

DETERMINATION OF THE METHANOGENIC POTENTIAL OF AN APPLE PROCESSING WASTEWATER TREATMENT SYSTEM

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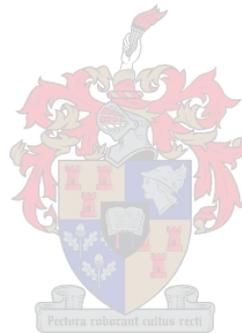
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DECLARATION

I, the undersigned, hereby declare that the work contained in this thesis is my own original work and has not previously in its entirety or in part been submitted at any university for a degree.

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ABSTRACT

The food and beverage industry generates large volumes of wastewater annually. The disposal of factory effluent from the fruit processing industry has always been a cause of concern to both the fruit processors and controlling bodies responsible for effluent management. Traditional disposal of wastewater into sewerage works has become undesirable due to its economical and environmental impacts. Therefore, on-site anaerobic treatment of wastewater has received considerable interest due to lower capital outlays and energy recovery possibilities. Thus, the aim of this study was to establish an operational treatment profile for an anaerobic pond system treating fruit-processing wastewater. The specific activity of the microbial populations was also monitored to determine the effect of the fruit processing seasons (peak and off-peak season). The biogas production potential at various temperatures was also assessed to determine the viability of methane recovery.

The influence of the processing and environmental conditions on the ponds' performance was established by monitoring various process parameters. The results showed that the chemical oxygen demand (COD) levels decreased during the off-peak season but the pond pH remained relatively stable between 6.0 and 6.4 during the entire year. Pond alkalinity was found to be dependent on the regular lime dosing to maintain the necessary alkalinity. The volatile fatty acid (VFA) concentrations indicated that the microbial populations of the pond were functioning well. However, a decrease in microbial activity and VFA concentrations were observed at the lower temperatures during the winter months. The temperature profile of the pond showed that the pond temperature was impacted by the fluctuations in the ambient air temperature. The general trend established by the operational treatment profile clearly showed the impact of the peak and off-peak season.

The sludge activity of the anaerobic pond was evaluated to determine the effect of the apple-processing peak and off-peak season on the specific activity of the acidogenic and methanogenic populations within the sludge. An activity test using four different test media was used during the activity assays. Sludge samples were taken at four different sampling positions across the pond's sludge bed. The sludge was also subjected to a biogas formation study, which was designed to simulate pond conditions on laboratory

scale in order to evaluate the biogas production potential of the anaerobic pond. The cumulative biogas volume and total CH₄ composition showed little or no difference between the four sludge sampling sites. A major difference was found between the activity of the microbial populations during the peak and off-peak seasons. The overall trend regarding the biogas production rate (S_B) and the methane production rate (S_M) values showed an increased activity during peak-season and a decreased activity during off-peak season. For the biogas formation test the highest incubation temperature (25 °C) resulted in the most biogas being produced, followed by 18 °C, and with 10 °C resulting in the lowest biogas volume. The biogas formation tests indicated that microbial activity and therefore biogas production was dependent on especially favourable temperature conditions. The pond and activity of the microbial populations are therefore influenced by factors like environmental changes such as decreased air temperatures and substrate changes such as decreased COD concentrations during the off-peak season. This in turn influences the rate of biogas production as well as the methane production rate.

The theoretical CH₄ calculations and estimates based on the results obtained during the biogas formation tests indicated that CH₄ recovery from the anaerobic pond would definitely be a worthwhile consideration. If it were assumed that the estimated CH₄ volumes (based on only 15% of the pond volume for practical reasons) obtained could be applied as an energy source, the minimum yearly savings in coal usage would amount to about R 665 000.

This study was valuable in evaluating the factors such as pond conditions, pond activity and air temperatures and the effect on the biogas production potential as well as more importantly, CH₄ production for the purpose of energy recovery.

UITTREKSEL

Die voedsel en drank industrie produseer jaarliks groot volumes uitvloeisel. Die behandeling en/of verwydering van uitvloeisel afkomstig van die vrugte verwerkingsindustrie is 'n bron van kommer vir die vrugteverwerker asook beheerliggame wat verantwoordelik is vir die bestuur van besoedelde uitvloeisel. Tradisionele verwydering van uitvloeisel deur munisipale suiweringswerke is deesdae ongewens as gevolg van die ekonomiese en omgewingsnadele wat dit inhou. Dit het veroorsaak dat behandelingsmetodes soos anaerobiese behandeling, wat op die verwerkingsperseel onderneem kan word, baie gewild raak. Hierdie behandelingsmetode het baie voordele in terme van laer kapitaal belegging en energie herwinningsmoontlikhede. Dus, die doel van die studie was om 'n bedryfsprofiel op te stel van 'n anaerobiese dam sisteem wat uitvloeisel behandel vanaf 'n vrugte verwerkingsfabriek. Die spesifieke aktiwiteit van die mikrobiële populasies is ook gemonitor om die effek van die vrugte seisoene (in en af-seisoen) te bepaal. Die biogas produksie potensiaal by verskeie temperature is ook ondersoek om die lewensvatbaarheid van metaan herwinning te bepaal.

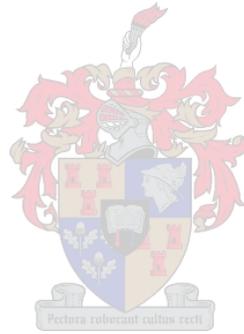
Die invloed van die verwerkingseisoene en omgewingsfaktore is bepaal deur verskeie bedryfsparameters te monitor. Die resultate het getoon dat die chemiese suurstof vereiste (COD) gedaal het tydens die af-seisoen, maar dat die pH van die dam relatief stabiel gebly het tussen 6.0 en 6.4 deur die verloop van die jaar. Die dam alkaliniteit was afhanklik van gereelde kalk-dosering om die noodsaaklike alkaliniteitsvlakke te handhaaf. Die kort ketting vetsuur (VFA) konsentrasie het getoon dat die mikrobiële populasies goed funksioneer in die dam. Alhoewel daar 'n afname in die mikrobiële aktiwiteit was sowel as die VFA konsentrasie tydens die wintermaande. Die temperatuur profiel het getoon dat die damtemperatuur beïnvloed word deur lugtemperatuur fluktuasies. Die algehele bedryfsprofiel het duidelik die impak van die prosesseringseisoene getoon.

Die slykaktiwiteit van die anaerobiese dam was ge-evalueer om die impak van verwerkingseisoene op die spesifieke aktiwiteit van die asidogene en metanogene populasies te bepaal. 'n Aktiwiteitstoets is gebruik met vier verskillende toets substrate. Die slykmonsters was geneem by vier verskillende posisies in die dam. Die slykmonsters is ook gebruik in 'n biogas formasie studie, wat die damtoestande gesimuleer in op klein

skaal. Hierdie studie was uitgevoer om die biogas produksie potensiaal van die dam vas te stel. Die kumulatiewe biogas volume en totale metaan inhoud van die vier slykmonsters het slegs klein verskille getoon. Daar was hoewel 'n groot verskil tussen die aktiwiteit van die mikrobiële populasies tydens die in- en af-seisoen. Die algehele tendens in die aktiwiteit het getoon op 'n afname in die S_B en S_M waardes tydens die af-seisoen en 'n toename tydens die in-seisoen. Die biogas formasie toetse het aangedui dat die hoogste inkubasiestemperatuur (25°C) die grootste hoeveelheid biogas oplewer, terwyl die laagste inkubasiestemperatuur die kleinste hoeveelheid oplewer het. Hierdie biogas formasie toetse het getoon dat die mikrobiële aktiwiteit en dus die biogas produksie potensiaal afhanklik was van gunstige temperatuur toestande. Dus word die dam en die aktiwiteit van die mikrobiële populasies grootliks beïnvloed deur omgewingsfaktore soos verlaagde lugtemperatuur asook veranderinge in substraat konsentrasies, veral verlaagde COD konsentrasies tydens die af-seisoene. Gevolglik word die biogas en metaangas produksie-tempo's beïnvloed.

Die teoretiese CH_4 berekening wat gebaseer was op 15% van die damvolume asook resultate verkry tydens die biogas formasie toetse het getoon dat CH_4 herwinning vanuit die anaerobiese dam 'n moeite werd is. Die berekening het getoon dat daar 'n jaarlikse minimum besparing van ongeveer R665 000 kan wees indien die herwinde CH_4 tesame met steenkool gebruik word.

Hierdie studie het waardevolle inligting verskaf ten opsigte van die damtoestande, mikrobiële aktiwiteit en lugtemperatuur wat gedurende die jaar voorkom bestaan. Inligting omtrent die biogas produksie potensiaal en CH_4 produksie is verkry wat belangrik is vir die herwinning van energie.



**Dedicated to my parents,
Mr. O.P.H. Paulsen & Mrs. C.G. Paulsen**

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Language and style in this thesis are in accordance with requirement of the *International Journal of Food Science and Technology*. This thesis represents a compilation of manuscripts where each chapter is an individual entity and some redundancy between chapters has, therefore, been unavoidable.

CHAPTER 1

INTRODUCTION

Large volumes of wastewater are generated by the food processing industry annually and consequently this results in severe environmental impacts (Trnovec & Britz, 1998). The fruit and vegetable canning industry as well as other agricultural industries are thus faced with two major problems (Sigge, 2005). Firstly, a profitable level of production needs to be maintained while reducing the intake of fresh, potable water, and secondly, the large volumes of generated wastewater need to be disposed or treated in an environmentally-friendly manner.

It has been reported that the South African apple juice processing industry is responsible for the production of 6.8 million m³ of wastewater annually (Van Schalkwyk, 2004). The peak-processing season is usually during the months of February to June when the apples are harvested and processed. Consequently, this is the period when most of the wastewater is generated. Towards the end of the peak-season (May to July) apples of a riper and softer nature are processed. The quality of these ripe apples is further influenced during the transport, washing and drying phases and the severely disintegrated apples end up in the wastewater (Van Schalkwyk, 2004). The use of overripe apples for juice extraction is, however, very popular since these apples cannot be sold on the fresh market but they do produce a high juice yield. Larger quantities of softer apples are thus processed during the latter periods of the processing season which subsequently leads to the production of larger volumes of wastewater that contain higher organic loads (Van Schalkwyk, 2004). These high wastewater volumes and organic loads put considerable stress on water treatment systems (Van Schalkwyk, 2004).

Various wastewater treatment options exist for the treatment of fruit and vegetable wastes. Anaerobic digestion (AD) is considered to be one of the major biological treatment processes in use today (Wang *et al.*, 1999). The AD process has several advantages over conventional disposal methods. Large areas of land are not required (Wood, 1992); bulking of the biomass within the digester is appreciably less than with other treatment methods (Lin & Yang, 1991); bad odours do not occur; the chemical oxygen demand (COD) is substantially lowered; nutrient requirements are low; and methane and carbon

dioxide are obtained as final metabolic end-products (Lettinga, 2001). The methane represents a form of combustible energy and can be used in the factory as boiler fuel, for incineration and or even to power forklifts, therefore facilitating considerable savings in energy (Lettinga, 2001).

Treatment of these fruit type effluents by upflow anaerobic sludge blanket (UASB) reactors has been shown to be a feasible option. Chemical oxygen demand reductions of up to 93% at organic loading rates (OLR) of $10.95 \text{ kgCOD}\cdot\text{m}^{-3}\cdot\text{d}^{-1}$ and hydraulic retention times (HRT) of $<12 \text{ h}$ may be achieved while treating fruit canning wastewaters (Trnovec & Britz, 1998). Therefore, the application of the UASB design is not only applicable as a wastewater treatment system but additionally can serve as an energy recovery system. This form of biogas technology offers an attractive method of meeting partial energy needs (Yadvika *et al.*, 2004; McGrath & Mason, 2004).

It is only recently that the South African food and beverage industries have come to realise the importance of long-term incentives for the development of sustainable energy forms, such as bio-energy from anaerobic digestion of the organic rich effluents. One such apple processing company has realised the importance of waste minimisation and energy recovery and is situated in the Overberg area, South Africa. This plant processes about 70 000 tons of apples and 9 000 tons of pears annually, producing concentrates and a variety of carbonated fruit juices for the local and international markets. The production process generates large volumes of wastewater that are treated on-site via a pond and wetland system (E. Muller, Appletiser SA, South Africa, personal communication, 2004). The treated wastewater is then irrigated onto the surrounding orchards. The current wastewater treatment process is considered to be efficient regarding pollutant removal. However, the possibility to produce renewable energy from the wastewater treatment process has received interest due to the economical and environmental benefits. Therefore, an investigation was set in motion to determine the feasibility of biogas recovery from an anaerobic pond which formed part of treating the apple factory wastewater.

The objectives of this study were firstly, to monitor the wastewater treatment system, specifically the anaerobic pond system (Pond A) during the peak and off-seasons in order to determine and evaluate the treatment efficiencies and pinpoint any environmental limitations, and then to characterise the operational profile for the anaerobic pond. The second objective was to determine the effect of the fruit processing seasons (peak and off-

season) on the specific activity of the acidogenic and methanogenic populations in the sludge of the anaerobic pond system. The biogas production potential at various temperatures will also be assessed to determine the viability of CH₄ recovery.

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CHAPTER 2

LITERATURE REVIEW

A. BACKGROUND

For many years the South African food and beverage industries have operated solely to be profitable and economically sustainable with very little concern for water usage, disposal of wastewater or even waste minimisation. This mindset has led to environmental impacts such as increased salination and eutrofication of water resources (Van Schoor, 2000). South Africa being a water scarce country with extended droughts and an unevenly distributed rainfall, has experienced a dramatic decline in annual rainfall patterns resulting in greater water shortages and the consequential implementation of water restrictions in certain regions of the country (Pybus, 1999). This occurrence has forced many industries to reduce water usage, implement wastewater treatment systems and introduce waste management or minimisation techniques.

The fruit processing industry is responsible for the generation of large quantities of liquid and solid wastes. Thus far, current disposal practices have included treatment of wastewater in lagoons or ponds before irrigation onto land, evaporation in basins, direct irrigation and treatment through wetland systems. Some companies dispose of their effluent into municipal sewers, but current environmental legislation (Van Schoor, 2000) and the associated escalating cost of water supply and discharge has shifted the emphasis to on-site wastewater recycling and or pre-treatment before discharging into municipal sewers.

