico-chemical characteristics of fresh faeces and faeces from different sanitation systems to VIP.

Average percentage moisture obtained for pit latrine faecal sludge suggests that moisture may not be a limiting factor to anaerobic microbial activities within the pit. This is in line with Lopez Zavala *et al.* (2004b) who state that high moisture levels ($\geq 64\%$) favour anaerobic digestion processes. This also is supported by the results from Couderc (2007) who showed that increasing moisture content of VIP contents enhances the rate of stabilisation of buried organic matter within the pit.

Possible causes of variations among the different layers of the pit, based on results from 16 pits

The following discussion on pit latrine characteristics should be seen in light of the local climate in eThekwini, which is subtropical, mild to cool winters and warm summers with high humidity. Different results can possibly be obtained in another climatic zone, for instance in a dry and hot area.

Significant variability (p<0.05) was observed in both physico-chemical for the different pit latrine faecal sludge within this study.

From the results presented for pit latrine faecal sludge, it is clear that although the VIPs were sampled from areas with similar location characteristics, significant differences in organic solids content, moisture content, and COD concentration existed among samples from the same pits and samples from different pits.

Moisture content of pit sludge was unexpectedly high. A possible reason for this is the presence of ingress water. The unexpectedly high moisture content of pit materials that were sampled from the bottom layer of some pits could have been caused by a nearby source of ingress water. This may have come from the water table. If the water table is located somewhere above the pit bottom, infiltration may occur. By contrast, contents from pit latrine that are located on a steep and sandy area are most likely to have lower moisture content as water drains out through the pit walls (Buckley *et al.*, 2008).

The water retaining capability of soil in the immediate vicinity of VIP pits can also affect the moisture content of pit sludge (Psarropoulos *et al.*, 2007). This has been explained in section 2.3.2. Observed difference in moisture content of pit latrine contents that were recorded in section 4.1.2 attests to this, and was as a result of the nature of the soil of the surrounding area. For areas farther away from the coast, the soil had a realtively high clay content and so has a greater ability to retain water than the very sandy soil encountered close to the coast. Consequently, contents from pits located closer to the coast were drier than contents from pits located in areas further inland.

User practices were observed to have an effect on the nature of pit latrine contents. This is because users throw domestic water and solid waste into the pits. This concurs with other reported studies (Taljaard *et al.*, 2003).

Furthermore, the non-homogeneous property of VIP contents contributes to variation among layers of the pit. This affects the microbial response to waste components in the VIP pit. Pit latrines that have more organic waste components (such as faeces) are expected to have a higher biological activity, than pit latrines consisting mostly of inorganic waste components (*e.g.* plastic bags, bottles and sand). Addition of foreign materials to the pit by users can result in inhibition of bacterial metabolism or bacterial growth. This applies particularly to methanogens, which are the rate-limiting bacterial population in anaerobic degradation. Chemicals that kill bacteria or inhibit bacterial growth can alter the environmental conditions, thereby making the pit environment hostile for methanogens. Commercial chemicals (*e.g.* products such as Jeyes fluid®), are bactericidal to the pit biomass (Buckley *et al.*, 2008). If methanogens are killed or inhibited, volatile organic acids from hydrolysis accumulate because the rate of the production of these acids is higher than the rate at which they can be utilised by the methanogens.

Finally, the variation in characteristics for pit latrine sludge of the same layer but from different pits can also be influenced by the diet of the individuals. The overall diet of the

individuals influences biological activity of the micro-organisms in the pit. The digestibility of the diets of individuals contributing to pit contents determines the chemical composition of the faeces. Depending on the fibre content of the diet of the individuals, the biodegradability of their faeces may differ (Stanogias and Pearce, 1987).

It should be noted that data obtained from soluble COD (SCOD) characterisation for the pit latrine faecal sludge showed no significant relationship with distribution of moisture amongst the different layers. Therefore the hypothesis that the area of greater concentrations of soluble organics within the pit layers indicates the direction of hydraulic transport is rejected.

4.2.2 Biodegradability studies (Serum bottle test)

4.2.2.1 Results

The mean COD and volatile solid contents obtained from characterisation of a typical anaerobic sludge (from Northern Works Waste Treatment Plant) that was used for all the experiments are presented in Table 4.2. These were used to determine loadings in the test bottles.

 Table 4.2 Mean values for total COD and volatile solids measured in anaerobic sludge used for the serum bottle test.

Property	n	Average	Standard deviation
Total COD	6	0.1g COD/g wet	0.01
Volatile solids	3	13.8mg VS/L	0.23

Total gas produced by the controls was compared to gas production from standards bottles (Figure 4.10). Allowance was made for the small sample size (n=3), by reading off the probability from the t-table, which was used to calculate the confidence limit. It is clear that all bottles incubated with acetate (*i.e.* standards) produced significantly higher volumes of gas than the control bottles. This shows that the anaerobic sludge was active but that the inoculums had little or no biodegradable COD to be used up as an inherent substrate. Gas production from the controls ceased, while the standards were still producing gas. Gas composition was not analysed due to technical problems with the gas chromatograph.



Figure 4.10 Cumulative total gas production from bottles containing 1 g COD acetate incubated with anaerobic sludge and nutrient solution (standards) and from bottles containing anaerobic sludge and nutrient solution only (controls) to verify the activity of the sludge. Error bars indicate the upper and lower boundaries of the 95 % confidence interval (n=3).

Figure 4.11 shows the average cumulative volume of gas produced from fresh faeces, VIP sludge and controls. These results were gathered from the initial exploratory testing. The large error bars (95 % confidence interval) indicate that gas production from fresh faeces was highly variable.

For this set of experiment, all bottles containing VIP sludge did not produce gas, except one bottle that contained sample from the middle layer. Therefore only cumulative gas production from that bottle is presented for VIP sludge. VIP sludge samples were inhibitory to anaerobic digestion. This could possibly be as a result of chemicals added into the pit by the toilet owner.

In comparison, fresh faecal samples produced significantly (p<0.05) more gas than both the VIP sludge samples and the control sets. There was no observable lag period in gas production. These results indicate that a portion of faeces is readily biodegradable by anaerobic digestion. Comparisons for the data obtained amongst the different pit layers could not be made for this set of serum bottle tests, because only one bottle from one pit layer (middle layer) produced gas.



Figure 4.11 Cumulative total gas production from bottles containing 0.89 g COD fresh faeces incubated with anaerobic sludge and nutrient solution; bottles containing 0.89 g COD of Tongaat VIP sludge incubated with anaerobic sludge and nutrient solution and from bottles containing anaerobic sludge and nutrient solution only (controls). Error bars indicate the upper and lower boundaries of the 95 % confidence interval (n=3).

Figure 4.12 shows average cumulative volume of gas produced from fresh faeces, pit latrine sludge from Pit 9 of the series of pits sampled (from surface, 0.5m, 1m, 1.5m layers) and controls. All the bottles containing fresh faeces and all the bottles containing VIP faecal sludge produced gas. Therefore cumulative gas production and average nett methane volume from fresh faeces and VIP sludge samples respectively are presented (Figures 4.12 and 4.13 respectively). Gas production from fresh faeces was significantly higher than that of VIP faecal sludge samples (p<0.05). In this set of experiments, all bottles containing VIP faecal sludge were able to produce gas, even though they

contained lesser amount of oxidisable substrate (0.3g COD per bottle) compared to the bottles in the first set of experiments (with 0.89g COD per bottle). This was not expected. It was expected that the greater the amount of COD (g COD per bottle), the greater the amount of gas that would be produced. The deviation from the expected behaviour in terms of gas production pattern is thought to be because the VIP sludge samples used in the two experiments depicted in Figures 4.11 and 4.12 respectively, came from two different pits. Furthermore, the second pit (Pit 9) contained faecal sludge that had more residual biodegradable organics than the first pit (Tongaat pit). Hence the sludge samples from the second pit were able to produce more gas when incorporated into the serum bottles. Therefore, the two experiments cannot be compared to one another.

Univariate analysis of variance indicated that there was no significant difference (p>0.05) in daily total gas produced from bottles containing VIP sludge, irrespective of the layer from which it was sampled from. However post-hoc Scheffe test indicated that bottles containing fresh faeces, produced significantly higher volume of gas (p<0.05) than all the bottles containing pit latrine faecal sludge. This is supported by a higher COD content of fresh faeces compared to pit latrine faecal sludge (Table 4.3). More so, no lag period was observed with fresh faeces. These results indicate that a portion of fresh faeces was readily biodegradable by anaerobic digestion.