Various wastewater treatment options exist for the treatment of fruit and vegetable wastes, and include both aerobic and anaerobic digestion processes (Chynoweth *et al.*, 2001). Anaerobic digestion has been successfully implemented as an on-site secondary treatment method for fruit and vegetable wastes and offers several additional advantages over conventional methods (Azad, 1976; Drysdale, 1981; Sterrit & Lester, 1982). The most significant advantage of the anaerobic digestion process is the production of a by-product, biogas, which can be a valuable energy source.

Biogas production from the biological conversion of organic waste has received increasing attention over the past few years mainly because of the ever-present risk of fossil fuel depletion (Shiralipour & Smith, 1984). The fossil fuel crisis and consequent price rises as well as the environmental impacts of fossil fuel usage have initiated the exploration of renewable energy sources (Chynoweth *et al.*, 2001). Methane (CH₄), an energetic constituent of biogas, is generated during anaerobic wastewater treatment as a by-product and can be used as fuel for boilers or reactor heating and for electricity generation (Mendonca & Campos, 2001). Therefore, anaerobic digestion can be considered as both a wastewater treatment process and a renewable energy production technology (Membrez & Fruteau de Laclos, 2001). Bioenergy has been cited as the most significant renewable energy source that can be used in the next few decades, at least until solar and wind power generation may offer a more economically viable large-scale alternative (Gunaseelan, 1997).

The use of renewable energy from organic waste as a resource is not only “greener” with respect to most pollutants but offers an effective method for the treatment and disposal of large quantities of municipal, industrial and agricultural organic wastes. Recovering energy during such wastewater treatments might reduce the cost and somewhat reduce our dependence on fossil fuels (Angenet *et al.*, 2004). The overall prospects of renewable energy may also offer the possibility of replacing fossil fuel derived energy and reduce environmental impacts such as global warming and acid rain (Chynoweth *et al.*, 2001).

Bio-energy and its exploitation from landfills and anaerobic digesters have been well demonstrated internationally and, to a lesser extent, in South Africa. Up to the end of 1991, the exploitation of CH₄ from landfills was carried out at only two sites in South Africa, namely Grahamstown and Robinson Deep (Letcher, 1992). Over the last two decades anaerobic digestion processes have been implemented in many South African food and beverage industries as a wastewater treatment process rather than an energy recovery process.

The South African food and beverage industry has recently realised that waste minimisation is the key to sustainable economic and environmental growth and development. Therefore, more and more companies are looking towards technologies offering both environmental and energy recovery advantages which comply with ISO 14 000 standards.

B. ORGANIC WASTEWATER TREATMENT METHODS

Almost all wastewaters can be treated biologically, but the wastewater must be properly analysed and the treatment process be environmentally controlled (Tchobanoglous & Burton, 1991). There are five major biological wastewater treatment processes, each varying in complexity and efficiency. The five include aerobic, anoxic, anaerobic, combined aerobic, anoxic and anaerobic, and pond processes. All these are based on degradation processes occurring naturally in nature (Tchobanoglous & Burton, 1991; Bitton, 1999).

Aerobic Processes

This biological treatment process can be subdivided into suspended-growth, attached-growth, and combined suspended and attached-growth processes. Suspended-growth processes include methods like activated sludge, aerated lagoons and aerobic digestion to name a few. Attached-growth processes refer to methods like trickling filters and rotating biological contactors (RBC) (Tchobanoglous & Burton, 1991; Bitton, 1999).

Activated Sludge Process - The activated sludge process is mainly used as a secondary biological treatment method for various wastewaters (Bitton, 1999). This process involves the production of an activated mass of microorganisms capable of stabilising wastewater aerobically and hence its name. It involves two characteristic features namely aeration and settling (Droste, 1997; Bitton, 1999).

Aerobic oxidation of the organic waste is carried out in an aeration tank. This tank contains the mixed liquor (ML), which consists of primary effluent that is mixed with return activated sludge (RAS). Aeration is provided by diffused or mechanical means and can be provided on a continuous or semi-continuous basis. It is during this time that the organic matter is oxidised to carbon dioxide (CO_2), water (H_2O), ammonia (NH_4) and new cell biomass. Aeration serves two purposes: the first is supplying oxygen to the aerobic microorganisms and, secondly to ensure that the activated sludge flocs are in constant agitation to provide adequate contact between the flocs and incoming wastewater (Tchobanoglous & Burton, 1991; Bitton, 1999). Aeration is also the major energy consuming operation of the activated sludge process. The aerobic conditions also allow energy recovery in terms of biomass per unit substrate

metabolised. This results in a large quantity of excess sludge production that requires processing and disposal (Munters, 1984) and directly leads to a major operating expense and is seen as a major disadvantage of this process (Droste, 1997; Bitton, 1999; Garrido *et al.*, 2001).

The microbial cells, produced during the oxidation phase form flocs that contain large numbers of bacteria held together by secreted polymers, which accumulate on their capsules. These flocs are then allowed to settle in a clarifier. The mixed liquor is transferred from the aeration tank into the sedimentation tank where the sludge is separated from the treated effluent (Bitton, 1999). A portion of the settled sludge is recycled back to the aeration tank while the remainder is disposed of or used in further treatments such as aerobic or anaerobic digestion. Secondary clarification follows floc sedimentation. Secondary clarification is the attachment of dispersed bacterial cells and small flocs to the settled flocs. In contrast, flocculation is a response of microorganisms to low nutrient conditions in their environment and flocculation serves as a survival mechanism due to a more efficient utilisation of food in the close proximity of cells. The sedimentation tanks of the activated sludge process serve as a selector for microorganisms that have suitable settleability characteristics. For instance, organisms growing in larger floc particles settle well and are returned to the aeration tank where they are able to proliferate while smaller floc particles that are not capable of settling are not separated from the effluent and are washed out of the process (Bitton, 1999). This is an important factor as problems can arise from insufficient sludge separation from the treated effluent and this leads to a loss of solids (Tchobanoglous & Burton, 1991; Bitton, 1990; Contreras *et al.*, 2000).

Several modifications of the conventional activated sludge process exist and include: an extended aeration system; a complete mixed aerated system; a high rate system; a pure oxygen sequencing batch reactor (SBR); contact stabilisation; oxidation ditches; deep tanks and deep shaft processes (Tchobanoglous & Burton, 1991; Droste, 1997; Bitton, 1999).

Pond Treatment Processes

Waste stabilisation pond technology is a flexible treatment process, which is usually simple in design and offers economical construction and low cost operation and maintenance options (Pearson, 1996; Bitton, 1999). Modern research has allowed waste stabilisation pond technology to be expanded in terms of its application and

reliability. Pond technology may be used in the treatment of domestic sewage as well as a wide range of industrial wastewaters, and may be designed to meet exacting effluent standards (Vanderholm, 1984; Springer & Goissis, 1988; Hickey *et al.*, 1989; (Tchobanoglous & Burton, 1991; McGrath & Mason, 2004). The existence of a wide range of pond types ensures that process optimisation is established to meet particular applications or conditions (Pearson, 1996; Bitton, 1999). Ponds can be used to combine wastewater treatment and storage and are considered to be the most inexpensive option for treating wastewater for subsequent re-use in aquaculture and agriculture (Hammer, 1986; Bitton, 1999). Various pond designs can offer other advantages such as the production of large quantities of algal biomass for animal and potential human consumption as well as energy production via methane. Ponds are thus regarded as a suitable method for wastewater and resource recovery technology compatible with current environmental thinking (Tchobanoglous & Burton, 1991; Pearson, 1996; Bitton, 1999).

Pond treatment processes can be classified into four groups with respect to the presence of oxygen. These pond systems are aerobic, maturation, facultative and anaerobic stabilisation ponds (Tchobanoglous & Burton, 1991; Bitton, 1999).

Aerobic stabilisation ponds – These ponds are described as large, shallow basins that are suitable for the treatment of wastewater by natural processes that involve the participation of both algae and bacteria (Bitton, 1999). The bacteria and algae are suspended in the pond whilst aerobic conditions exist throughout the depth of the pond. Furthermore, two types of basic aerobic ponds exist, both differing in objective. The operation for the first type is to maximise algae production and the design is usually limited to a depth of 150 to 450 mm. The second type is used to maximise the amount of oxygen produced and pond depths of up to 1.5 m are used. In both types oxygen or aerobic conditions are established through oxygen production via the algae and through the atmospheric diffusion of oxygen into the liquid (Bitton, 1999). Factors such as organic loading rate (OLR), degree of pond mixing, pH, nutrients, sunlight, and temperature influences the particular algal and bacterial species that are present in any section of an aerobic stabilisation pond (Tchobanoglous & Burton, 1991). In general aerobic ponds exert a high BOD₅ conversion efficiency ranging up to 95%. The soluble BOD₅ in an aerobic pond system will be removed from the influent wastewater but the pond effluent will contain an equivalent or higher concentration of algae and bacteria

and therefore ultimately have a higher BOD₅ than the original waste stream (Tchobanoglous & Burton, 1991; Pearson, 1996; Bitton, 1999).

Facultative ponds - These ponds are used for the stabilisation of wastewater by a combination of aerobic, anaerobic and facultative bacteria. A facultative pond is comprised of three zones (Tchobanoglous & Burton, 1991; Bitton, 1999). A surface zone is formed by a symbiotic relationship between aerobic bacteria and algae. This symbiotic relationship is responsible for the production of oxygen required by the facultative bacteria. The intermediate zone is partly aerobic and partly anaerobic and contains facultative bacteria responsible for the decomposition or oxidation of the soluble and colloidal organic materials. The carbon dioxide produced during this organic oxidation serves as a carbon source for the algae. Lastly, an anaerobic bottom zone exists where anaerobic bacteria decompose the accumulated solids that settle out to form the anaerobic sludge layer. This anaerobic breakdown results in the production of dissolved gases such as CO₂, H₂S and CH₄, which are subsequently oxidised by the aerobic bacteria or vented to the atmosphere (Tchobanoglous & Schroeder, 1985). BOD removals range from 70 – 95%. Higher removal rates are obtained with longer retention times. Facultative stabilisation ponds are conventionally used for the treatment of screened or primary wastewater. In some cases surface aerators are used to maintain the oxygen rich surface layer. In these cases the use of algae is eliminated and as a result higher organic loads may be applied (Tchobanoglous & Burton, 1991; Bitton, 1999).

Facultative ponds have been used for the treatment of food processing and canning wastes (Azad, 1976; Parker, 1978). For example it has been reported that a laboratory-scale facultative pond treating yoghurt waste removed 70 and 80% of the total and soluble COD, respectively, at retention times of 7.9 d and OLR's as high as 450 kg COD.ha⁻¹.d⁻¹ (Kilani, 1992). Another laboratory-scale facultative pond system treating "synthetic" milk wastewater at loading rates of 300 kg BOD₅.ha⁻¹.d⁻¹, achieved 90% COD removal (Al-Khateeb & Tebbutt, 1992). An aerated/facultative lagoon successfully treated corn wet-milling wastewater and achieved a 95% BOD removal while the system was receiving 3 450 kg COD.d⁻¹ (Muirhead, 1990). Therefore, key applications considered for facultative stabilisation ponds are organic solids removal and pathogen removal (Mara *et al.*, 1996; Pearson, 1996; Pescod, 1996).

Anaerobic stabilisation ponds - These ponds serve as sedimentation basins as well as anaerobic treatment devices for high-strength organic wastewaters that contain high solid concentrations (Bitton, 1999). Anaerobic ponds are usually deep earthen ponds with appropriate inlet and outlet piping. Anaerobic ponds can be as deep as 9.0 m to ensure that the anaerobic conditions are maintained throughout the pond and to conserve heat energy (Hammer, 1986; Droste, 1997; Bitton, 1999). In an anaerobic pond system organic waste is converted to CO₂, CH₄, and other gaseous end-products, organic acids and cell biomass. Conversion efficiencies can range from 75 to 85% when operating at optimal conditions (Drysdale, 1981; Pearson, 1996; Pescod 1996; Bitton, 1999). Anaerobic ponds have considerable advantages over facultative ponds in terms of land use and have a low surface area to volume ratio. Anaerobic ponds can sustain much higher loads than facultative ponds. Land requirements have also been greatly reduced by recent design innovations. Modern anaerobic ponds operate with minimum hydraulic retention times of one day and their inclusion in a pond system can give a land area saving of over 75% at design temperatures above 16°C (Pearson, 1996). There is no requirement with regards to expensive mechanical aeration and only small amounts of sludge are generated (Hammer, 1986; Bitton, 1999). Anaerobic stabilisation pond systems usually are operated in conjunction with other treatment processes for follow-up treatment of the partially clarified effluent (Pearson, 1996; Pescod, 1996; Bitton, 1999). The drawbacks associated with anaerobic stabilisation ponds are the production of odorous compounds (e.g. H₂S), sensitivity to toxicants, and the requirement of relatively high temperatures. Anaerobic digestion of wastewater is virtually halted below 10°C.

Anaerobic lagoons have been employed to treat wastewaters arising from sugarbeet processing, fruit and vegetable canning, milk processing (Azad, 1976; Drysdale, 1981; Sterrit & Lester, 1982), coffee production (Gathuo *et al.*, 1991), slaughterhouse workings, meat and poultry processing and sugar processing wastewater (Droste, 1997). BOD removals that have been achieved, have ranged from 20 - 95% at BOD loading rates of up to 3 360 kg.ha⁻¹.d⁻¹ were recorded (Azad, 1976; Droste, 1997).

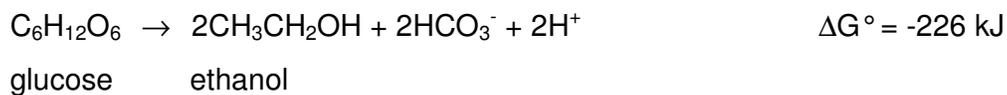
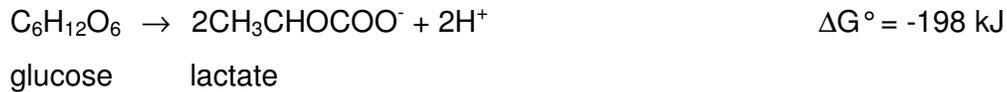
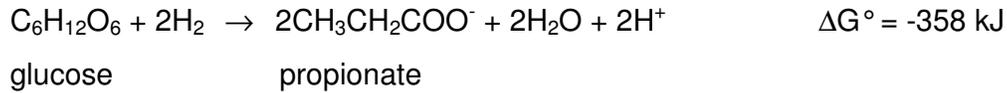
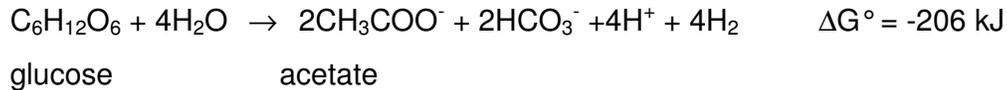
Pearson (1996) also emphasised the role that waste stabilisation pond technology plays in energy recovery. He reported that CH₄ recovery from facultative pre-treatment ponds was possible using submerged gas collectors. The CH₄ recovered from pond systems is of a good quality and not only represents energy recovery but

also prevents the release of CH₄ into the environment. Conventional pond systems are seen as economical because of lower costs in terms of construction, operation and maintenance. The microbiological quality of the effluent treated using pond systems is such that it is suitable for the use in unrestricted irrigation. This is allowed through a natural disinfection process, without the addition of chemicals or the use of physical disinfection technology. Any other form of wastewater treatment technology cannot match this important characteristic. The water, nutrients and algae in pond effluents all represent energy recovery when used directly for crop irrigation (Pearson, 1996).