The experiment was terminated on day 24. However gas production had already plateued by day 20 for all the bottles.

Only biodegradability results for fresh faeces samples and samples from Pit 9 of the 16 pits described in Section 1.2.1 are presented here (Figures 4.12 and 4.13; Table 4.3), because serum bottle tests carried out using VIP sludge from other pit latrines failed. Visual inspection of the contents of the pits other than Pit 9, in addition to the lack of significant gas production, indicated that these were probably fully stabilised. Figure 4.13, shows that although all the bottles containing VIP faecal sludge produced gas, the total nett volume produced was not significantly different among the layers sampled. There was no significant difference in total gas production among the layers either. Bottles containing fresh faeces produced a significantly higher volume of methane gas than VIP sludge. The same trend was observed for total gas production.

Analysis of variance indicated that there was no significant difference (p<0.05) in the calculated biodegradability for the different layers of Pit 9 even though COD (dry basis)

of the surface layer was significantly higher than that of the bottom layer of the pit contents (Table 4.3).



Figure 4.12 Cumulative total gas production from bottles containing 0.3 g COD fresh faeces incubated with anaerobic sludge and nutrient solution; bottles containing 0.3 g COD VIP sludge (sampled from the different layers of Pit 9) incubated with anaerobic sludge and nutrient solution from bottles containing anaerobic sludge and nutrient solution only (controls). Error bars indicate the upper and lower boundaries of the 95 % confidence interval (n=3).



Figure 4.13 Average nett methane volume produced from samples as per Figure 4.12, after subtraction of the volume of gas produced by blanks from volume of gas produced by serum bottles containing 0.3 g COD fresh faeces and by bottles containing 0.3 g COD pit latrine faecal sludge, respectively, after 20 days.. Pit latrine sludge was sampled from four different layers (by depth) of Pit 9. Error bars show the upper and lower boundaries of the 95 % confidence interval (n=3).

Table 4.3: Calculated percentage biodegradability of pit latrine sludge, sampled from pit 9 at different depths (as per Figure 4.12). Results are presented as mean value ± standard deviation, n=3 for all results reported for VIP sludge, and n=5 for all results reported for fresh faeces.

Property	Fresh	Surface	0.5m layer	1m layer	1.5m layer
	faeces	layer			
COD g/g dry	1.11±0.07	0.72±0.01	0.38±0.00	0.25±0.028	0.23±0.00
Sample					
Percentage	70±16.73	11.11±2.88	12.64 ± 6.88	11.70 ± 1.93	6.28±0.01
biodegradability					

One reason for the low gas production overall in results reported here for the anaerobic biodegradability test could be because gas production was measurable in a single full pit that had not been in use and had been standing for an unspecified period of time. It is possible that all the readily biodegradable COD within the pit contents had been degraded, leaving behind only the slowly and the non-biodegradable COD. Similarly, the lack of gas production from the remaining 15 pits could have been the result of a long standing period during which all the biodegradable COD had been degraded prior to sampling. But it is important to note that gas production measurements for Pit 9 (Figures 4.11 and 4.12) showed large variability even though they contained the same amount of sample on the basis of COD. Furthermore, although inhibition of Tongaat VIP samples (Figure 4.11) to anaerobic digestion was tentatively attributed to chemical agents added to the pit, this was not confirmed. In addition, differences in biodegradability were expected among the pit layers. These were not observed. The cumulative evidence suggests that unsuitability of the anaerobic biodegradability test for VIP sludge should not be ruled out.

Since only Pit 9 exhibited methanogenic activity, the sample size was too small, to draw conclusions on overall biodegradability of VIP sludge.

Results presented here for the serum bottle test, as performed on VIP sludge, suggest that the hypothesis that this test can be used to measure anaerobic biodegradability of pit latrine faecal sludge should not be accepted. This is considered further in the Discussion below.

The average biodegradability obtained for faeces in this study showed that 70 % of total COD in fresh faeces was biodegradable and 30 % was non-biodegradable. Analysis of variance conducted to compare the biodegradability values obtained for fresh faecal samples and pit latrine faecal sludge, showed that biodegradability of faeces differed significantly (p<0.05) from that of VIP sludge. This implies that the faeces undergo degradation during the time they are inside the pit. Furthermore, evaluation of anaerobic biodegradability of fresh faeces by the serum bottle test appears to be a valid technique, unlike indications from the same test performed on VIP sludge.

4.2.2.2 Discussion

Comparing results for both physico-chemical properties and (reported in Section 4.2.1 above) and biodegradability of fresh faeces and pit latrine faecal sludge, these values

were consistently higher for fresh faeces than for pit latrine contents. These observations can be explained in terms of changes with time and environmental factors, such as moisture, temperature, carbon content and nutrient availability, that occur between fresh faeces immediately upon deposition in the pit and the contents of the top layer of the pit (Nordin, 2006).

Significant similarity was observed for data on the physico-chemical characteristics of faeces obtained within this study and that of literature. Results obtained in this study showed that, in general, fresh faeces were biodegradable. This, too, agrees with the literature, that raw faeces are capable of undergoing natural breakdown (Lopez Zavala *et al.*, 2002; Lopez Zavala, 2005; Hotta and Funamizu, 2007; Buckley *et al.*, 2008). Also, Conversely, the outcomes of the serum bottle tests from this study did not indicate that the VIP sludge was significantly anaerobically biodegradable. This last result is unexpected.

There are two possible interpretations of this outcome. One is that VIP pit contents indeed do not undergo significant biodegradation under the conditions in pit latrines, which are predominantly anaerobic. The other is that the serum bottle test is not suitable for the measurement of biodegradability of pit latrine faecal sludge. The following discussion considers the evidence for each of these possible explanations.

Evidence for biodegradability of pit latrine sludge

Previous experiments which formed part of a larger Water Research Commission project entitled "Scientific Support for the Design and Operation of Ventilated Improved Pit Latrines" (VIPs), showed that reduction in the volume of organic material in pit latrines is possible to some extent if the environmental conditions are conducive for the appropriate micro-organisms to thrive (Buckley *et al.*, 2008).

This is not in accordance with the outcomes of the serum bottle tests from this study. This suggests that pit latrine faecal contents are indeed biodegradable and hence that present results point to the unsuitability of the serum bottle test for VIP sludge samples.

Evidence for suitability of serum bottle test for measurement of biodegradability of pit latrine sludge

The serum bottle test, if conducted under conditions of ideal temperature, pH, and alkalinity (owing to nature of the substrates inside the bottles), has been shown to be suitable for monitoring the anaerobic degradation of substrates (Angelidaki *et al.*,

2008). However, the suitability of the serum bottle test method in testing the biodegradability of the samples depends on whether samples are able to produce sufficient volume of methane to calculate their biodegradability, and results for gas production measurements do not have large variability (high standard deviation). Gas production measurements in the present study (Figures 4.14 and 4.15) showed large variability for cumulative volume of gas produced for triplicates, even though they contained the same sample. This was the case for both fresh faeces and VIP sludge sampled from the different layers within the pit. Also, the experiment using 0.89 g COD of Tongaat VIP sludge revealed that volume of methane produced by the samples from the test was not sufficient to calculate biodegradability of samples. This suggests that the serum bottle test was not suitable for measuring the biodegradability of the VIP faecal sludges investigated in this study.

Results in Table 4.3 show no marked difference in biodegradability of pit latrine faecal sludge sampled from four different layers within the pit. Using analysis of variance, the initial COD (on dry mass basis) of the pit layers differed significantly (p<0.05), with the surface layer being higher in COD than the bottom layer. These results do not agree with those of a parallel study to this by Bakare (unpublished). In that study, an aerobic biodegradability assay (aeration test) was carried out on the *same* VIP samples that were used in the present study. Bakare showed a strong relationship between results of the aerobic biodegradability assay and those obtained for COD and volatile solids measurements, *i.e.* the same trend was observed for these three measurements amongst the four different layers of the pit. Using analysis of variance, a significant decrease (p<0.05) in COD, volatile solids and biodegradability was demonstrated from the topmost layer to the bottom layer was demonstrated. This significant decrease in biodegradability was not observed with anaerobic serum bottle tests as performed in the present study, and is strong evidence that the serum bottle test is not suitable for pit latrine faecal sludge samples.

Therefore the hypothesis that serum bottle test technique can be used to measure the anaerobic biodegradability of both fresh faeces and pit latrine faecal sludge should be rejected.

Study limitations

The major limitations of this study relate to the non-homogenous nature of the samples. Although efforts were made to homogenise the samples before adding them to the bottles, the difference in factors such as the initial availability of organic matter possibly contributed to high variability of results in replicates, as demonstrated by the large error bars in Figures 4.14 and 4.15.

Non-homeogeneity of samples incorporated in individual replicates relates to nonhomogeneity in the source material. For VIP samples, it is possible that in some cases a part of the sample that was incorporated into a replicate bottle may not have been of faecal origin, and in worst cases may not have been organic in nature at all. A bottle containing sample of such nature would have had low gas production ability if compared with other replicates of predominently organic nature and of faecal origin. For fresh faecal samples, it is possible that sample incorporated into one of the replicate bottles was completely slowly biodegradable, while the majority of the sample in the others, constituted readily biodegradable organics (although readily biodegradable organics in fresh faeces are reported to be negligible - Lopez Zavala *et al.*, 2004a). This type of limitation can be avoided if larger volumes of samples could be tested at a time. However, this is not possible with this type of test, because the bottles are small (125 mL) in size.

Another limitation of this study could have been the inoculum used. The activity of the biomass is crucial to the success of the test, and can be affected by factors such as the source and the age of the sludge. Although efforts were made to ensure that the anaerobic sludge used came from a source fed with substrates of faecal origin and also was active, VIP faecal sludge samples were still inhibitory to anaerobic digestion, and so were unable to produce sufficient volume of methane to calculate biodegradability of samples.

Evidence presented above strongly supports the conclusion that the biodegradability values obtained for the VIP sludge samples in this study were low because of unsuitability of the serum bottle test, and not because samples could not be biodegraded. Therefore, it is recommended that an alternative method is used for measuring the biodegradability of VIP sludge samples.

5 Development of a model description of biological activity in the different layers within the pit

At the outset of this study, it was hoped that measurements of anaerobic biodegradability of fresh faeces and of VIP contents at several depths would allow construction of a theoretical model of biological activity in a pit. Unfortunately, this was not possible in light of the results obtained. Results of biodegradability measurement of fresh faeces have been presented in Chapter 4. Biodegradability data obtained for pit latrine faecal sludge could not be used to make general conclusions about processes in pit latrines since only Pit 9 of the 16 pits investigated exhibited methanogenic activity. As a result of this, the sample size was too small (n=3) for statistically valid generalisations based on biodegradability.

However, data gathered for COD and organic solids measurements can be used as a substitute for biodegradability measurements to provide the basis for a model of biological acitvity in VIP pits. This is in line with the hypothesis that the measurement of physico-chemical and biological characteristics of VIP sludge and fresh faeces allows conclusions to be drawn regarding the extent of biodegradation that has occurred in a pit latrine faecal sludge. The model description of biological activity in the different layers in the pit latrine, developed on the basis of measurements of COD and organic (volatile) solids, is presented in this chapter.

The results obtained so far suggest that changes do take place in the biodegradable organic material of faecal sludge found in a pit latrine with time. COD measurements suggest that immediately after defecation, rapid degradation occurs during which microorganisms inherent in the faecal matter and those present at topmost layer of the pit contents carry out a quick aerobic degradation of the readily biodegradable COD in fresh faeces. Consequently, there is a loss of a large fraction of the biodegradable components in faeces through aerobic degradation while it is sitting at the topmost layer of the pit contents. This justifies, the applicability of the aerobic biodegradability test. Furthermore, this aerobic biological activity occurs on the top surface of the pit, before the "fresh" faeces are overlaid with new material. This layer is too small to be easily be sampled. Once it is overlaid by new pit material, anaerobic degradation occurs at a far slower rate, resulting in gradual reduction of organic material. This is supported by the following observations. In comparing COD value of fresh faeces to that of faecal sludge at the surface layer of the pit, it was shown that about half of the COD of fresh faeces had already been lost. Further reductions in COD were seen in the other layers beyond the surface layer, compared to the surface layer.

Thus a description of biological activity in the different layers in pit latrine sludge within a pit is proposed. Moving from the top of the pit downward, the following layers can be distinguished:

- i. A first layer that is too small to be measured and is composed of fresh faeces where the readily biodegradable components are still unchanged.
- ii. A second layer made up of the topmost aerobic part of the pit, where aerobic degradation of hydrolysable organic material takes place at a rate limited by the aerobic hydrolysis of large organic molecules into simpler compounds.
- iii. A third layer that is anaerobic as a result of the elimination of oxygen by the covering material, where anaerobic digestion of hydrolysable organic material takes place at a rate limited by the anaerobic hydrolysis of large organic molecules into simpler compounds.
- iv. A bottom layer, where no further stabilisation of organic material can occur as materials are already stabilised.

This is shown diagrammatically in Figure 5.1.



Figure 5.1: Diagram of the theoretical model of biological activity in a pit latrine showing the proposed layers: (i) fresh faeces; (ii) partially degraded aerobic surface layer; (iii) partially degraded anaerobic layer beneath the surface; (iv) completely stabilised anaerobic layer. Source: Buckley *et al.* (2008)

6 General Discussion, Conclusions and Recommendations

The main focus of this study was to obtain information on the stabilisation processes in VIP contents (pit latrine faecal sludge). The method devised to obtain a scientific understanding of the biological conversion of the pit latrine faecal sludge was based on the assumption that the characteristics of pit latrine contents at varying depths can be related to their extent of conversion. Faeces is the major organic feed material that goes into the pit, and so a baseline assessment of physico-chemical properties and biological characteristics of fresh faeces and pit latrine faecal sludge were measured in this study.

User practices are amongst the most important factors affecting the properties of the contents of the pit. This was addressed in the present study through an informal questionnaire survey of a small sample of households in the area from which pit latrine samples were collected.

Biodegradability measurement of pit latrine faecal sludge using the serum bottle test showed little gas production from samples, which was assumed to be due to an inhibition of anaerobic digestion under the test conditions. It was concluded that the serum bottle test was not suitable for measuring biodegradability of VIP sludge in this study. Future studies on biodegradability of VIP sludge are recommended, in which the aerobic biodegradability test is used in place of the serum bottle test, since the ultimate goal is to quantify biodegradability of samples irrespective of the environmental conditions (*i.e.* quantifying anaerobic biodegradability of the samples is not specifically required). The aerobic biodegradability test allows the use of larger volume of sample, and no inoculums are required. The results obtained from the different assays (i.e. aerobic and anaerobic) must be compared. Biodegradability should also be compared to physical tests such as COD and volatile solids measurements, because these tests can be used as indirect substitute test for biodegradability analysis. In other words, COD and volatile solids measurements can be used to measure the biodegradable organic content of a material, and also potential changes that have occurred in the organic content of the material over time.

Although the questionnaire survey was an informal one and despite the small number of respondents used for this survey, data obtained was sufficient to indicate qualitatively the experiences of VIP owners and maintenance approaches towards the system.

The questionnaire survey revealed daily user operational practices amongst the households, which are carried out in ignorance by householders with the intention of improving the conditions of the pit, and especially to reduce the volume of the pit contents, and to reduce unpleasant smell from the pit. The survey revealed that VIP users around eThekwini Municipality throw household wastes and different kinds of greywater, in addition to tap water, into the pit. Consequently, depending on what is dumped into the pit, the physical, chemical and biological properties of contents from pit latrines can vary. Inadequate education of users about the importance and purpose of the VIP latrine is most likely a cause of these attitudes. Users need to be educated on how to use the VIPs, if they expect them to accomplish the requirements of improved sanitation.

Although lack of adequate user education may have contributed to the scenario, the majority of bad practices by VIP users may have originated from what users believe or make out their needs. They tend to use the VIP to satisfy their needs, thereby affecting their daily routines when using these toilets. Households that are located far way from an efficient solid waste removal system are most likely to use the pit, not only for defecation, but as a dumping site for household solid waste. Dumping of solid waste accelerates pit filling, thereby preventing the pit from providing a functioning system for improved sanitation. To counteract filling, an informal means of pit desludging is commonly practised. In order to promote good behavior and hence reasonable pit filling rates and efficient functioning of pits, users should be educated not to dispose of their household solid waste into the pit. This can only be achieved in practise if authorities provide an efficient solid waste removal system that is accessible to all VIP owners.