Anaerobic digestion

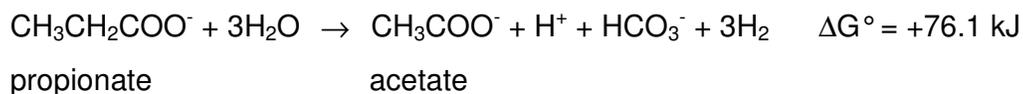
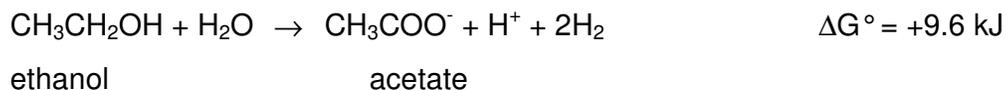
Anaerobic digestion (AD) is the decomposition of organic material and the subsequent formation of CH₄ and CO₂ by microorganisms in the absence of oxygen (Bryant & McInerney, 1981; Chynoweth *et al.*, 2001; Mata-Alvarez, 2003). Anaerobic digestion relies on the presence and activity of specific bacterial species that form a community, each with specialized ecological roles and complex nutritional requirements (Bryant & McInerney, 1981; Iannotti *et al.*, 1987; Barnett *et al.*, 1994). Four different bacterial groups as shown in Fig. 2.1 with their synergistic relationship are known to be responsible for the AD process (Bryant & McInerney, 1981; Blaut, 1994; Mata-Alvarez, 2003).

Hydrolytic and fermentation bacteria (Acidogens) - The acidogens catabolize carbohydrates, proteins, lipids and other minor components of complex organic molecules to soluble monomer molecules such as amino acids, glucose, glycerol, short chain fatty acids, hydrogen and carbon dioxide by a process of hydrolysis (Gander *et al.*, 1993; Mata-Alvarez, 2003). Hydrolysis of the complex molecules is governed by extracellular enzymes, especially the hydrolases. Depending on the type of reaction catalysed, the extracellular enzymes can also include cellulase, protease and lipases (Gander *et al.*, 1993). The soluble monomers are readily available to microbial cells and can be metabolised (Forday & Greenfield, 1983; Bitton, 1999). The principal volatile fatty acids that are formed include acetic, propionic and butyric acids, however, small quantities of valeric acid may also be present (Chen *et al.*, 1980; Duarte & Anderson, 1982; Forday & Greenfield, 1983; Batstone *et al.*, 2002; Mata-Alvarez, 2003). The main reactions take place as follows:



During this phase acidification occurs but the chemical oxygen demand (COD) reduction is minimal. However, some COD reduction may occur when large amounts of H_2 and CO_2 are produced but this COD reduction is seldom higher than 10% (Noike *et al.*, 1985). Hydrolysis can be considered as the rate-limiting step in the overall anaerobic digestion process because it is a relatively slow step (Noike *et al.*, 1985). This can be especially true for wastes such as raw cellulosic wastes containing lignin (Bitton, 1999). The efficiency of the hydrolysis step also directly contributes to the ultimate CH_4 yield.

Acetogenic Bacteria - The hydrogen-producing acetogenic bacteria catabolize certain fatty acids and neutral end-products to acetate, formate, CO_2 and H_2 . Other fatty acids and metabolites such as propionate, butyrate, lactate, succinate and alcohol may also be produced from odd-numbered carbon skeletons (Chen *et al.*, 1980; Zinder, 1990). Ethanol, propionate and butyrate are converted to acetate according to the following reactions (Forday & Greenfield, 1983; Bitton, 1999; Batstone *et al.*, 2002; Mata-Alvarez, 2003):



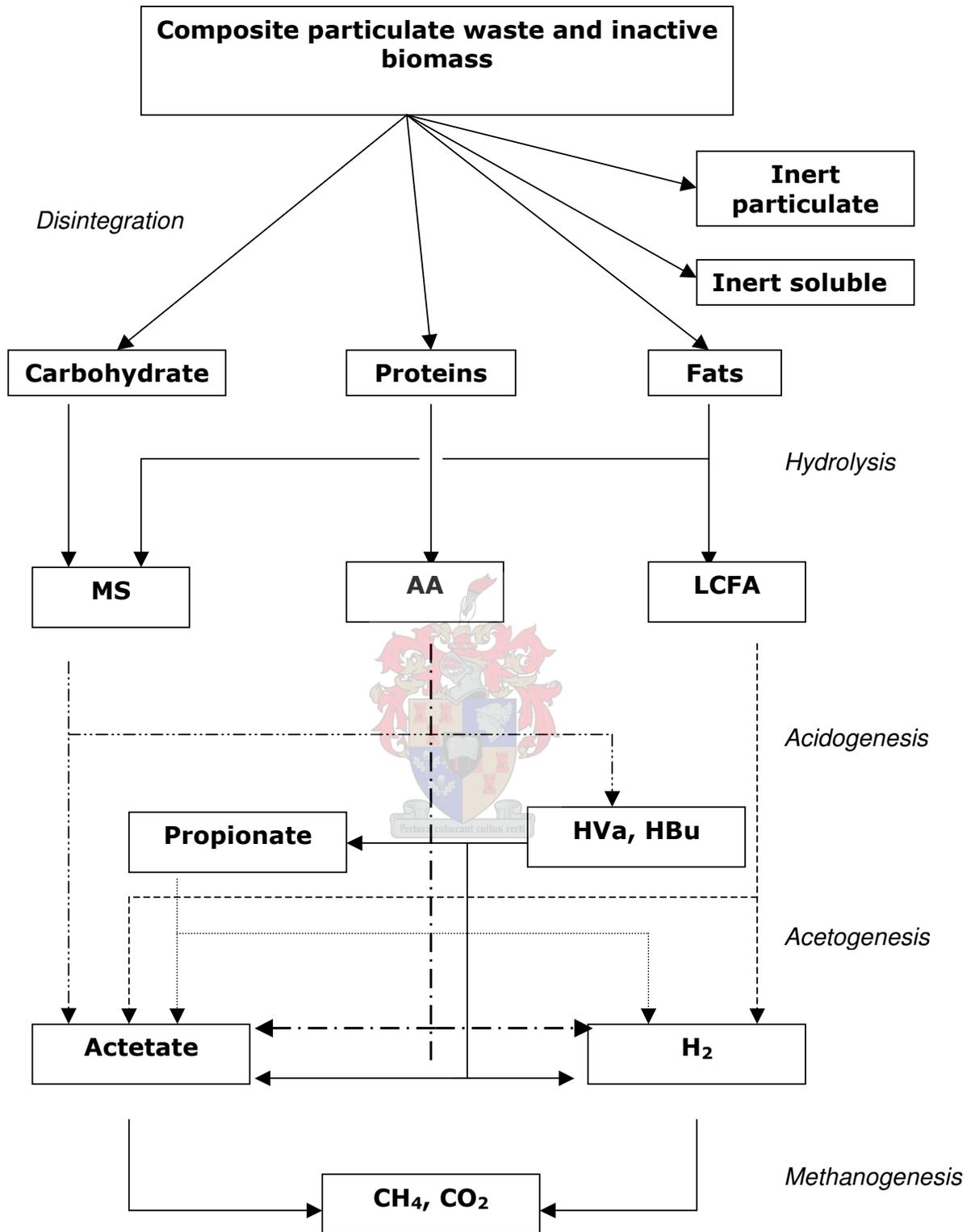


Figure 2.1 Outline of the conversion processes in anaerobic digestion according to the Anaerobic Digestion Model No. 1 (ADM1) (Batstone *et al.*, 2002). Abbreviations include MS (monosaccharides); AA (amino acids); LCFA (long chain fatty acids); HVa (valeric acid); HBu (butyric acid).

two main sub-categories exist for the methanogens. The first is the hydrogen utilising group or hydrogenotrophic group that splits acetate and convert H_2 and CO_2 to CH_4 as follows (Blaut, 1994):



This methanogenic group is also responsible for maintaining the very low H_2 partial pressure, which is necessary for the conversion of volatile fatty acids and alcohols to acetate (Iannotti *et al.*, 1987; Bitton, 1999).

The second group, the acetotrophic methanogens or acetoclastic methanogenic methanogens, are responsible for the conversion of acetate to CH_4 and CO_2 as follows (Zinder, 1993; Bitton, 1999):



This group grows much slower than the acetoclastic methanogens, with doubling times of 2 –12 days. They are responsible for the generation of at least two-thirds of the CH_4 , generated during the conversion of acetate (Jetten *et al.*, 1992). The remaining third is the result of CO_2 reduction by H_2 (Ditchfield, 1986; Mackie & Bryant, 1981).

C. IMPACT OF ENVIRONMENTAL FACTORS

The efficiency of the AD process is governed by specific conditions that must be favourable to the bacterial consortium. These specific conditions include the pH, temperature, alkalinity, volatile acid concentration, nutrient availability, retention time, competition of methanogens with sulphate-reducing bacteria, composition of the wastewater and absence of toxic materials (Chen *et al.*, 1980; Bryant & McInerney, 1981; Forday & Greenfield, 1983; Mata-Alvarez, 2003).

pH - Research has shown that one of the most important environmental factors associated with optimising methanogenic production of biogas is the system pH (Mawson *et al.*, 1991; Speece, 1996). The methanogens and acidogens are extremely sensitive to pH changes and in most cases have an optimum pH growth range of 7.0-7.4. Similarly for optimum CH_4 production, a pH range between 6.7 and 7.4 is essential

(Chen *et al.*, 1980; Bryant & McInerney, 1981; Koster, 1986; Mata-Alvarez, 2003). Acute toxicity occurs when the pH level falls below 6.0 and this is usually associated with the presence of undissociated volatile fatty acids produced by the acidogenic bacteria (Chen *et al.*, 1980; Wang *et al.*, 1999; McMahon *et al.*, 2001). The pH is also a function of the bicarbonate alkalinity, CO₂ partial pressure and the concentration of volatile acids (Chen *et al.*, 1980; Nel & Britz, 1986; Mata-Alvarez, 2003). Methanogens are more susceptible to acidity than the acidogenic bacteria and thus an increase in volatile acid concentration may indicate system upset (Bitton, 1999). The maintenance of an acceptable pH range is brought about by the combined activities of the acetogenic and methanogenic populations. Bicarbonate produced by methanogens serves as a buffering agent during pH reduction (Bitton, 1999; Mata-Alvarez, 2003).

Alkalinity - The alkalinity of an anaerobic system has been shown to be important as it is a measure of the buffering capacity that in an AD normally may consist of bicarbonate, carbonate, ammonia, phosphate and hydroxide components (Ahring, 1995). Organic acids and acids salts may also contribute to the buffering capacity in anaerobic systems. A bicarbonate alkalinity in the range of 2.5 to 5.0 g of CaCO₃.L⁻¹ provides sufficient and a safe buffering capacity for the anaerobic treatment of waste (Chen *et al.*, 1980; Mata-Alvarez, 2003).

Organic acids - The organic acid level of anaerobic systems is also an important determinant of the digestion efficiency (Ahring, 1995; Wang *et al.*, 1999). It has been well documented that the organic acid level should remain below 2000 mg.L⁻¹ of acetate for efficient digestion and that higher levels have been shown to be toxic (Chen *et al.*, 1980; Mata-Alvarez, 2003). Acute methanogenic toxicity occurs at unionised volatile acid concentrations of between 30 and 60 mg.L⁻¹ as acetic acid and this, under certain conditions corresponds to a total volatile acid concentration of between 1 650 – 2 600 g.L⁻¹ (Chen *et al.*, 1980; Duarte & Anderson, 1982; Mata-Alvarez, 2003). At this concentration the pH is usually well below 6.3 (Taconi, 2004).

Temperature - Temperature is another important environmental factor that will negatively impact the anaerobic digestion processes. Two optimum temperatures have been reported for the anaerobic treatment of organic wastewater. The optimum for mesophilic anaerobic digestion is 35°C (usually ranges between 25° and 40°C) and for

the thermophilic is 60 °C (usually ranges between 50 ° and 65 °C) (Patel & Madamwar, 1984; Uemera & Harada, 1995; Bitton, 1999). It has also been shown that it is less expensive to produce CH₄ at higher temperatures. Higher temperatures during digestion give rise to faster fermentation rates, which directly impact the loading rates and also leads to a minimisation of bacterial and viral pathogens (Bitton, 1999). Increases in temperature, especially between 35 ° and 65 °C, may also cause increases in total volatile acid concentrations and therefore induce a higher sensitivity to toxicants (Bitton, 1999). At very low temperatures (below 10 °C) very little CH₄ is produced. The microbial cells, however, remain viable and continue to grow and will actively produce CH₄ when the incubation temperature is increased to 35 °C. It has been reported that during incubation of mesophilic methanogens at 45 °C no growth or CH₄ production occurs and the cells were found to become non-viable after four weeks of incubation at this temperature (Patel & Madamwar, 1984; Taconi, 2004).