This research also revealed that VIP owners usually add chemicals and pit latrine additives alleged to reduce smell and pit heap respectively into the pit. There have been no scientific evidence that supports the efficacy of these pit latrine additives. This suggests that users collect information from inappropriate sources. It is essential that correct information on the operation and maintenance of the ventilated improved pit latrine is transferred. Proper training in operation and maintenance is important, and should involve both users and implementing agents.

However, the pilot questionnaire study reported here is not without limitations. Outcomes of the questionnaire survey give an indication, but not conclusive understanding, of the experiences of VIP owners and maintenance approaches towards the system. Firstly, sample size was not statistically representative of the community sampled (only 10 households out of a population of approximately 600) and certainly not of the entire population of VIP users in eThekwini Municipality, because it is small. Therefore, it cannot be assumed that similar practices were carried out by VIP users throughout the eThekwini Municipality. Finally the demographic homogeneity of this survey excludes the generalization to more diverse VIP users and practices, such as in other countries. However, the results of this survey serve as a basis for broader exploration. It is recommended that this survey be considered as a pilot survey, to be confirmed by studies including larger number of households.

The laboratory investigation of the physico-chemical characteristics and biodegradability measured for both pit latrine faecal sludge and fresh faeces leads to the following conclusions regarding the nature of pit latrine contents and appropriate methods of measuring these:

- There was a regular decrease in total COD content, moisture content and organic solids fraction with increase in the depth of pit contents.
- Contrary to expectation, there was no significant correlation between the concentration of soluble organics and the mositure gradient in pit contents.
- Also contrary to expectation, the serum bottle test for anaerobic biodegradability proved to be unsuitable for testing the biodegradability of pit latrine contents.
- A description of biological activity in the different layers in faecal sludge found within a pit latrine was developed, based on measures of physico-chemical characteristics of pit contents at varying depths.
- Large variations exist in the physical and chemical composition both within a pit and from different pits.

The information on the nature of pit latrine contents gathered in this study provides a platform for decision-making in terms of managing pit latrines during their normal lifespan, and managing pit emptying and associated sludge management.

Management of pit latrine during normal life span encompasses efforts that should be made to improve the ability of the VIP to provide an improved sanitation service to the users of the latrine: Proper education of users is fundamental, to ensure that pit latrines are properly used and maintained, thereby eliminating problems associated with VIP latrines including rapid filling rates. Furthermore, users should recognize that a pit latrine is a biological system and therefore avoiding addition of potentially bactericidal additives, or activities that can interfere with the biological activity in the pit.

Besides, the municipality must consider the potential role of ingress water before pit can be located.

From the conclusions of this study, most biological activity occurs at the surface and upper layer of the pit, therefore any interventions aimed at improving the lifespan of pits should be most active at these points.

In addition, this study concluded that conditions vary among pits, therefore interventions to improve the lifespan of pits must be sufficiently flexible to adapt to a range of conditions.

Because of variability among pits, a reliable form of testing the extent of stabilisation of particularly the upper pit layers is needed. Physico-chemical characteristics are more reliable in this respect than anaerobic biodegradability.

On the other hand, sludge management plans involve management and control of emptied pit latrine sludge in an environmentally sustainable way, and should consider all re-use options, storage and transportation based on the sludge quality. It is clear from what has been obtained in this study that the nature of pit latrine contents varies within a pit latrine and between different pit latrines. These findings are important because they highlight the reality that effective management of pit latrine sludge requires different approaches that are dependent on the nature of the pit contents. Proper management is necessary to prevent negative impacts of feacal sludge on health and environment:

Information about pit latrine sludge characteristics is essential for design and improvement of pit latrine system. As faecal sludge characteristics have been shown to vary for different pits depending on many factors, it is most likely that similar variation is also inherent to other types of on-site sanitation facilities. Therefore, one cannot rely on literature data only for design parameters such as organic load and inherent biodegradability. These parameters may be quite different from other recorded cases and are likely to vary widely, both among geographical locations and among pits at any given location.

Also, treatment requirement of exhumed faecal sludge is usually based on the solids content of the sludge. The degree of stabilization of the sludge indicates whether further digestion of sludge is necessary. This is measured by the COD and volatile solids content of the sludge. The conclusion of this study suggests that samples from the upper layers may require further digestion to achieve a similar degree of stabilisation as the bottom layer samples.

Faecal sludge is full of all infective organism excreted with human faeces (give a reference here). The load of potentially infective micro-organisms present in the pit latrine faecal sludge was not measured in this study. However, the load of the infective micro-organisms is linked to the degree of stabilisation of the pit contents: the greater the degree of stabilisation, the more likely it is that significant die-off of potentially harmful micro-organisms has occurred. Furthermore, the potential for infection is a function of both the load of potentially infective organisms and the exposure of humans to these organisms. Handling of sludge co-occurs during pit emptying. Baseline information on the properties of the pit latrine sludge determines how sludge should be handled. Hygiene and safe handling is necessary to prevent disease transmission.

Proper management plans for pit latrine sludge are essential for the safety of both humans and the environment. Exhumed faecal sludge, if not formally managed, may be discharged into the environment without considering the physico-chemical quality of the sludge (give a reference here). This can lead to the pollution and deterioration of the surrounding environment.

Finally, although, faecal sludge is a potential good organic fertilizer and soil conditioner and can therefore be re-used in agriculture (Klingel et al., 2002), it is not hygienically safe, if it not biochemically stable before use. From the conclusions given above, it can be suggested that samples from the bottom layer of the pit is likely to be safer to be handled and potentially re-used than the upper layers of the pit , because the bottom layer of the pit is the most stable.

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Appendices

Appendix A: List of materials used for Laboratory analysis and site visits

Materials used for sampling pit latrine faecal sludge from the field

- Shovel
- 15 Honey jars (~300mL each)
- Refuse bags (~400 mm ×900 mm)
- Latex Gloves

Materials for determination of solids

- Prepared 100mL Porcelain crucible (90 mm in diameter)
- 30 g of each sample
- Desiccator with silica crystals as desiccant
- Balance (Metler, model; AE 160 fine balance)
- Oven (Gallekampt Hotbox Oven size 1) at 105° C
- Muffle furnace at 550 ° C

Materials used for measuring Total Chemical Oxygen Demand

- 1 L Pyrex glass volumetric flask
- Three I L round bottom Pyrex flask
- 10 mL pipette
- 50 mL burette and stand
- Glass condenser
- Twelve 250 mL flat bottom volumetric flasks, each with ground glass 24/29 neck
- Twelve glass condensers with 24/29 ground-glass joint
- Hot plates (Labcon HPE 3118U)
- Concentrated sulfuric acid solution (H₂SO₄) (Chemically pure)
- 98% sulphuric acid reagent
- 0.0417 M potassium dichromate reagent (K₂Cr₂O₇).
- 0.04 g mercury sulphate

- 0.25 M ferrous ammonium sulphate reagent (FAS)
- Distilled water
- 50 g of each sample
- Electrical blender
- Ferroin indicator

Extra materials used for measuring Soluble and Particulate Organic content (Soluble and Particulate Chemical Oxygen Demand)

- Centrifuge (Beckman J2-21 with JA10 rotor)
- 1L Pyrex measuring cylinder

Materials used for serum bottle test

- 18 serum bottles
- Rubber septa
- Anaerobic sludge
- Nutrient solution
- Measured mass of sample of known COD
- Distilled water
- Gas mixture of 50% CO₂ and N₂ each
- 50mL glass syringe
- Gas chromatograph

Appendix B Reagent and equipment preparation

Reagents

0. 417 M (K2Cr2O7) Potassium dichromate solution

Procedure

- K₂Cr₂O₇ (primary standard grade, 12.259 g) was dried in an oven at 103 ° C for 3 hours.
- This was allowed to cool in a dessicator and then dissolved in distilled water and made it up to 1000 mL in volumetric flask.

0.25 M Ferrous ammonium sulphate [Fe(NH4)2(SO4)2]

Procedure

- 15 g of silver sulphate (Ag₂SO₄) were dissolved in 2 500 mL of concentrated (>98 %) H₂SO₄
- The solution was stirred using a magnetic stirrer and allowed to stand for 3 days.

Ferroin indicator

Procedure

1.485 g of 1,10 phenanthroline monohydrate and 695 mg of FeSO_{4.}7H₂O was dissolved in distilled water, and diluted to 100 mL.

Mecuric Sulphate (HgSO4) solution

Procedure

40 g of red mercuric oxide (HgO) was dissolved in 250 mL of 1 H₂SO₄ : 5 water (50 mL :250 mL) and diluted to 800 mL with distilled water.

Sulphuric acid (H_2SO_4) – mercuric sulphate $(HgSO_4)$ – potassium sulphate (K2SO4) solution.

Procedure

- 333.75 g of K2SO4 was dissolved in 1 800 mL of distilled water and to it, 500 mL of concentrated H₂SO₄ .was added.
- To the mixture above, 62.5 mL of the HgSO4 prepared in above.
- Dilute to 2 500 mL with distilled water.

7 N Sulphuric acid (H2SO4)

Procedure
435 mL of concentrated H₂SO₄ (98%) was diluted in distilled water and made up to 2 500 mL using distilled water.

The nutrient medium was prepared as described in Remigi and Buckley, (2005).

Composition of stock solutions used for the preparation of the nutrients medium incorporated into the bottles. Source: Remigi and Buckley, (2005).

Stock solution	Composition	Concentration (g/L)
S2	Resazurin	1
S3	(NH ₄)2HPO ₄	26.7
	CaCla 2HaO	167
		26.6
	ΝΠ ₄ C1	20.0
	MgCl ₂ .4H ₂ O	120
	KCl	86.7
	MnCl ₂ .4H ₂ O	1.33
	CoCl ₂ .6H ₂ O	2
S4	H ₃ BO ₃	0.38
	CuCl ₂ .2H ₂ O	0.18
	NaMoO ₄ .2H ₂ 0	0.17
	ZnCl ₂	0.14
S5	FeCl ₂ .4H ₂ O	370
S6	Na ₂ S.9H ₂ O	500
	Biotin	0.002
	Folic acid	0.002
	Pyridoxine hydrochloride	0.01
S7	Riboflavin	0.005
	Thiamine	0.005
	Nicotinic acid	0.005
	Panthotenic acid	0.005

Equipments

Gas chromatograph (GC)

The gas composition in the serum bottle was analysed by gas chromatography.

Specifications of the GC

Туре	GOW-MAC 350,
Detector type	Thermal Conductivity Detector
Carrier gas	Helium
Calibration gases	nitrogen, methane, and carbon dioxide production
Column type	Haysep D stainless steel column

Separation conditions:

- Gas pressure 400 k Pa
- Gas flow rate of 40mL/min.
- Column temperature of 80°C
- Detector and injector port temperature at 95°C
- Detector bridge current at 100 m A

The retention times for nitrogen, methane and carbon dioxide under these conditions were approximately 0.98, 1.31 and 1.84 min respectively. Clarity Lite[®] software was used to analyse the chromatographs.

GC Calibration

Calibration curves were prepared by injecting volumes $(20 \ \mu l \ to \ 100 \ \mu l)$ of calibration gas into the GC. Assuming that the peak area is proportional to the quantity of individual gasses injected, the peak areas were plotted against the volumes injected. It is recommended that the r² value be closest to unity.

		H	IOU	SE							
		NUMBERS									
QUESTIONS	OPTIONS	1	2	3	4	5	6	7	8	9	10
User-behaviour questions											
How many people does the pit	weekly										
latrine serve											
	weekends										
	on average										
How many of these people are children	Give number										
How many of the users	reside on-site										
	are visitors										
How long have you been using the	months										
pit											
	years										
Do you throw wash water into the pit	Y										
	Ν										
Do you throw other domestic	Y										
water into the pit											
	Ν										
Management related questions											
Have the pit been emptied before	Y										
	Ν										
If Y when was the last emptying											
What type of pit emptying	manual										
	mechanical										
How often do you empty the pit											
Have you been adding	Domestos										
-	Jeyes fluid										

Appendix C: Questions used for the survey as input into Excel sheet

	Jik
Give reason for using any of these	
How often do you use	Domestos
	Jeyes fluid
	Jik
Have you been adding any other chemical	Y
	Ν
Give the name for the chemical	
Give reason for using this chemical	
How often do you use this chemical	
Where do you buy	Chemical (additive)
	Jik / Domestos / jeyes fluid
When do you buy them	month end
	more regular
Do you keep or dispose the empty container	
How do you dispose the empty containers	
When was the last time you added	
Have the pit ever been adjusted to	Y
enhance its performance	Ν
How	
Do you know of any other ways	Y
	Ν
Do you experience unpleasant	Y

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odour from pit

	Ν
Do you know any way to make	
toilet smell better	
What can you say about the	
general performance of the pit	
toilet	
Technology/Construction	
questions	
Does the toilet have a door	Y
	Ν
Is the door broken	Y
	Ν
If door is broken ,who fixes it	
Does it have a fly screen	Y
	Ν
Is there a water inlet from sides of	Y
the pit	
	Ν
Does any rain or storm water enter	Y
the pit	
	Ν
Through where, if Y	
Is toilet built in an accessible place	Y
	Ν
Does the pit have a cover	Y
	Ν
Did you seal the back flaps	Y
	Ν
if Y why	
Are you worried about the pit	Y
filling up fast	
	Ν
If Y what can you do about it	
Observations	
Does the toilet have a hand	Y

cleaning material

	Ν
Do they use soap to wash their	Y
hands	
	Ν
Do they throw the hand-wash	Y
water into the pit	
	Ν
General condition of the toilet	Clean
inside	
	Unclean
Describe the condition of the toilet	
inside	
General condition of the toilet	Clean
outside	
	Unclean
Describe the condition of the toilet	
outside	

Appendix D: Stepwise estimation of the equivalence in mass of sample added into the bottles

If sample COD = X g COD / g wet sample

Sample amount in g COD / bottle:

Initial mass of bottle = A

Final mass of bottle after adding sample (but not capped) = B

Mass of sample in bottle (C) = B - A

Thus sample COD per bottle = $(X_gCOD / gsample \times C)$

= YgCOD/bottle

Calculate volume of methane (L) added to bottles at STP

 $1 \text{ g COD} = 0.35 \text{ L CH}_4$

Thus vol. of CH₄ added at STP per bottle:

$$= YgCOD/bottle \times 0.35LCH_4$$
$$= Z L CH_4$$

Calculate vol. of CH_4 at working temperature (T_w) and pressure and assuming working pressure = 1atm

 $= Z L CH_4 \times (273.15 + T_w) / 273.15$

Appendix E: Tables of data obtained within this study and used for plotting the <u>Figures.</u>

Results obtained for moisture content, solids content and COD analysis for samples obtained from top middle and bottom layer of VIP in Tongaat area. Results are presented as value ± standard deviation. n=3 for all results reported.

				Sample ID					
Property	7			Тор	Middle	Bottom			
Average	Moistu	re (%)		79.74±0.07	72.81±1.54	65.80±3.58			
Average volatile solids (%)				61.54±5.05	52.95±2.33	41.92±2.08			
Sample	total	COD	(mg COD/g wet	150±0.09	199±0.04	172±0.05			
sample)									
Sample	total	COD	(mg COD/g dry	738±0.09	733±0.04	503±0.05			
sample)									

Values obtained for mean total COD in mg/mg dry samples COD for fresh faeces against VIP samples from 4 different layers of the 16 different pits. Results are presented at n=48 for all results reported.

Sample	Mean	Std.	Std.	95% Confidence		Mini-	Maxi-
source		Devi-	Error	Interval for		mum	mum
		ation		Mean			
				Lower	Upper		
				Bound	Bound		
fresh faeces	1.11	0.05	0.03	0.99	1.24	1.07	1.17
Pit surface layer	0.57	0.25	0.04	0.50	0.64	0.12	1.22
0.5 m layer of							
pit	0.38	0.12	0.02	0.35	0.42	0.05	0.56
1m layer of pit	0.25	0.12	0.02	0.22	0.29	0.09	0.55
1.5 m layer of							
pit	0.25	0.13	0.02	0.21	0.28	0.09	0.49

Values obtained for mean solids in % g VS per g wet sample/ g TS per g wet sample for fresh faeces against VIP samples from 4 different layers of the 16 different pits. Results are presented at n=48 for all results reported.Organic

Sample	Mean	Std.	Std.	95% Confidence		Mini-	Maxi-
source		Devi-	Error	Interval for Mean		mum	mum
		ation		Lower	Upper		
				Bound	Bound		
fresh faeces	84.26	5.20	2.12	78.81	89.72	79.27	89.25
Pit surface layer	57.89	14.92	2.15	53.56	62.22	23.60	94.64
0.5 m layer of	47.74	17.50	2.53	42.66	52.82	3.67	75.62
pit							
1m layer of pit	33.95	16.53	2.39	29.15	38.75	4.89	73.57
1.5 m layer of	36.57	17.82	2.57	31.40	41.74	3.94	74.46
pit							

Values obtained for mean moisture in % perg wet sample for fresh faeces against VIP samples from 4 different layers of the 16 different pits. Results are presented at n=48 for all results reported.