Nutrients - To maintain the anaerobic digestion process, the wastewater being treated must be nutritionally balanced (Bitton, 1999). The presence of nutrients, such as nitrogen, phosphorus and sulphur as well as other trace elements needed by the bacteria play a crucial role in this process (Speece, 1996). The best C:N:P ratio for the AD process is recommended to be 700:5:1 (Bitton, 1999; Mata-Alvarez, 2003). It has also been suggested that for optimal gas production the C:N ratio should be 25-30:1 (Bitton, 1999; Mata-Alvarez, 2003). Some minerals have been found to exhibit a stimulatory effect at low concentrations and these include sodium, potassium, calcium, magnesium and iron (Bitton, 1999; Azbar & Speece, 2000; Mata-Alvarez, 2003). In contrast, at higher concentrations these elements may show inhibitory effects (Bitton, 1999; Mata-Alvarez, 2003). High concentrations of sulphate are known to especially retard the production of methane. The mechanism by which sulphate inhibits methanogenesis is by the competition for the available H₂. The sulphate-reducing bacteria are able to scavenge the available H₂ faster than the CH₄ bacteria (Lusk, 1998). Therefore, this results in the shunting of electrons from the CH₄ generation pathway to sulphate reduction (Bitton, 1999). Methanogenic bacteria, in general, have simple nutrient requirements and those species that require organic materials such as B-vitamins, fatty acids and amino acids for growth, obtain it from other bacterial species that produce them during wastewater catabolism (Jarrel & Kalmokoff, 1988; Bitton, 1999; Mata-Alvarez, 2003).

HRT - The hydraulic retention time is another important operational factor in the anaerobic digestion process. The HRT is dependent on the wastewater properties, carbon concentration and environmental conditions and must be long enough to allow for sufficient digestion of the waste by the anaerobic bacteria (Bitton, 1999). The HRT usually varies with the different types of digesters and is also directly dependent on the operational temperature (Bitton, 1999; Mata-Alvarez, 2003). It usually varies from 10 h to several days (10 – 30). In contrast, in anaerobic digestion systems the minimum solids retention time (SRT) is in the range of 2 - 6 days, depending on the temperature (Forday & Greenfield, 1983).

D. MANAGEMENT OF THE ANAEROBIC DIGESTION PROCESS

From a superficial point of view, biomethanisation appears to be a simple process because it looks as if it only consists of two-steps. As described earlier, the first major step involves the conversion of organic matter to intermediates such as VFAs, CO₂ and H₂. During the second major step the intermediates are metabolised into CH₄ by the methanogens (Mata-Alvarez, 2003). However, these two major steps of the anaerobic digestion process are very sensitive to disturbances in the environment (Table 2.1) and even minor changes can result in digester organic overload and subsequent process failure (Mata-Alvarez, 2003). Causes of digester overload can be ascribed to the substrate containing excess organic biodegradable compounds as well as any environmental occurrence that will result in a decreased concentration of active microorganisms. These can include dramatic organic loads, temperature and pH changes, toxic substances introduced into the system and sudden increased flow rates (Mata-Alvarez, 2003). Under disturbed operational conditions the acidogens show more tolerance and continue to function, metabolising and producing more acids. The methanogens on the other hand are negatively affected by disturbances in their environment and their activity decreases (Mata-Alvarez, 2003). The discrepancy between the activity of the acidogens and methanogens during adverse conditions results in an antagonistic relationship. The higher concentration of acids produced by the acidogens are not removed and thus directly inhibits the activity of the methanogens and subsequently other intermediates are formed, in order to metabolise the accumulated H₂ and formate (Mata-Alvarez, 2003).

Table 2.1. Examples of anaerobic digester operational parameter changes and their impacts (Mata-Alvarez, 2003).

Parameter change	Effect on microbial population	Impact on digester efficiency
Increased flow rate	Decrease in concentration of microorganisms; methanogens growth inhibited - long doubling time	Reduced: % CH ₄ in biogas; lower pH and alkalinity; lower CH ₄ production rate
Change in feed type	Acidogens produce more or less acids;	pH declines Alkalinity declines
Increased feed concentration	Methanogens affected; acidogens produce more acids that methanogens cannot remove	Increased VFA concentration; lower pH and alkalinity; other metabolic pathways needed to degrade compounds.
Presence of toxic substances	pH and alkalinity influenced Specific population destroyed/damaged or inhibited	Consortium balance destroyed
Temperature fluctuations	Microbial activity decreases; might die off	Consortium balance destroyed

Consequently, the pH is lowered and the alkalinity levels decline and digester failure is inevitable if this imbalance persists.

In order to overcome problems that may arise as a result of operational disturbances such as feeding high strength waste at short HRTs, the management or control of the digester needs to be sustained. An anaerobic digestion control system must be set up so as to detect disturbances and even be used to calculate indirect variables from direct measurements. The “health state” of the system should be calculated by measuring process variables such as substrate concentration, flow rate, temperature, hydrogen pressure, gas levels and more specific measurements like pH, alkalinity and VFAs (Mata-Alvarez, 2003). The process stability and efficiency can therefore be established by regularly monitoring the variables as given in (Table 2.2). As some of these variables are more sensitive to disturbances, an early indication of process imbalance must be identified as soon as possible (Mata-Alvarez, 2003). The control of the anaerobic digestion process is essential in establishing efficient handling of unstable processes and for managing and maintaining the operation of systems at optimal conditions (Mata-Alvarez, 2003).

E. APPLICATION OF ANAEROBIC DIGESTION TO DIFFERENT WASTEWATERS

Anaerobic digestion has in the past been used for the dual purpose of treating wastewater and for energy conservation in the form of CH₄ recovery. The treatment of a variety of wastewaters has been studied for many years. In 1986, Koster reported that when using an Upflow Anaerobic Sludge Blanket (UASB) reactor to treat an acidic tomato waste stream to produce methane, as much as 80% of the influent COD was converted to methane. The highest loading rate obtained in this case was 22 kgCOD.m³.d⁻¹ and the treatment efficiency resulted in a 90% COD reduction. Hemming (1981) also reported that biogas containing 70% CH₄ was produced in a full-scale anaerobic digestion plant treating potato processing effluent. The BOD concentration was reduced by 60% at an HRT of 7 d and a total biogas production of 1 800 m³.d⁻¹.

Szendrey & Dorion (1986) reported the successful operation of a 13 million litre fixed-film downflow anaerobic filter while treating distillery wastewater. During this study an optimum loading rate of 12.84 kg COD.m⁻³.d⁻¹ was achieved with a wastewater pH range of 7.2 - 7.4. The volatile fatty acid concentration was considered to be the most important operational parameter and ranged between 3 000 and 4 000 mg.L⁻¹. The average biogas production was 0.56 m³.kgCOD⁻¹_{removed} during the full-

Table 2.2. Operational variables important for controlling the anaerobic digestion process (Mata Alvarez, 2003).

Liquid phase	Gas phase
VFA (type and concentration)	Gas production rate
pH (low, optimum, high)	CH ₄ / CO ₂ production rates & concentrations
Alkalinity (low, high)	CO content in biogas
OLR	Gas pressure
Nutrient level and additions	H ₂ content in biogas
HRT	Temperature
C:N:P ratios	Gas phase volume
Temperature	
COD (type and concentration)	
Liquid level	

scale study. They also reported that the CH₄ content (50 – 65%) of the biogas was sensitive to the pH and volatile acid concentrations.

Nand *et al.* (1991) used canteen and mess wastewater in a study to determine their biogas generation potential. The anaerobic digestion was performed in a floating dome type digester and certain of the environmental and operational parameters influencing the biogas yield and the CH₄ content were optimised. During this study a high gas yield of 0.981 m³.kg⁻¹ VS_{added} was obtained with a CH₄ content of 50% and 65% substrate utilisation. The gas yield was directly proportional to the HRT at 27° - 33°C, and an OLR of 100 kg total solids m⁻³.d⁻¹ was achieved.

Anaerobic treatment of winery and distillery wastewaters has also been researched (García-Bernet *et al.*, 1998; Ronquest & Britz, 1999; O’Kennedy, 2000; Ruiz *et al.*, 2002). O’Kennedy (2000) used a mesophilic lab-scale UASB reactor to treat high strength distillery effluent. The average COD removed was >90% at an OLR of 30 kgCOD.m⁻³.d⁻¹ with a pH of 7.8 and biogas production of 18.5 L.d⁻¹.

The anaerobic treatment of various fruit and vegetable wastewaters has also been documented. Viswanath *et al.* (1992) determined the effect of different fruit and vegetable wastewater on digester performance in terms of biogas production in a 60 L digester. Different OLRs and different HRTs were studied. The maximum biogas yield was 0.6 m³.kg⁻¹ VS_{added} at a 20 day HRT and an OLR of 40 kgTS.m⁻³.d⁻¹.

Austermann-Haun *et al.* (1994) treated fruit juice factory effluent using an UASB reactor. A space loading rate of 3.6 kgCOD.m⁻³.d⁻¹ and an influent COD concentration of 2 337 mg.L⁻¹ was used. The COD removal efficiency achieved was 80 – 90%. Trnovec & Britz (1998) also treated fruit canning wastewater using a mesophilic-laboratory scale UASB reactor. The OLR was 10.95 kgCOD.m⁻³.d⁻¹ with COD removals of 90 – 93 % and HRT of 10 h. More recently Britz *et al.* (2000) reported that an UASB reactor treating fruit cannery wastewater devoid of lye, can achieve OLR’s of 9.2 kgCOD.m⁻³.d⁻¹ with an average COD removal of 81 – 84 % and HRT of 10 h. The influent COD levels ranged between 3 800 – 4 300 mg.L⁻¹ at an influent pH of 5.0.

Some effluents are an ideal substrate for UASB digestion, like wastewater originating from the sugar beet industry (Van Lier *et al.*, 2001). However, other wastewaters that contain complex organic material may not be treated as easily using the UASB process (Lettinga *et al.*, 1980). Problematic constituents such as pectin found in effluents originating from the fruit processing industry will influence the process and could lead to various problems in the UASB reactor (Fedirici *et al.*, 1988). Other

effluents such as apple juice processing wastewater require pH adjustment before the UASB treatment process can proceed. This is due to the wastewater's low pH range (pH 3 – 5) (Wayman, 1996). However, Britz *et al.* (2004) showed that the use of ozone as a pre-treatment improves the suitability of apple juice processing wastewater for UASB degradation by breaking the pectin gel that would otherwise interfere during normal UASB digestion.

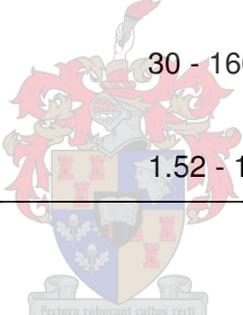
F. BIOGAS FROM ANAEROBIC DIGESTION

Anaerobic digestion of biodegradable waste results in both potential energy generation and reduction of greenhouse gas emissions (Baldasano & Soriano, 2000; Keller & Hartley, 2003). Methane, an energetic constituent of biogas, is generated during anaerobic wastewater treatment as a by-product and can be used as fuel for boilers or reactor heating and for electricity generation (Mendonca & Campos, 2001). Biogas is a renewable source of energy with much lower environmental impacts than conventional fossil fuel (Fan *et al.*, 2001; Ho, 2005).

Biogas – It has been reported that anaerobic digestion of organic waste releases between 500 – 800 million tons CH₄ annually into the atmosphere (Bitton, 1999). Biogas is comprised of four major constituents: methane, carbon dioxide, moisture and hydrogen sulphide. The concentrations are given in Table 2.3. The normal composition of biogas from efficient operating anaerobic digestion systems ranges from 60 – 70% CH₄ with the balance being CO₂. The energy content of biogas is entirely associated with the CH₄, which has an energy value of 37 MJ.m⁻³. The amount of CH₄ that can be produced during the anaerobic digestion process from organic material is directly proportional to the substrate content of convertible COD. Since no oxidation by atmospheric O₂ can occur the biodegradable COD from the substrate will be preserved in the end-products. Stoichiometrically CH₄ has a COD of 2 moles (= 64 g of COD) of oxygen per mole (= 16 g) of CH₄. Thus 1g of CH₄ is equivalent to 4 g of COD. Further calculations show that from 1 Kg of COD 0.355 m³ CH₄ at STP can be produced which is equivalent to 14 132 kJ usable energy (as CH₄). Methane has a critical temperature and pressure of - 82°C and 4.6 MPa, respectively (Chen *et al.*, 1980; Chynoweth *et al.*, 2001; Mata-Alvarez, 2003). It is not possible to liquefy or compress CH₄ at higher temperatures (Mata-Alvarez, 2003). Biogas can be

Table 2.3. Composition of Biogas.

Constituent	Value (range)
Methane	50 - 65%
Carbon Dioxide	35 - 50%
Moisture	30 - 160 g.m ⁻³
Hydrogen Sulphide	1.52 - 12.5 g.m ⁻³

A watermark of a university crest is centered on the page. It features a shield with various symbols, topped by a crown and supported by two figures. Below the shield is a banner with the Latin motto "Pectora roborant cultus recti".

stored for long periods at reasonable costs this property allows for the conversion of biogas into electricity during hours when the electricity is expensive or periods of high electricity demands (Mata-Alvarez, 2003).

Carbon dioxide is the other major constituent of biogas. Its characteristics are dependent on different factors such as substrate composition, pH, reactor pressure, temperature, HRT, process design, etc (Mata-Alvarez, 2003). The effect of CO₂ in biogas is that it dilutes the energy value of biogas and increases the volume to be handled and stored. CO₂ removal is not necessary for thermal application (Mata-Alvarez, 2003). However, when feeding gas into a public gas pipeline and for its storage under pressure (fuel for cars) the CO₂ has to be removed (Mata-Alvarez, 2003).

Biogas also contains water vapour, which is an important contaminant that should be removed before application (Table 2.3). Condensed water poses a problem when it accumulates in gas handling equipment and meters. This causes problems such as frozen pipes and corrosion of metal parts when combined with the H₂S also present in the gas (Chynoweth *et al.*, 2001). The water can be condensed by water-traps (Mata-Alvarez, 2003).

Biogas has to be purified in order to meet natural gas pipeline standards. The most economical and commercial cleaning process used is water scrubbing (Chynoweth *et al.*, 2001). Other methods such as the phosphate buffer and membrane separation processes are comparable in cost but neither has been extensively field-tested. Water scrubbing is a relatively simple process in which the H₂S-free biogas is compressed and then flows “counter current” to water in a pressurized packed column. The scrubbed CO₂ is vented to the atmosphere and the water recycled back to the stripping column. The scrubbed biogas is then dehumidified to meet natural gas pipeline standards. H₂S can be removed with a simple and common method called the iron oxide (or iron sponge) process (Chynoweth *et al.*, 2001).

Biogas Carbon Trading - The benefits of biogas recovery from anaerobic digesters to rural communities have been well demonstrated in developing countries such as India, China, Sri Lanka and Nepal (Ho, 2005). The rural energy benefits provided by biogas recovery includes energy for cooking and lighting. Other benefits of promoting biogas recovery have to do with “carbon trading”. The United Nations Framework Convention on Climate Change has set up a “Clean Development Fund”, and the World Bank has provided a “Carbon Finance Unit”, which allows developed countries to buy emissions

when more carbon is released into the atmosphere than what is allowed according to the Kyoto Protocol from developing countries. This will prevent carbon emissions through conserving forests and renewable energy (Ho, 2005).