Sample	Mean	Std.	Std.	95% Confidence		Mini-	Maxi-
source		Devi-	Error	Interval for		mum	mum
		ation		Mean			
				Lower	Upper		
				Bound	Bound		
fresh faeces	78.20	1.90	0.78	76.20	80.19	75.25	80.06
Pit surface layer	76.88	5.68	0.82	75.23	78.52	57.58	85.71
0.5 m layer of pit	71.63	11.18	1.61	68.39	74.88	30.06	86.06
1m layer of pit	64.60	12.33	1.78	61.02	68.18	30.72	84.83
1.5 m layer of pit	67.22	12.56	1.81	63.57	70.86	34.71	87.48

Layer	Pit	N	Mean	Std.	Std.	95% Co	95% Confidence		Maxi -
	no.			Devia	Error	Interva	l for	mum	mum
				-tion		Mean			
						Lower	Upper		
						Bound	Bound		
surface	1	3.00	0.37	0.07	0.04	0.20	0.54	0.29	0.42
	2	3.00	0.45	0.02	0.01	0.39	0.51	0.44	0.48
	3	3.00	0.37	0.08	0.05	0.17	0.57	0.27	0.42
	4	3.00	1.15	0.07	0.04	0.98	1.32	1.08	1.22
	5	3.00	0.49	0.02	0.01	0.43	0.55	0.47	0.52
	6	3.00	0.64	0.12	0.07	0.34	0.94	0.50	0.71
	7	3.00	0.33	0.04	0.02	0.22	0.43	0.28	0.36
	8	3.00	0.70	0.01	0.01	0.68	0.73	0.69	0.71
	9	3.00	0.72	0.00	0.00	0.71	0.73	0.72	0.73
	10	3.00	0.59	0.01	0.00	0.58	0.60	0.58	0.59
	11	3.00	0.12	0.01	0.00	0.11	0.14	0.12	0.13
	12	3.00	0.40	0.10	0.06	0.15	0.65	0.30	0.50
	13	3.00	0.81	0.04	0.02	0.71	0.91	0.77	0.85
	14	3.00	0.55	0.04	0.02	0.47	0.64	0.52	0.59
	15	3.00	0.86	0.06	0.03	0.71	1.01	0.83	0.93
	16	3.00	0.57	0.07	0.04	0.39	0.75	0.49	0.63
	Total	48.00	0.57	0.25	0.04	0.50	0.64	0.12	1.22
0.5m	1	3.00	0.39	0.02	0.01	0.34	0.44	0.37	0.40
	2	3.00	0.28	0.01	0.01	0.24	0.31	0.26	0.29
	3	3.00	0.41	0.00	0.00	0.40	0.41	0.40	0.41
	4	3.00	0.39	0.01	0.01	0.36	0.42	0.38	0.41
	5	3.00	0.30	0.01	0.01	0.27	0.32	0.29	0.31
	6	3.00	0.43	0.02	0.01	0.38	0.47	0.41	0.45
	7	3.00	0.06	0.01	0.01	0.04	0.08	0.05	0.07
	8	3.00	0.53	0.00	0.00	0.52	0.53	0.53	0.53
	9	3.00	0.38	0.00	0.00	0.38	0.38	0.38	0.38
	10	3.00	0.55	0.00	0.00	0.55	0.56	0.55	0.56
	11	3.00	0.41	0.09	0.05	0.18	0.64	0.33	0.51

Total COD (in mg/mg dry sample) distribution for faecal sludge sampled from different layers within the same pit and sampled from different pits

	12	3.00	0.41	0.02	0.01	0.36	0.47	0.39	0.43
	13	3.00	0.39	0.04	0.02	0.29	0.49	0.34	0.42
	14	3.00	0.31	0.10	0.06	0.07	0.55	0.20	0.39
	15	3.00	0.34	0.03	0.01	0.28	0.40	0.31	0.36
	16	3.00	0.52	0.03	0.02	0.46	0.59	0.50	0.55
	Total	48.00	0.38	0.12	0.02	0.35	0.42	0.05	0.56
	1	3.00	0.17	0.01	0.01	0.14	0.20	0.16	0.18
	2	3.00	0.22	0.02	0.01	0.18	0.27	0.21	0.24
	3	3.00	0.39	0.03	0.02	0.31	0.47	0.36	0.42
	4	3.00	0.33	0.01	0.00	0.31	0.34	0.32	0.33
	5	3.00	0.13	0.02	0.01	0.08	0.18	0.11	0.15
	6	3.00	0.18	0.02	0.01	0.13	0.23	0.16	0.20
	7	3.00	0.26	0.01	0.01	0.23	0.28	0.24	0.27
	8	3.00	0.42	0.00	0.00	0.42	0.42	0.42	0.42
	9	3.00	0.25	0.07	0.04	0.07	0.42	0.20	0.33
	10	3.00	0.55	0.00	0.00	0.55	0.56	0.55	0.55
	11	3.00	0.14	0.04	0.03	0.03	0.25	0.09	0.17
	12	3.00	0.21	0.02	0.01	0.16	0.27	0.20	0.24
	13	3.00	0.12	0.02	0.01	0.07	0.17	0.10	0.14
	14	3.00	0.15	0.01	0.00	0.13	0.16	0.14	0.15
	15	3.00	0.20	0.00	0.00	0.19	0.21	0.20	0.20
	16	3.00	0.31	0.03	0.02	0.24	0.39	0.28	0.34
	Total	48.00	0.25	0.12	0.02	0.22	0.29	0.09	0.55
m	1	2.00	0.34	0.00	0.00	0.33	0.36	0.34	0.34
	2	3.00	0.10	0.01	0.01	0.07	0.12	0.09	0.11
	3	3.00	0.14	0.01	0.00	0.12	0.16	0.13	0.15
	4	3.00	0.33	0.08	0.05	0.12	0.55	0.28	0.43
	5	3.00	0.46	0.04	0.02	0.36	0.56	0.42	0.49
	6	3.00	0.45	0.03	0.02	0.38	0.52	0.41	0.47
	7	3.00	0.25	0.00	0.00	0.24	0.25	0.24	0.25
	8	3.00	0.33	0.02	0.01	0.27	0.39	0.31	0.36
	9	3.00	0.23	0.01	0.01	0.21	0.25	0.22	0.24
	10	3.00	0.46	0.00	0.00	0.45	0.46	0.45	0.46
	11	3.00	0.21	0.02	0.01	0.16	0.26	0.19	0.23
	12	3.00	0.11	0.01	0.01	0.07	0.14	0.09	0.12
	13	3.00	0.11	0.02	0.01	0.07	0.15	0.09	0.13

1m

1.5m

14	3.00	0.10	0.01	0.01	0.07	0.12	0.09	0.11
15	3.00	0.13	0.00	0.00	0.12	0.14	0.13	0.13
16	3.00	0.21	0.02	0.01	0.16	0.27	0.19	0.24
Total	47.00	0.25	0.13	0.02	0.21	0.28	0.09	0.49

Layers		Ν	Mean	Std.	Std.	95% Co	nfidence	Mini	Maxi
	Pit			Deviation	Error	Interval	for	mum	mum
	No.					Mean			
						Lower	Upper		
						Bound	Bound		
surface	1.00	3.00	57.78	8.86	5.11	35.77	79.78	50.55	67.66
layer									
	2.00	3.00	51.65	1.96	1.13	46.78	56.51	49.76	53.67
	3.00	3.00	41.78	2.15	1.24	36.43	47.13	39.35	43.46
	4.00	3.00	71.20	2.84	1.64	64.16	78.24	69.12	74.43
	5.00	3.00	55.26	11.92	6.88	25.65	84.88	42.47	66.06
	6.00	3.00	64.73	3.03	1.75	57.20	72.26	62.84	68.23
	7.00	3.00	46.32	14.62	8.44	10.01	82.63	29.44	54.94
	8.00	3.00	75.37	1.15	0.67	72.51	78.24	74.11	76.37
	9.00	3.00	81.91	11.10	6.41	54.34	109.49	74.22	94.64
	10.00	3.00	63.45	1.11	0.64	60.69	66.21	62.72	64.73
	11.00	3.00	32.18	9.54	5.51	8.49	55.87	23.60	42.45
	12.00	3.00	43.02	14.22	8.21	7.68	78.35	27.82	56.01
	13.00	3.00	65.99	3.06	1.77	58.38	73.60	63.29	69.32
	14.00	3.00	57.53	5.78	3.34	43.18	71.89	51.95	63.49
	15.00	3.00	71.49	2.74	1.58	64.67	78.30	68.42	73.71
	16.00	3.00	46.60	6.95	4.01	29.34	63.85	39.08	52.78
	Total	48.00	57.89	14.92	2.15	53.56	62.22	23.60	94.64
0.5m layer	1.00	3.00	44.42	22.62	13.06	-11.77	100.61	28.30	70.28
	2.00	3.00	29.77	6.94	4.01	12.53	47.00	23.64	37.30
	3.00	3.00	59.75	1.87	1.08	55.11	64.38	57.60	60.97
	4.00	3.00	57.14	7.90	4.56	37.52	76.76	48.79	64.49
	5.00	3.00	47.02	8.17	4.72	26.73	67.30	41.60	56.41
	6.00	3.00	72.66	3.06	1.77	65.06	80.26	69.51	75.62
	7.00	3.00	6.98	2.88	1.66	-0.17	14.13	3.67	8.87
	8.00	3.00	64.70	4.98	2.88	52.32	77.08	59.48	69.41
	9.00	3.00	53.03	10.00	5.77	28.18	77.87	43.31	63.29
	10.00	3.00	54.70	2.05	1.18	49.60	59.79	53.18	57.03

Organic solids (in % g VS per g wet sample/ g TS per g wet) distribution

for faecal sludge sampled from different layers within the same pit and sampled from different pits xxxiii

	11.00	3.00	44.91	14.58	8.42	8.70	81.12	28.32	55.66
	12.00	3.00	45.07	4.23	2.44	34.56	55.58	41.22	49.60
	13.00	3.00	41.50	7.93	4.58	21.79	61.20	32.86	48.46
	14.00	3.00	29.37	15.45	8.92	-9.00	67.74	12.00	41.57
	15.00	3.00	51.63	9.98	5.76	26.84	76.42	44.70	63.07
	16.00	3.00	61.16	4.84	2.80	49.13	73.19	55.94	65.51
	Total	48.00	47.74	17.50	2.53	42.66	52.82	3.67	75.62
1m layer	1.00	3.00	20.84	7.05	4.07	3.32	38.35	14.23	28.26
	2.00	3.00	32.03	4.04	2.33	21.99	42.07	28.05	36.13
	3.00	3.00	51.52	4.80	2.77	39.58	63.45	46.40	55.93
	4.00	3.00	43.77	1.24	0.71	40.70	46.84	42.54	45.01
	5.00	3.00	26.87	1.89	1.09	22.17	31.58	25.73	29.06
	6.00	3.00	20.87	7.78	4.49	1.54	40.21	13.97	29.31
	7.00	3.00	28.90	5.12	2.96	16.17	41.63	24.21	34.37
	8.00	3.00	28.55	2.59	1.50	22.10	34.99	26.74	31.52
	9.00	3.00	28.30	4.28	2.47	17.68	38.93	25.37	33.21
	10.00	3.00	68.44	5.06	2.92	55.87	81.02	63.45	73.57
	11.00	3.00	43.66	4.60	2.65	32.24	55.08	39.58	48.64
	12.00	3.00	31.30	1.80	1.04	26.83	35.78	29.92	33.34
	13.00	3.00	14.90	6.00	3.46	-0.01	29.81	8.63	20.59
	14.00	3.00	6.25	1.19	0.69	3.30	9.21	4.89	7.07
	15.00	3.00	35.44	0.64	0.37	33.84	37.03	34.83	36.11
	16.00	3.00	61.59	4.14	2.39	51.31	71.86	57.53	65.80
	Total	48.00	33.95	16.53	2.39	29.15	38.75	4.89	73.57
1.5m layer	1.00	3.00	47.03	12.36	7.14	16.33	77.73	37.81	61.07
	2.00	3.00	12.63	3.53	2.04	3.87	21.39	8.73	15.59
	3.00	3.00	18.44	2.25	1.30	12.86	24.02	15.95	20.31
	4.00	3.00	39.49	2.59	1.49	33.07	45.92	37.96	42.48
	5.00	3.00	42.92	3.77	2.18	33.55	52.28	38.57	45.28
	6.00	3.00	28.90	1.28	0.74	25.73	32.07	28.08	30.37
	7.00	3.00	6.74	2.69	1.55	0.06	13.42	3.94	9.30
	8.00	3.00	43.95	8.57	4.95	22.65	65.25	36.69	53.41
	9.00	3.00	29.99	3.31	1.91	21.77	38.22	27.42	33.73
	10.00	3.00	54.25	4.27	2.47	43.64	64.86	49.38	57.35
	11.00	3.00	42.24	7.18	4.14	24.42	60.07	35.31	49.64
	12.00	3.00	23.84	2.31	1.34	18.10	29.58	21.35	25.92

V1	\$7
- A I	v
	xi

13.00	3.00	26.59	6.20	3.58	11.20	41.98	19.47	30.77
14.00	3.00	33.04	4.97	2.87	20.70	45.37	28.42	38.29
15.00	3.00	63.57	4.45	2.57	52.51	74.62	58.90	67.76
16.00	3.00	71.50	2.64	1.52	64.95	78.06	69.39	74.46
Total	48.00	36.57	17.82	2.57	31.40	41.74	3.94	74.46

Layer	Pit	Ν	Mean	Std.	Std.	95% Co	onfidence	Mini	Maxi
	No.			Deviation	Error	Interva	l for	mum	mum
						Mean			
						Lower	Upper		
						Bound	Bound		
surface	1.00	3.00	77.31	2.27	1.31	71.67	82.94	74.70	78.82
	2.00	3.00	72.11	0.94	0.54	69.77	74.45	71.10	72.96
	3.00	3.00	73.77	1.13	0.65	70.97	76.58	72.52	74.71
	4.00	3.00	80.64	0.14	0.08	80.28	80.99	80.53	80.80
	5.00	3.00	79.61	1.05	0.61	77.01	82.22	78.47	80.53
	6.00	3.00	81.79	0.19	0.11	81.31	82.27	81.62	82.00
	7.00	3.00	75.54	6.22	3.59	60.10	90.98	68.36	79.23
	8.00	3.00	78.58	0.30	0.17	77.84	79.31	78.34	78.91
	9.00	3.00	82.22	1.53	0.89	78.40	86.03	81.18	83.98
	10.00	3.00	76.74	0.21	0.12	76.22	77.26	76.50	76.86
	11.00	3.00	68.33	8.48	4.89	47.28	89.39	60.71	77.46
	12.00	3.00	66.32	8.02	4.63	46.40	86.24	57.58	73.34
	13.00	3.00	80.19	0.68	0.39	78.49	81.88	79.47	80.83
	14.00	3.00	74.43	1.33	0.77	71.12	77.74	73.18	75.83
	15.00	3.00	84.52	1.04	0.60	81.94	87.10	83.80	85.71
	16.00	3.00	77.92	3.04	1.76	70.37	85.47	74.45	80.12
	Total	48.00	76.88	5.68	0.82	75.23	78.52	57.58	85.71
.5m	1.00	3.00	67.72	5.69	3.29	53.58	81.87	63.55	74.21
	2.00	3.00	63.07	5.70	3.29	48.91	77.23	57.78	69.11
	3.00	3.00	77.90	0.89	0.51	75.70	80.10	76.94	78.69
	4.00	3.00	78.64	2.35	1.35	72.81	84.47	76.24	80.93
	5.00	3.00	76.15	0.44	0.25	75.06	77.23	75.73	76.60
	6.00	3.00	84.95	0.96	0.56	82.56	87.34	84.35	86.06
	7.00	3.00	39.70	8.43	4.87	18.75	60.65	30.06	45.71
	8.00	3.00	78.87	0.89	0.51	76.67	81.07	78.08	79.83
	9.00	3.00	76.60	0.87	0.50	74.44	78.76	75.73	77.47
	10.00	3.00	70.20	1.70	0.98	65.97	74.42	68.31	71.61
	11.00	3.00	76.40	9.03	5.21	53.97	98.83	66.00	82.24

Moisture (in % per g wet sample) distribution for faecal sludge samp

from different layers within the same pit and sampled from

different pit.