Internationally, Nepal's biogas programme is regarded as a model for the successful use of alternative energy for the rural Third World. Nepal has 125 000 functioning anaerobic digesters treating various domestic, agricultural and industrial wastes which prevents 5 tonnes of carbon dioxide equivalents annually from being emitted into the atmosphere. This amounts to US\$ 5 million in greenhouse gas emissions that can be traded (Ho, 2005). This money can be invested back into clean energy production, therefore preventing even more greenhouse gas emissions.

Biogas recovery from anaerobic digesters yields resources with significant financial and intangible value. Community biogas plants, especially in rural Third World countries, may serve as the most useful decentralised sources of energy supply and may to some degree reduce our dependence on fossil fuels (Angenet *et al.*, 2004; Ho, 2005).

G. DISCUSSION

In order to overcome the major problem of water scarcity and processing effluent disposal in the South African fruit processing industry, initiatives such as on-site wastewater treatment technologies and energy recovery technologies should be undertaken.

Biological treatment is one of the most efficient and economical ways to reduce high COD values. Aerobic digestion is the traditional and most widely used option to treat oxygen consuming wastewater. Various problems are, however, associated with aerobic processes such as high energy requirements ($>100 \text{ kW}\cdot\text{ton}^{-1}$ of COD reduced) and the production and disposal of large sludge quantities (50% of the COD). Anaerobic treatment of wastewater may be seen as a technology which is an energy-efficient approach to waste management (Bitton, 1999). A positive energy balance due to the production of biogas and low sludge volume productions are of the main advantages of AD. Anaerobic digestion can be seen as a mature technology and many successful systems have been installed worldwide.

On-site anaerobic treatment of effluents offers several advantages to the industries. It allows for water re-use, therefore enabling lower capital outlays for water

as well as lower sur-charges for waste disposal. The main advantage is energy recovery via CH₄.

Methane is an ideal fuel because it is the cleanest most non-polluting fuel currently in use. Methane is used to provide the US with at least 20% of its energy supply and is mainly for domestic, industrial and municipal application (Price & Cheremisinoff, 1985). The beneficial use of CH₄ compared to other fossil fuels has set the trend toward its increased application for operation of general appliances, vehicles, industrial purposes and power generation (Chynoweth *et al.*, 2001). Recovering energy by the food and beverage industry will help reduce the energy cost and somewhat reduce the industries dependence on fossil fuels (Angenet *et al.*, 2004).

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CHAPTER 3

ESTABLISHMENT OF AN OPERATIONAL TREATMENT PROFILE FOR AN ANAEROBIC POND SYSTEM TREATING FRUIT-PROCESSING WASTEWATER

Summary

A pond system treating wastewater from a fruit-processing factory was monitored during 2004. The influence of processing and environmental conditions on the pond performance were established by monitoring various process parameters. Four domes were placed and secured on the surface of the pond to facilitate four horizontal sampling positions. Beneath each dome two vertical sampling positions were established at 1 and 3 m from the surface. Various process parameters including chemical oxygen demand (COD); pH; alkalinity; volatile fatty acids (VFA's); temperature; sludge volume; mineral content, and nutrient content (COD:N:P) were monitored. The results showed that the COD levels decreased during the off-peak season but the pond pH remained relatively stable between 6.0 and 6.4 during the entire year. Pond alkalinity was found to be dependant on the regular lime dosing to maintain the necessary alkalinity. The VFA concentrations indicated that the microbial populations inside the pond were functioning well. However, a decrease in microbial activity and VFA concentrations were observed at the lower temperatures during the winter months. The temperature profile of the pond showed that the pond temperature is impacted by the fluctuations in the ambient air temperature. The settled sludge volume of the pond was also found to vary according to the peak and off-peak seasons. The mineral content throughout the year was maintained at sufficient levels and appeared to exert no inhibitory effects on the pond activity. The COD:N:P ratio varied throughout the year, mainly due to variations in the N and P levels, brought about by differences in the fruit being processed.

Introduction

The Western Cape Province is known for its extensive deciduous fruit industry and is home to a variety of fruit processing plants (Hurndall, 2005). This fruit industry caters for the export and local market as well as for juice processing industries. The export market generates billions of Rand in foreign exchange annually (Birch, 1993; Hurndall, 2005) and accounts for 85% of South Africa's total agricultural exports (Wesgro, 2002). The local market is supplied with fruit not suitable for export and the processing industry receives the surplus fruit.

One of these fruit processing factories is situated in the Overberg area. The factory processes 70 000 tons of apples and 9 000 tons of pears annually, producing concentrates and a variety of carbonated fruit juices for the local and international markets (E. Muller, Appletiser SA, South Africa, personal communication, 2004). Processing and peak season usually starts in mid-February and continues until mid-July. The fruit-processing factory is supplied with fresh water from a nearby river. The fresh water is treated at a water treatment plant to produce water of a quality suitable for production, according to certain standards required by the processing plant (E. Muller, Appletiser SA, South Africa, personal communication, 2004). The production process in turn, generates large volumes of wastewater that are treated on-site via a pond and wetland system. The treated wastewater is then irrigated onto the surrounding orchards.

The aim of this study was to monitor the wastewater treatment system, specifically the anaerobic pond system (Pond A), during the peak and off-peak seasons in order to evaluate the treatment efficiencies and limitations and establish a characteristic operational profile for the anaerobic pond. In this chapter no biogas monitoring was undertaken as it will be included in Chapter 4 of this thesis.

Materials and methods

Wastewater treatment system design

The fruit factory wastewater treatment plant consists of an intricate pond system (Fig. 3.1). The wastewater is pumped from the processing plant to the screening and lime dosing facility. Here the wastewater passes through a 0.5 mm screen to remove large particles and lime is dosed. The wastewater is pumped to Pond A, an anaerobic pond of 6 600 m³,

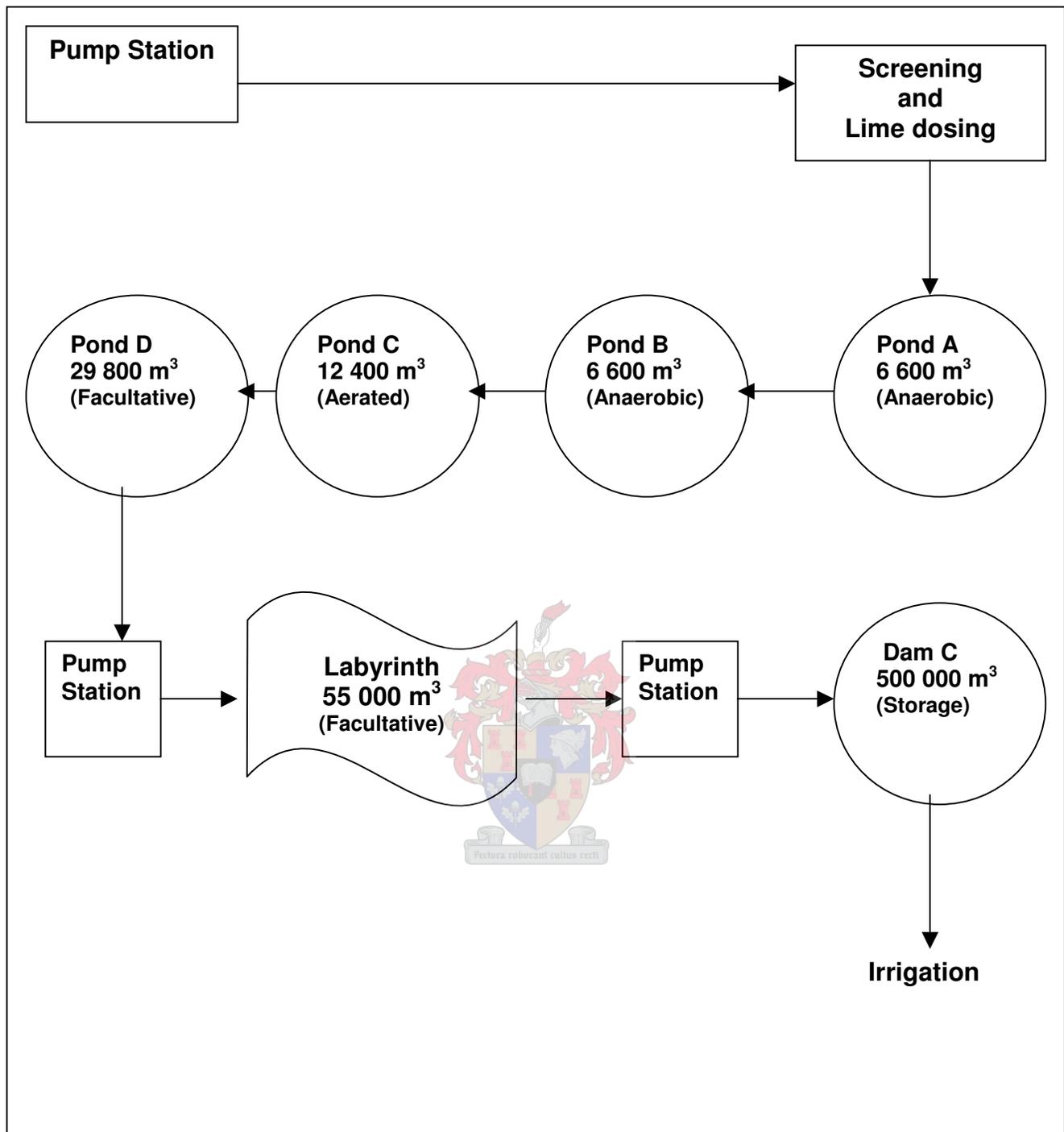


Figure 3.1 A schematic representation of the fruit factory wastewater treatment system.

which is 4 m deep. Wastewater is fed into the anaerobic pond by a distribution system consisting of a nine-point distribution manifold evenly spread over the base of the pond (Fig. 3.2). The wastewater percolates upward through a blanket of sludge containing the different microbial populations that are responsible for the treatment process. The pond has a surface area of 2 100 m², an overflow rate of 1.4 m.d⁻¹ at a flow rate of 3 000 m³.d⁻¹. The treated effluent overflows into launders situated on the sides of the pond. The anaerobic pond is designed for a chemical oxygen demand (COD) loading rate capacity of 5 kg COD.m⁻³.d⁻¹ and, as far as possible, to imitate the operation of an Upflow Anaerobic Sludge Blanket (UASB) process (E. Muller, Appletiser SA, South Africa, personal communication, 2004). The anaerobic pond is followed by a secondary anaerobic pond, an aerobic pond equipped with two 55 KW surface aerators, and a facultative pond (Fig. 3.1). The effluent then flows through the wetland labyrinth, which is also facultative, into the storage dam (Dam C) before the treated wastewater is irrigated onto surrounding orchards during the summer months.

Experimental design

For clarification purposes summer was taken as the period January to April (day 0 – 149) and September to December (day 250 – 350), winter was taken as May to August (day 150 – 249), the peak season was taken as the period mid-February to mid-July (day 50 – 200) and off-peak season was taken as from day 201. It was extremely difficult to define the end of the peak season and the start of the off-peak season as a result of the availability of fruit affecting the duration of the daily plant operation. Thus for practical reasons day 200 was arbitrarily taken as the end of the peak season. A similar problem occurred towards the end of the off-peak season when the availability of fruit gradually increases at the onset of summer.

Initially four domes ($\theta = 1$ m, with stainless steel frames, covered with PVC) were constructed to serve as gas collection vessels on Pond A. The floating domes were positioned on the surface across Pond A and secured by a cable fixed over the pond. The four domes were colour coded as green, red, blue and yellow to facilitate identification. The green, red, yellow and blue domes were positioned approximately at 30, 26, 24 and 14 m from the side of Pond A, respectively (Fig. 3.2). Each dome was fitted with a black PVC pipe, which functioned as the gas carrier and to vent the biogas to the atmosphere.

Sampling took place beneath each dome at a depth of 1 and 3 m from the surface of Pond A. Thus, in total there were 8 sampling points. For the incoming COD values (COD_{IN})

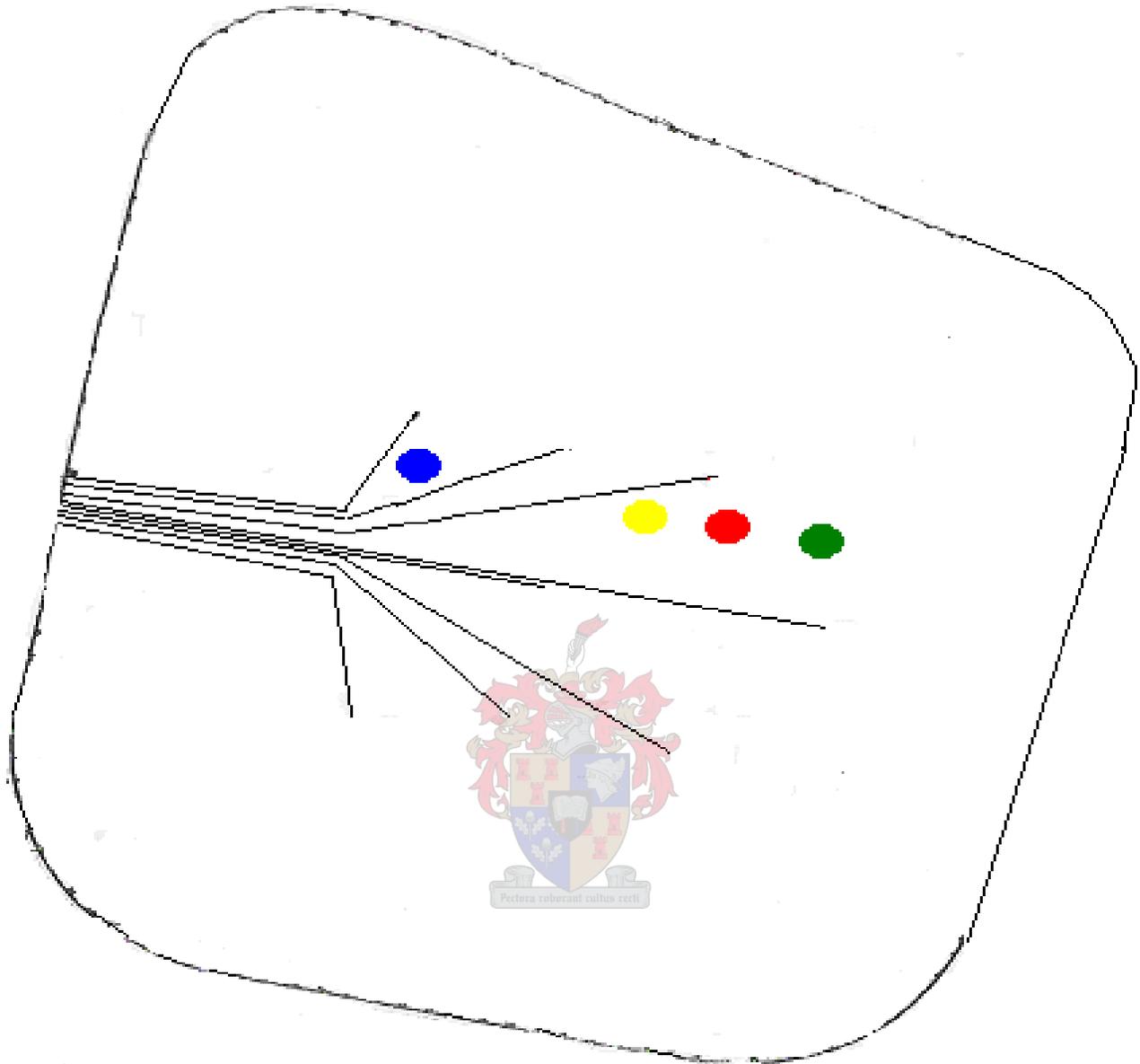


Figure 3.2 An outlined representation of Pond A, showing the nine-point distribution manifold and dome positioning.

as well as the COD of Pond A overflow (COD_{OUT}) sampling was done twice each week over a period of a year. These analyses were done as part of the processing plants' routine analysis. During the period of day 286 – 322 no values were obtained due to instrument failure. For the 1 and 3 m depths COD sampling was done weekly from day 75 to day 348 (in total 30 weeks) at the four sampling positions (for each of the four domes). This ensured that the data collected from each dome was representative and that comparisons could be made between different positions and depths in the pond.

Sampling

Water samples (1 L) at the different vertical positions were taken with the aid of a submersible water pump. The water pump was attached to a second cable that spanned the pond. The cable served as a guide to position the water pump with the aid of a pulley system. Once the pump was stabilised at the required dome, the pump was lowered to the required sampling depth. Samples were collected from a garden hose pipe connected to the pump. At the required sampling point, the pump was allowed to pump water through the garden hose for a few minutes before the 1 L samples were taken. This was done to ensure that the pipe was cleared and that the sample being collected was representative of the specific sampling point.

Process parameters

The following operational parameters were monitored on a weekly basis according to Standard Methods (APHA, 1998): COD; pH; alkalinity; volatile fatty acids (VFA's); and settled sludge volumes. The pH of the incoming wastewater was monitored daily over a period of a year and was done as part of the processing plants' routine analysis. The pH values at the 1 and 3 m depths were monitored weekly for 30 weeks at each of the four domes. Temperature was determined using a minimum and maximum thermometer and was monitored weekly for 30 weeks.

Mineral analyses were carried out on a monthly basis at the Central Analytical Facility of Stellenbosch University. The mineral composition of the wastewater was determined with a Varian Liberty II Radial ICP-AES at 1.2 kW power with a flow rate of $15 \text{ L}\cdot\text{min}^{-1}$ and a viewing height of 7 mm. The sample introduction was with a V-groove type nebulizer and a Sturman Masters spraychamber. The following ions: Ca^{2+} , Fe^{2+} , K^+ , Mg^{2+} and Na^+ were determined at the corresponding wavelengths (λ): 317.933, 259.94, 769.896, 276.553 and 589.592, respectively. The total Kjeldahl nitrogen (TKN) and total phosphorous were

analysed by the Council for Scientific Industrial Research (CSIR) according to Standard Methods (APHA, 1998).

Results and discussion

Operational profiles

The process parameter data were used to set-up a series of profiles over the trial period in order to characterise the overall “system profile”. For these profiles the peak and off-peak season periods were taken from February to mid-July to end-December, respectively.

COD profile

The COD profile for Pond A over a 350 d period is shown in Fig. 3.3. The average COD_{IN} , COD_{OUT} and % COD removal is given in Table 3.1. From the data it is evident that the COD concentration of the wastewater flowing into Pond A fluctuates greatly during the peak season (Fig. 3.3 A). The highest COD concentration recorded was $16\,500\text{ mg.L}^{-1}$, while an average of $5\,020\text{ mg.L}^{-1}$ was maintained during the peak season. COD samples from the incoming wastewater were taken on a daily basis and are therefore not representative of the average influent COD over a 24 h period. The data also showed that there was a decrease in the COD levels of the incoming wastewater during the off-peak season, which is also mostly during the winter months (Fig. 3.3 A). This was attributed to a less intense production schedule during the winter.

During the peak season the average COD concentration at a depth of 1 m in the pond (Fig. 3.3 B) was 521 mg.L^{-1} , while the average COD concentration was 251 mg.L^{-1} during the off-peak season. The maximum COD concentration recorded at 1 m was $1\,834\text{ mg.L}^{-1}$ during the peak season and 754 mg.L^{-1} during the off-peak season (Fig. 3.3 B). The abnormally high COD concentrations in Fig. 3.3 A (day 106, 148 and 201) can possibly be ascribed to the very high COD concentrations of the incoming wastewater and were all correlated with production spills in the processing plant.

The average COD concentrations at the 3 m level (Fig. 3.3 C) for the peak and off-peak season were 813 mg.L^{-1} and 558 mg.L^{-1} , respectively. The maximum COD concentration recorded during the peak season for the 3 m depth was $2\,214\text{ mg.L}^{-1}$, while the maximum COD concentration during the off-peak season was 516 mg.L^{-1} .

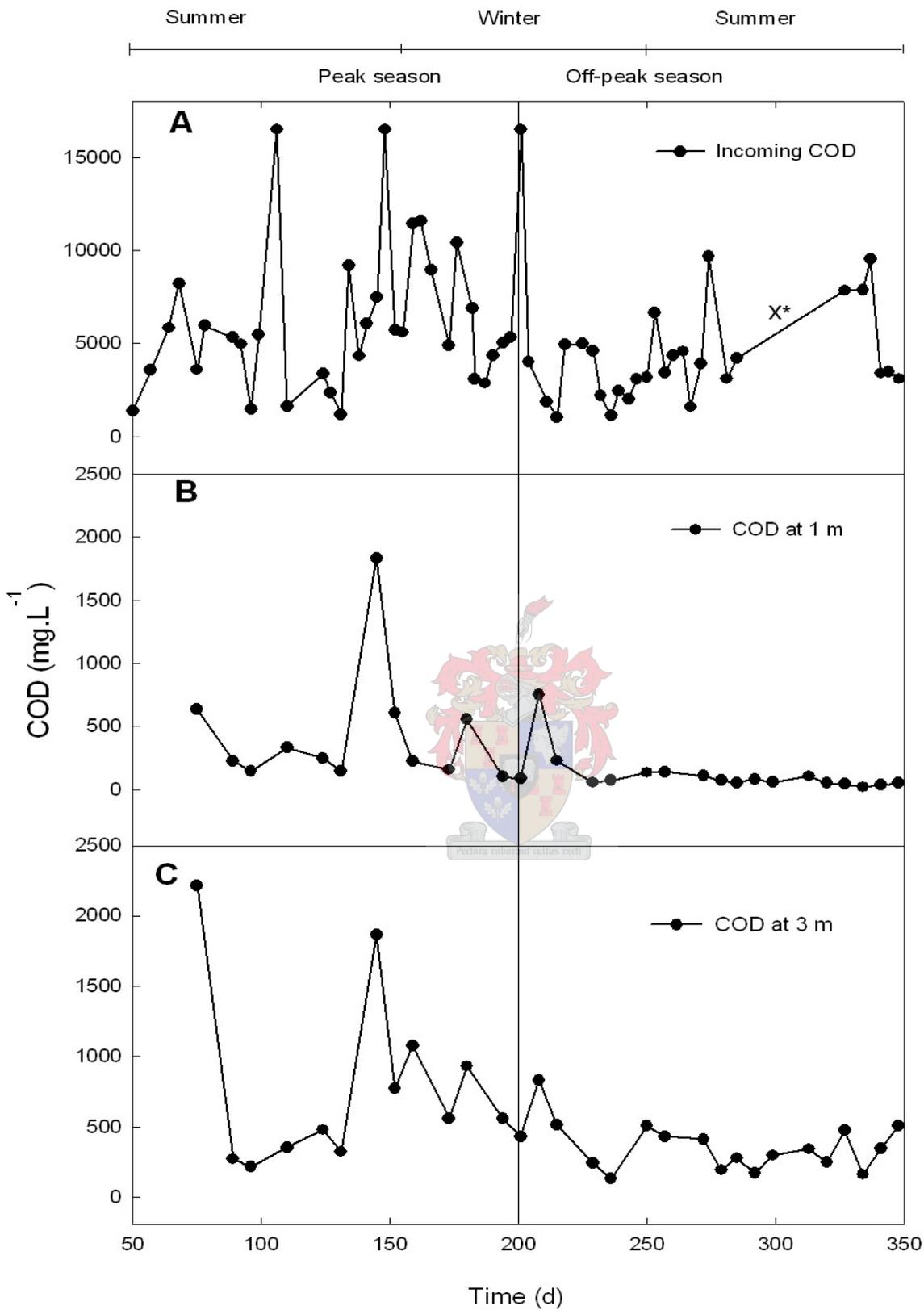
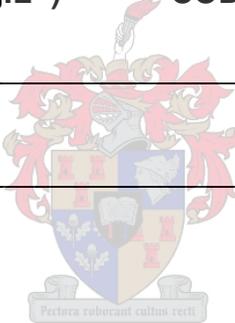


Figure 3.3 The COD concentrations of the incoming fruit processing wastewater (A), at a depth of 1 m (B), and at a depth of 3 m (C) during the peak and off-peak season in Pond A. (Variation per sampling point set for each depth showed a variation of less than 5%) (X*= no values for this period)

Table 3.1 The average (of the years data) COD_{IN} (incoming processing wastewater, twice weekly for 30 weeks), COD_{OUT} (Pond A overflow, twice weekly for 30 weeks) and % COD reduction for Pond A for the 2004 year.

	COD_{IN} (mg.L⁻¹)	COD_{OUT} (mg.L⁻¹)	% COD Removal
Pond A	5 450	419	91 %



The higher average COD concentration at both the 1 and 3 m depths during the peak season was expected, as a continuous 24-hour production schedule took place in the processing plant. This intense production program, which generates large volumes of wastewater, led to relatively short hydraulic retention times (HRT) of about 3 days in Pond A. This in turn, directly resulted in the higher COD concentrations at the 1 and 3 m levels. The lower average COD concentration at both the 1 and 3 m depths during the off-peak season was ascribed to the lower incoming COD concentrations as a result of the less intense production schedule, which then gave longer HRT's (between 5 and 8 d). The initial COD concentrations being lower resulting in longer HRT's and subsequently resulting in more efficient COD reductions. The stable COD concentration during the off-peak season (Fig. 3.3 B and C) can also be due to sedimentation of the biodegradable settleable solids. This may be influenced by the pond temperature and HRT (Pescod, 1996).

pH

The pH profile of the incoming wastewater and of the pond at the 1 and 3 m depths is shown in Fig. 3.4. The incoming wastewater pH varied widely from 3.8 to 11.9 and this was ascribed to the nature of the production process, cleaning operations and the fruit quality.

The pond pH, however, was found to be very stable at both the 1 and 3 m depths. The pH at 1 m (Fig. 3.4 B) ranged from 5.6 to 6.8 throughout the year, while the pH at 3 m ranged from 5.7 to 6.7 (Fig. 3.4 C).

The stability in pond pH at both the 1 and 3 m depths throughout peak and off-peak season was thought to be significant, facilitating overall process efficiency (Mawson *et al.*, 1991; Speece, 1996; Azbar *et al.*, 2000; Reith *et al.*, 2003). This stability was also taken as an indication of sufficient buffer capacity, which is important when the incoming wastewater pH varied so widely.

Alkalinity

A representation of the alkalinity profile of Pond A at the 1 and 3 m depths is shown in Fig. 3.5. The alkalinity at 1 and 3 m exhibited a similar trend and indicated that there was very little difference in alkalinity at the two depths. This suggests that sufficient mixing and welling took place in the pond.