12.00	3.00	68.56	1.32	0.76	65.28	71.83	67.60	70.06
13.00	3.00	72.37	2.94	1.70	65.07	79.66	68.98	74.23
14.00	3.00	62.93	13.92	8.04	28.34	97.52	47.05	73.04
15.00	3.00	71.24	2.05	1.19	66.14	76.34	68.98	72.99
16.00	3.00	80.83	0.80	0.46	78.83	82.83	80.08	81.68
Total	48.00	71.63	11.18	1.61	68.39	74.88	30.06	86.06
1.00	3.00	53.63	4.74	2.74	41.85	65.42	48.45	57.76
2.00	3.00	68.18	4.06	2.34	58.11	78.26	64.00	72.10
3.00	3.00	74.03	1.24	0.72	70.95	77.11	72.96	75.39
4.00	3.00	73.17	1.27	0.73	70.02	76.33	71.92	74.46
5.00	3.00	69.47	0.78	0.45	67.52	71.42	68.76	70.31
6.00	3.00	63.23	7.77	4.49	43.92	82.55	55.75	71.27
7.00	3.00	66.48	4.98	2.88	54.11	78.85	63.19	72.21
8.00	3.00	59.22	1.01	0.58	56.70	61.73	58.44	60.36
9.00	3.00	58.41	0.78	0.45	56.46	60.36	57.53	59.03
10.00	3.00	73.75	0.77	0.44	71.85	75.66	72.97	74.50
11.00	3.00	77.46	1.61	0.93	73.47	81.44	75.87	79.08
12.00	3.00	63.22	1.07	0.62	60.57	65.87	61.99	63.92
13.00	3.00	49.12	10.37	5.99	23.37	74.88	37.77	58.09
14.00	3.00	33.71	2.60	1.50	27.25	40.17	30.72	35.43
15.00	3.00	66.53	0.56	0.32	65.15	67.91	66.05	67.14
16.00	3.00	83.98	1.00	0.58	81.50	86.47	82.88	84.83
Total	48.00	64.60	12.33	1.78	61.02	68.18	30.72	84.83
1.00	3.00	75.99	2.05	1.18	70.90	81.08	73.63	77.29
2.00	3.00	42.16	6.02	3.47	27.21	57.10	35.41	46.96
3.00	3.00	60.41	2.24	1.29	54.85	65.97	57.91	62.22
4.00	3.00	69.64	2.32	1.34	63.87	75.41	67.01	71.42
5.00	3.00	87.07	0.53	0.31	85.75	88.39	86.47	87.48
6.00	3.00	71.16	0.89	0.51	68.95	73.37	70.13	71.67
7.00	3.00	42.52	7.14	4.12	24.78	60.26	34.71	48.72
8.00	3.00	74.78	3.62	2.09	65.79	83.77	71.51	78.67
9.00	3.00	64.74	1.44	0.83	61.18	68.31	63.11	65.81
10.00	3.00	69.34	0.88	0.51	67.14	71.53	68.53	70.28
11.00	3.00	74.69	3.29	1.90	66.51	82.87	72.45	78.47
12.00	3.00	59.19	1.53	0.89	55.38	63.00	57.42	60.09

1m

1.5m

13.00

3.00

62.35

5.72

3.31

48.13

76.57

55.82 66.49

xxxvi

xxxvii

14.00	3.00	63.66	3.32	1.92	55.40	71.92	60.13	66.73
15.00	3.00	72.71	1.74	1.00	68.40	77.02	70.76	74.09
16.00	3.00	85.06	0.94	0.54	82.73	87.39	84.15	86.02
Total	48.00	67.22	12.56	1.81	63.57	70.86	34.71	87.48

sampled from the four different layers (depth) within the pit of VIP latrine. (N=30).										
Sample	Mean	Std.	Std.	95% Cor	nfidence	Mini-	Maxi-			
source		Devi-	Error	Interval	for	mum	mum			
		ation		Mean						
				Lower	Upper					
				Bound	Bound					

0.02

0.01

0.01

0.01

0.00

Mean soluble COD (in mg/mg dry sample) obtained for faecal sludge

fresh faeces

pit

pit

Pit surface layer

0.5 m layer of

1m layer of pit

1.5 m layer of

0.37

0.09

0.06

0.05

0.04

0.05

0.04

0.04

0.03

0.02

Analyses of first set serum bottle test (FF= fresh faeces, an.sludge=
anaerobic sludge, COD_0 = initial COD at the beginning of test,
CH4 ₀ methane equivalence of COD ₀ at the beginning of test,
CH4 ∞ = methane produced in the course of assay).

Sample	Bottle	Bottle	COD ₀	CH4 ₀	CH4	CH4(mL)	Nett	CH4∞	Ultimate	Biod
identity/source	Temp.		in test	(L)	(mL)	produced	CH4	(L) At	Biodeg -	grada
	°C	Temp.	Bottle		produced	from	(mL)	STP	radability	(%)
		°K			from	bottles	for			
					sample	Blanks	sample			
					bottles		bottles			
FF + an. sludge	38.00	311.15	0.85	0.30	195.10	12.34	182.76	0.21	0.70	69.64
FF + an. sludge	38.00	311.15	0.87	0.30	260.87	12.34	248.53	0.28	0.93	92.89
FF + an. sludge	38.00	311.15	0.89	0.31	162.56	12.34	150.22	0.17	0.55	55.01
VIP mid layer	38.00	311.15	0.85	0.30	33.79	12.34	21.45	0.02	0.08	8.20
+ an. sludge										

0.42

0.10

0.08

0.06

0.05

0.32

0.02

0.00

0.00

0.00

0.43

0.18

0.12

0.09

0.07

0.32

0.07

0.05

0.03

0.03

Analyses of second set (pit 9) serum bottle test (FF= fresh faeces, an.sludge= anaerobic sludge, COD_0 = initial COD at the beginning of test, CH4₀ methane equivalence of COD_0 at the beginning of test, CH4 ∞ = methane produced in the course of assay).

Bottle	Sample	COD ₀	CH4 ₀	CH4	CH4	Nett	(Nett)	Ultimate	
ID no	Identity	in test	(L)	(mL)	produced	CH4	CH4∞	Biodegr-	Biodegr-
	/source	Bottle		Produced	from	(mL)	(L)	adability	adability
				from	Blanks	for	At		(%)
				sample		sample	STP		
				bottles		bottles			
ff2+ an.	fresh faeces	0.28	0.10	55.40	1.15	54.26	0.05	0.56	55.60
sludge									
ff3+ an.	fresh faeces	0.28	0.10	83.73	1.15	82.58	0.08	0.83	83.10
sludge									
surf1+	VIP surface	0.27	0.09	15.18	1.15	14.03	0.01	0.15	15.03
an.	layer								
sludge									
surf2+	VIP surface	0.27	0.09	10.49	1.15	9.34	0.01	0.10	9.95
an.	layer								
sludge									
surf3+	VIP surface	0.27	0.10	15.37	1.15	14.23	0.01	0.15	14.89
an.	layer								
sludge									
0.5m1+	VIP	0.29	0.10	24.18	1.15	23.03	0.02	0.23	22.50
an.	0.5m depth								
sludge									
0.5m2+	VIP 0.5m	0.30	0.10	13.30	1.15	12.15	0.01	0.12	11.66
an.	depth								
sludge									
0.5m3+	VIP 0.5m	0.29	0.10	11.13	1.15	9.98	0.01	0.10	9.71
an.	depth								
sludge									
1m1+	VIP 1m	0.32	0.11	17.94	1.15	16.80	0.02	0.15	14.79
an.	depth								
sludge									

1m2+	VIP 1m	0.33	0.11	14.01	1.15	12.87	0.01	0.11	11.27	
an.	depth									
sludge										
1m3+	VIP 1m	0.33	0.11	17.60	1.15	16.45	0.02	0.14	14.45	
an.	depth									
sludge										
1.5m1+	VIP 1.5m	0.28	0.10	10.80	1.15	9.65	0.01	0.10	9.81	
an.	depth									
sludge										
1.5m2+	VIP 1.5m	0.28	0.10	8.75	1.15	7.60	0.01	0.08	7.70	
an.	depth									
sludge										

gis2.durban.gov.za/website/atest/Run.htm)