The alkalinity values at the 1 m depth (Fig. 3.5 A) ranged from 437 to 756 mg.L⁻¹, during the peak season. Whereas a lower alkalinity range of 168 to 512 mg.L⁻¹ was obtained

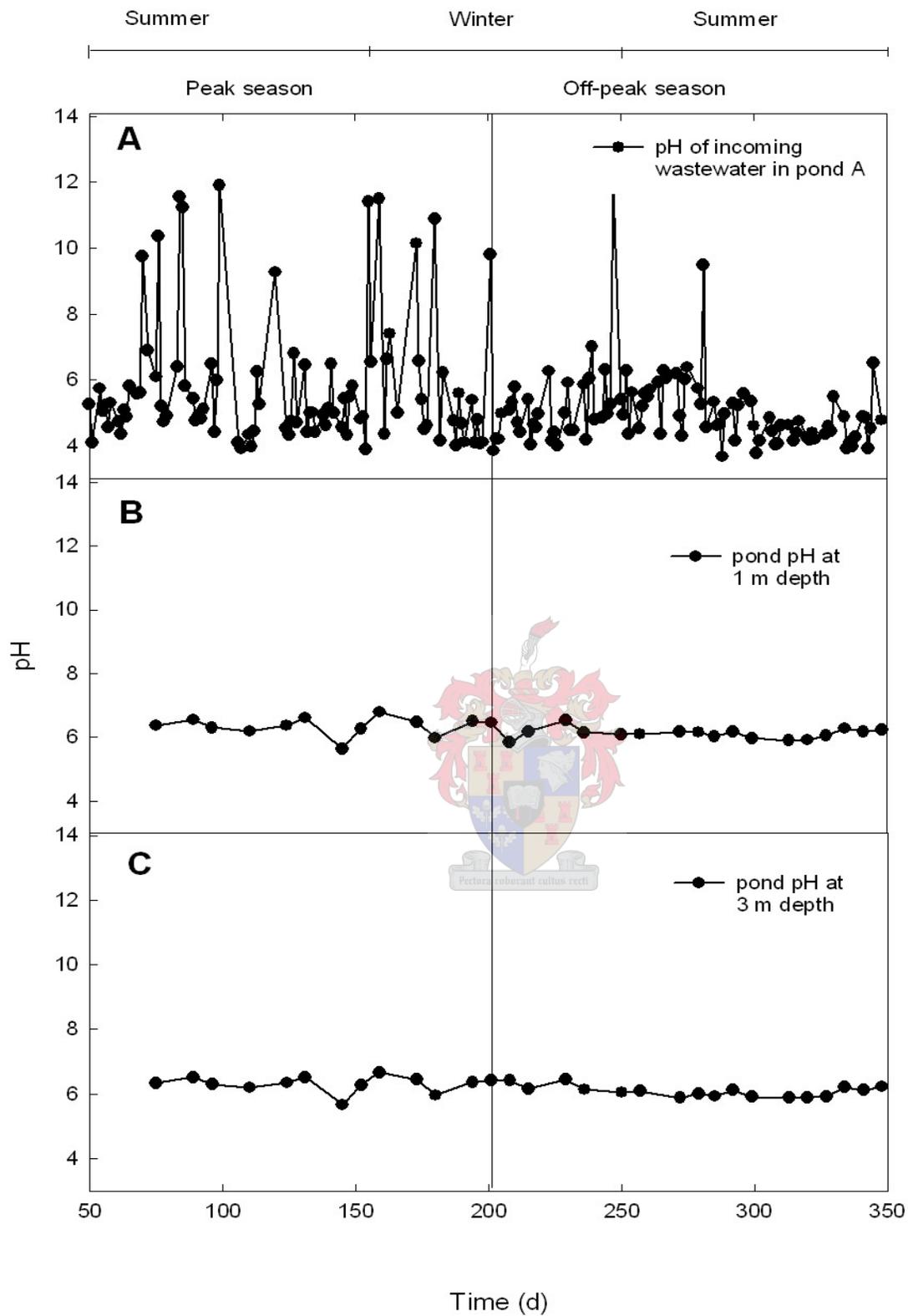


Figure 3.4 The pH of the incoming fruit processing wastewater (A), pH at a depth of 1 m (B), and the pH at 3 m depth (C) during the peak and off-peak season in Pond A. (Variation per sampling point set for each depth showed a variation of less than 0.2 units).

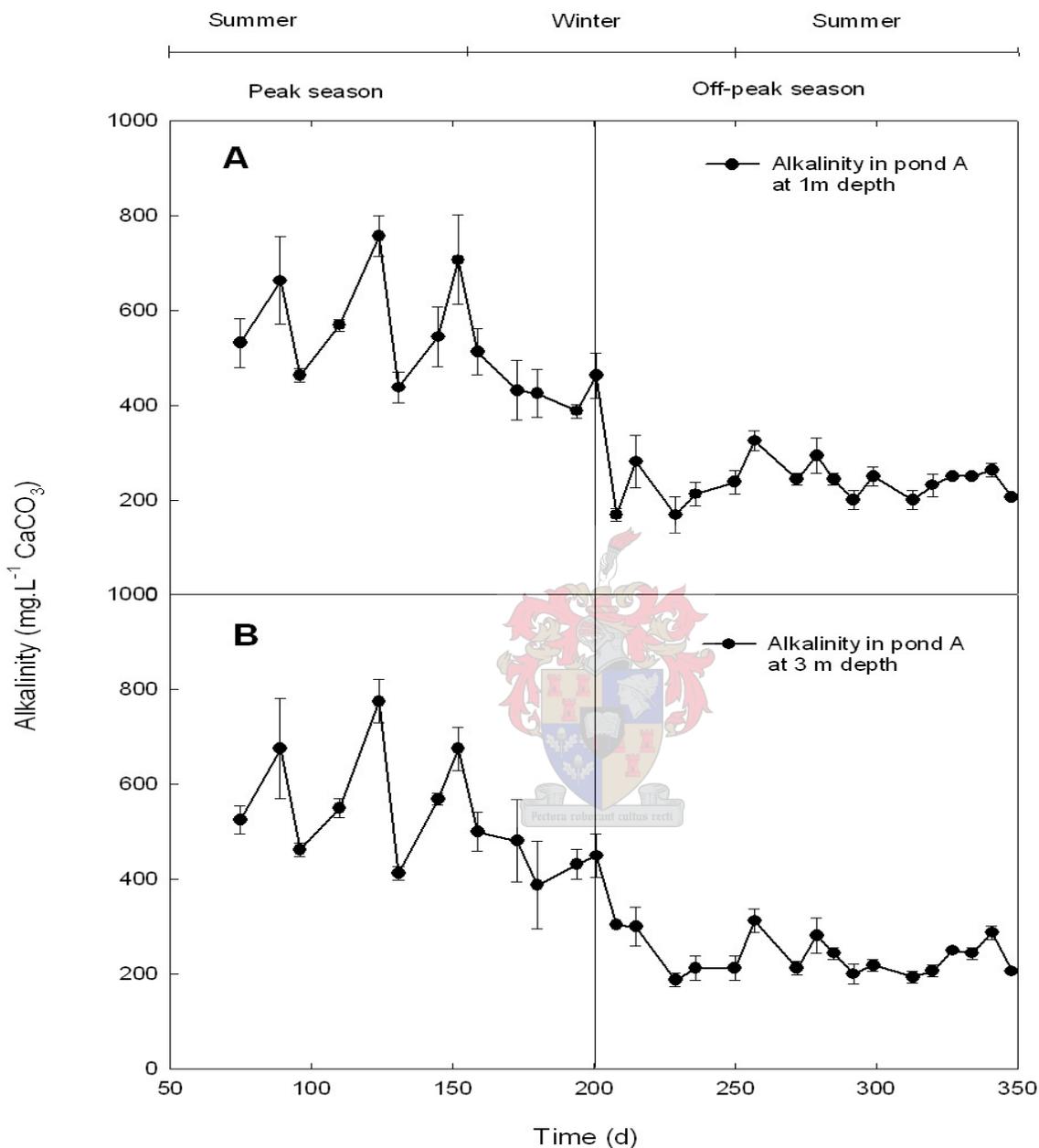


Figure 3.5 The pond alkalinity at a depth of 1 m (A) and at a depth of 3 m (B) during the peak and off-peak season in Pond A. Data points represent samples taken at the four different domes at both the 1 and 3 m depths. The error bars are the standard deviations based on data from the four monitoring points.

during the off-peak season. The alkalinity at 3 m (Fig. 3.5 B) showed an alkalinity range of 412 to 775 mg.L⁻¹ during the peak season and 187 to 500 mg.L⁻¹ range during the off-peak season.

It is clear from the data in Fig. 3.5 that the alkalinity is higher during the peak season than during the off-peak season. This was ascribed to regular lime dosing. The data in Fig. 3.6 shows the direct correlation between the lime dosing and alkalinity in Pond A. Therefore, it was concluded that the alkalinity profile is a direct consequence of the lime dosing intervals. The alkalinity of the pond during peak and off-peak season is much lower than specified in the literature for optimum operating conditions for anaerobic digestion (Chen et al., 1980; Bitton, 1999). However, the alkalinity levels in Pond A appear to be effective for the digestion taking place in this particular anaerobic pond. This was also correlated to the stable pH profile found at the 1 and 3 m depths (Fig. 3.4).

Temperature

The temperature profile of Pond A (based on the weekly measurements Fig. 3.7) shows the variation in pond temperature during the year. It was interesting to note that climatic temperature changes during the summer and winter months did influence the pond temperature. During the summer months the pond temperature could reach 32°C whereas during the winter months a temperature as low as 13°C was reached (Fig. 3.7). It is also important to note that the peak season lapped into the winter months (June – July) and similarly the off-peak season lapped into the summer months (November to January and some years even February).

The temperature profile of Pond A (Fig. 3.7) was further found to resemble the average daily air temperature-curve as shown in Fig. 3.8. It was found that the average daily air temperature decreased from January (day 0) to July (day 183) (Fig. 3.8) and similarly the pond temperature also decreased (Fig. 3.7). Thus, it was concluded that the environmental impacts, especially temperature, on the pond system may impact the treatment efficiency of the pond system.

The atmospheric air temperature variations throughout the year were thus found to impact the pond temperature and subsequently the efficiency of the anaerobic digestion process. It is known that low temperatures (10° – 15°C) during winter months are not favourable for the microbial populations in the sludge layer and digestion and methane production will decrease (Patel, 1984). It is also well known that methanogenesis rarely

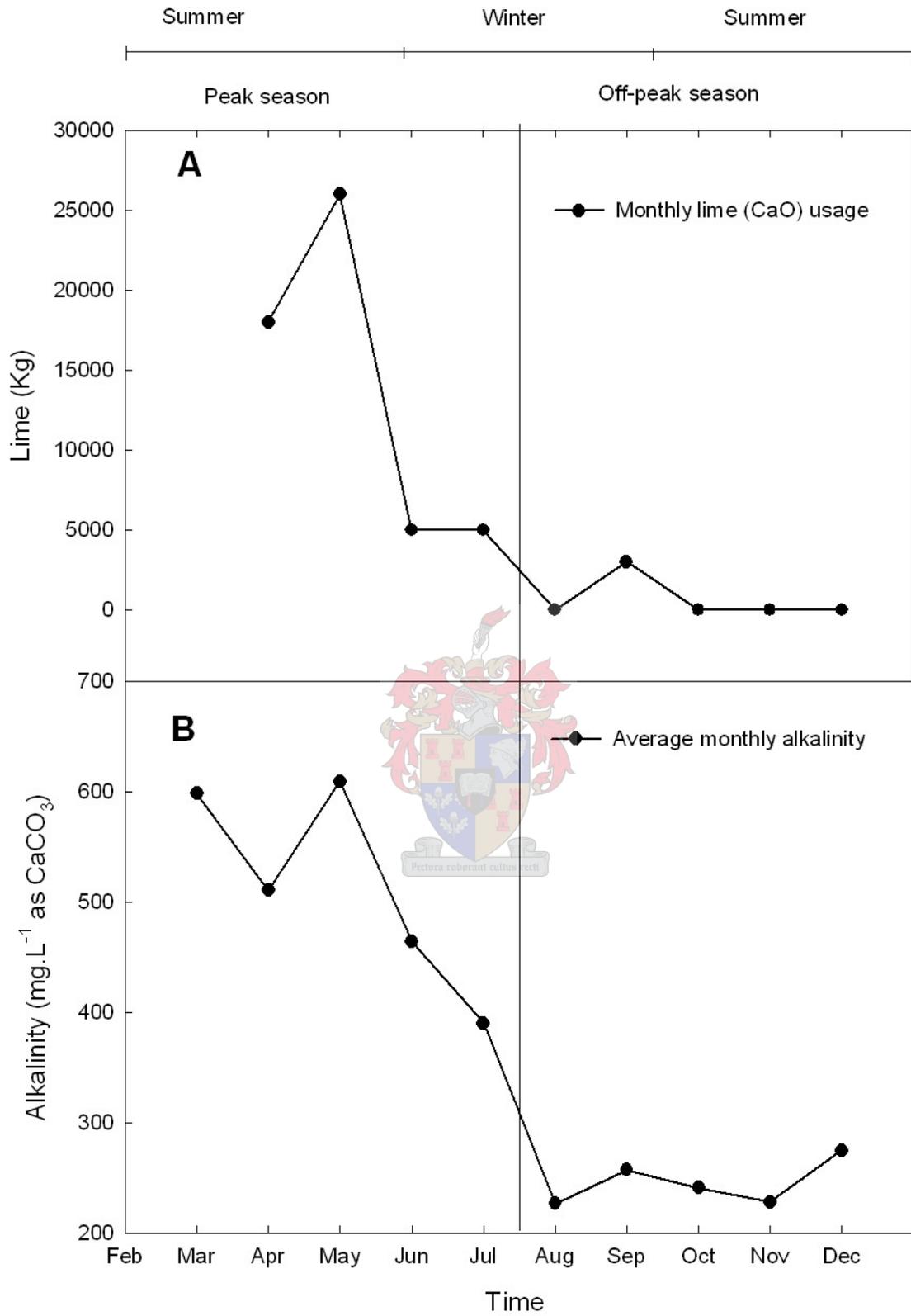


Figure 3.6 The monthly lime (CaO) usage (A) versus the average pond alkalinity (B).

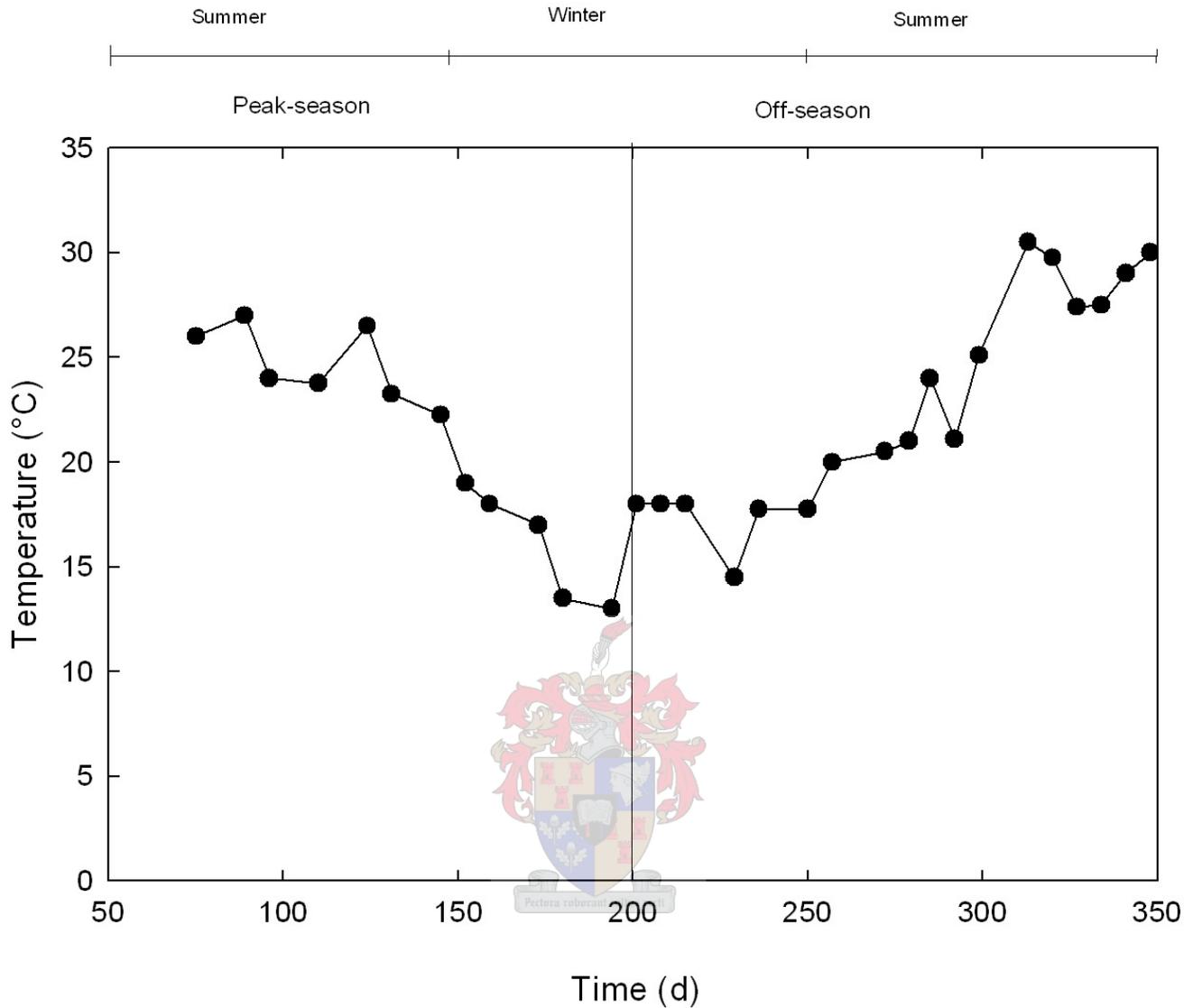


Figure 3.7 The weekly average temperature variation in Pond A during peak and off-peak season. Average of 8 values measured on a weekly basis over 30 weeks. (Variation per sampling point set for each depth showed a variation of less than 1°C).

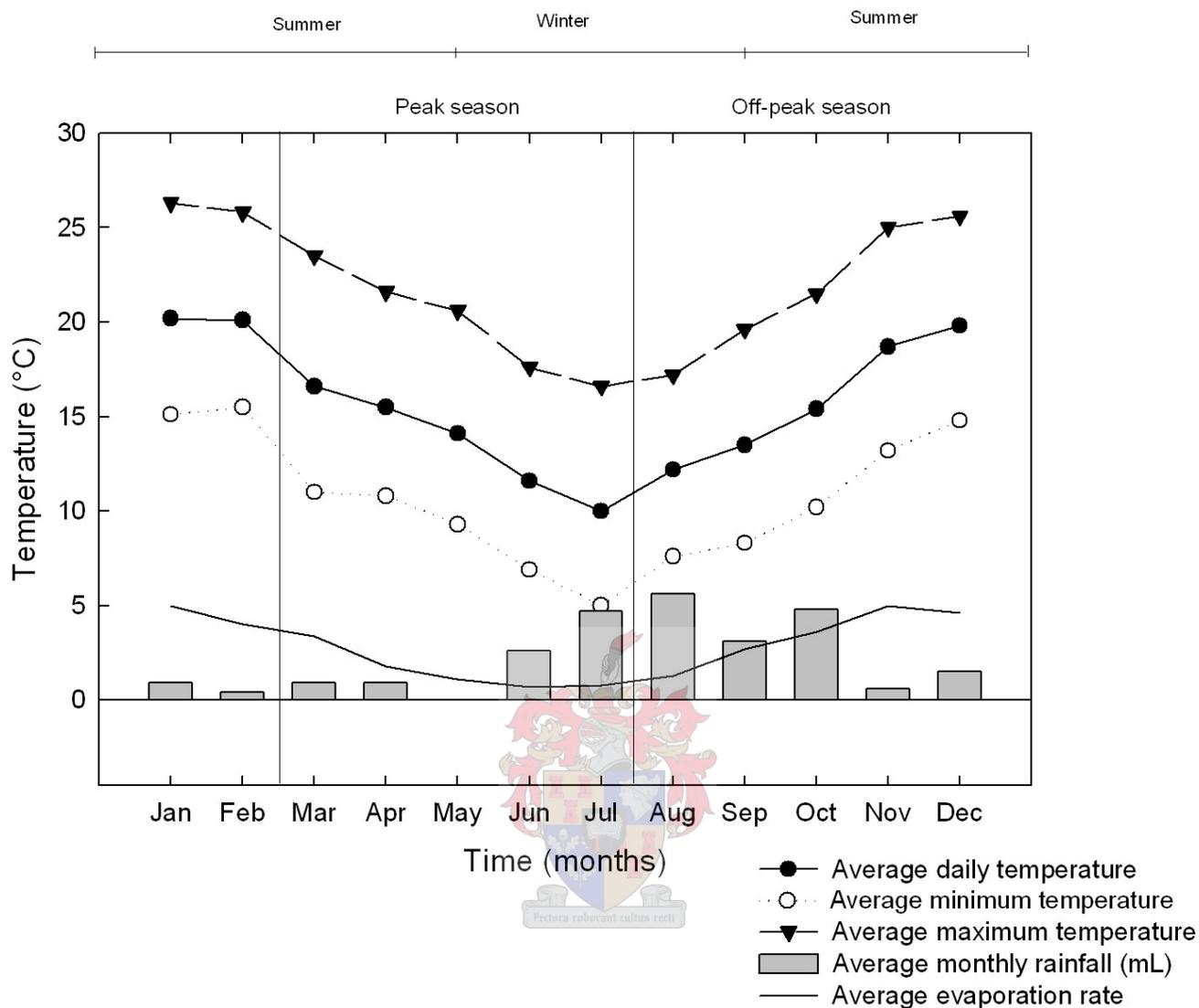


Figure 3.8 Daily minimum, maximum and average temperatures and monthly rainfall during 2004 at Applethwaite Farm where Pond A is situated (D van Zyl, Applethwaite Farm-Electronic Weather Station, Grabouw, South Africa, personal communication, 2005).

occurs at such low temperatures (Chen *et al.*, 1980; Pescod, 1996; Bitton, 1999; Bouallagui *et al.*, 2004; Taconi, 2004). Another factor that must be considered when comparing COD reduction efficiencies in the case of Pond A is that during the off-peak season wastewater with lower COD concentrations are produced which will lead to lower biogas production volume and rates. It must be remembered that these conditions lead to a slower anaerobic digestion rate, which may result in pond failure or even upsets if shock-loading should suddenly occur (Pescod, 1996; Bouallagui *et al.*, 2004).

Settled sludge volume

The average settled sludge volumes (as mL.L⁻¹)(Fig. 3.9), as determined using APHA (1998) methods, of Pond A at the 1 and 3 m depths were measured and used as an indication of microbial activity potential. The average settled sludge volume at 1 m (Fig. 3.9 A) varied remarkably between the peak season and off-peak season. An average settled sludge volume of 400 mL.L⁻¹ during the peak season was obtained, while an average settled sludge volume of 35 mL.L⁻¹ was obtained during the off-peak season.

The average settled sludge volume at the 3 m depth (Fig. 3.9 B) indicated overall higher settled sludge volumes for both the peak and off-peak seasons, than the average settled sludge volumes at 1 m depth. The average settled sludge volume obtained during the peak season was 700 mL.L⁻¹, while an average settled sludge volume of 680 mL.L⁻¹ was obtained during the off-peak season.

The decrease in settled sludge volume at 1 m during the winter months (off-peak season) may probably be attributed to a combination of factors, such as lower temperatures and decreased COD concentrations in the feed wastewater. These influenced the microbial consortium activity and production of biogas and biomass. The decrease in COD concentration together with the decrease temperature lead to lower biogas production due to a lower microbial activity and the decrease in biogas production led to less mixing and therefore a decrease in the suspended sludge bed.

The relatively small variations in the average settled sludge volume at 3 m (Fig. 3.9 B) between the peak and off-peak season can be ascribed to samples being taken nearer to the permanent sludge bed. This is due to the total depth of the pond being 4 m with a settled sludge layer of approximately 1 m. The fluctuation of the sludge volume at 3 m can be ascribed to flow volumes and strength of the incoming wastewater, activity of the microbial populations as well as gas production.

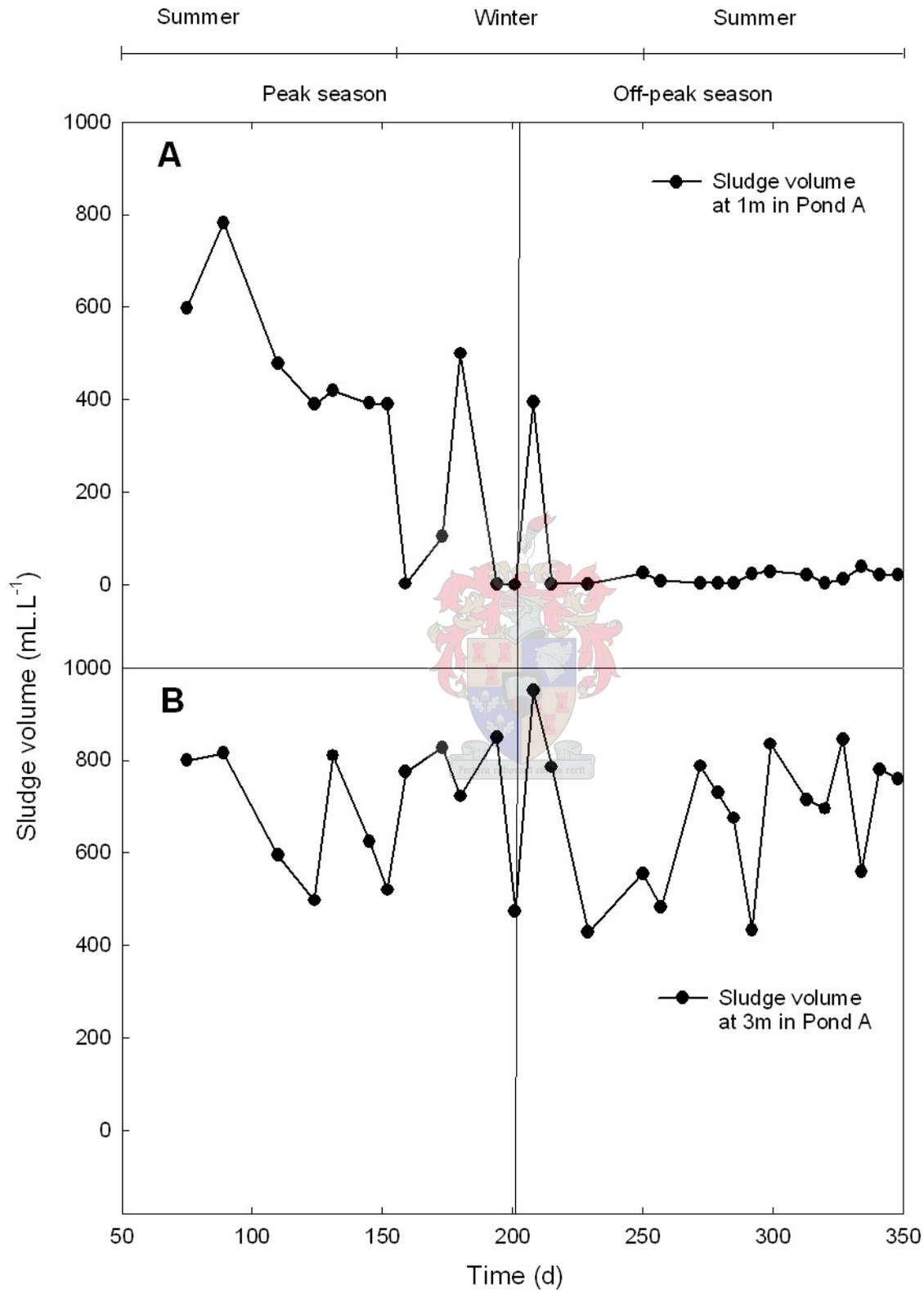


Figure 3.9 The average sludge volume in Pond A at depths of 1 m (A) and 3 m (B) during the peak and off-peak season.

Volatile Fatty Acids

The total VFA content of Pond A at 1 m depth ranged from 0 to 740 mg.L⁻¹ during the peak season and from 0 to 300 mg.L⁻¹ during the off-peak season. At the 3 m depth the total VFA content ranged from 34 to 914 mg.L⁻¹ during the peak season and from 0 to 340 mg.L⁻¹ during the off-peak season. The prevalent VFA at both the 1 and 3 m depths was acetic acid followed by propionic acid. The other VFA's, iso-butyric, butyric, iso-valeric and valeric acid, were present at very low concentrations throughout the peak and off-peak seasons. The organic acid concentration in an anaerobic system is indicative of the digestion efficiency (Griessel, 2002). It has been suggested that the organic acid concentration should be below 2 000 mg.L⁻¹ of acetate for efficient digestion to take place as higher levels have been shown to be toxic (Chen *et al.*, 1980; Jetten, 1992; Bitton, 1999; Mata-Alvarez, 2003). In the literature it has also been reported that acute methanogenic toxicity occurs at unionised volatile acid concentrations of between 10 – 60 mg.L⁻¹ as acetic acid. The unionised forms are usually found at pH values below 6.3 (Chen *et al.*, 1980; Duarte & Anderson, 1982; Mata-Alvarez, 2003) and in the case of Pond A with the relatively high pH, this should not be a problem.

Mineral analysis

The sodium-ion (Na⁺) content of the incoming wastewater (Fig. 3.10 A) ranged from 25 to 125 mg.L⁻¹ and the varying levels were ascribed to the fact that a caustic solution (NaOH) was used during factory cleaning regimes. The apples that are processed are also a natural source of Na⁺ (AIJN, 2001) but the Na⁺ concentration present in the pond (Fig. 3.10 B) is believed to be mainly due to the NaOH cleaning solution used throughout the year. Therefore, it was expected that the average Na⁺ concentration in Pond A would be higher during the peak season (147 mg.L⁻¹) due to more cleaning applications. The decrease in the average Na⁺ concentration (88 mg.L⁻¹) during the off-peak season probably was due to a lower frequency of NaOH applications during the off-peak season. The overall Na⁺ concentration in Pond

A should have no inhibitory effects on the anaerobic system, as low concentrations persist throughout the year (Rinzema *et al.*, 1998). However, high Na⁺ concentrations in excess of 2 000 mg.L⁻¹ are known to pose a problem. High concentrations may cause acute toxicity to the microbial species responsible for the anaerobic digestion process, but may also have

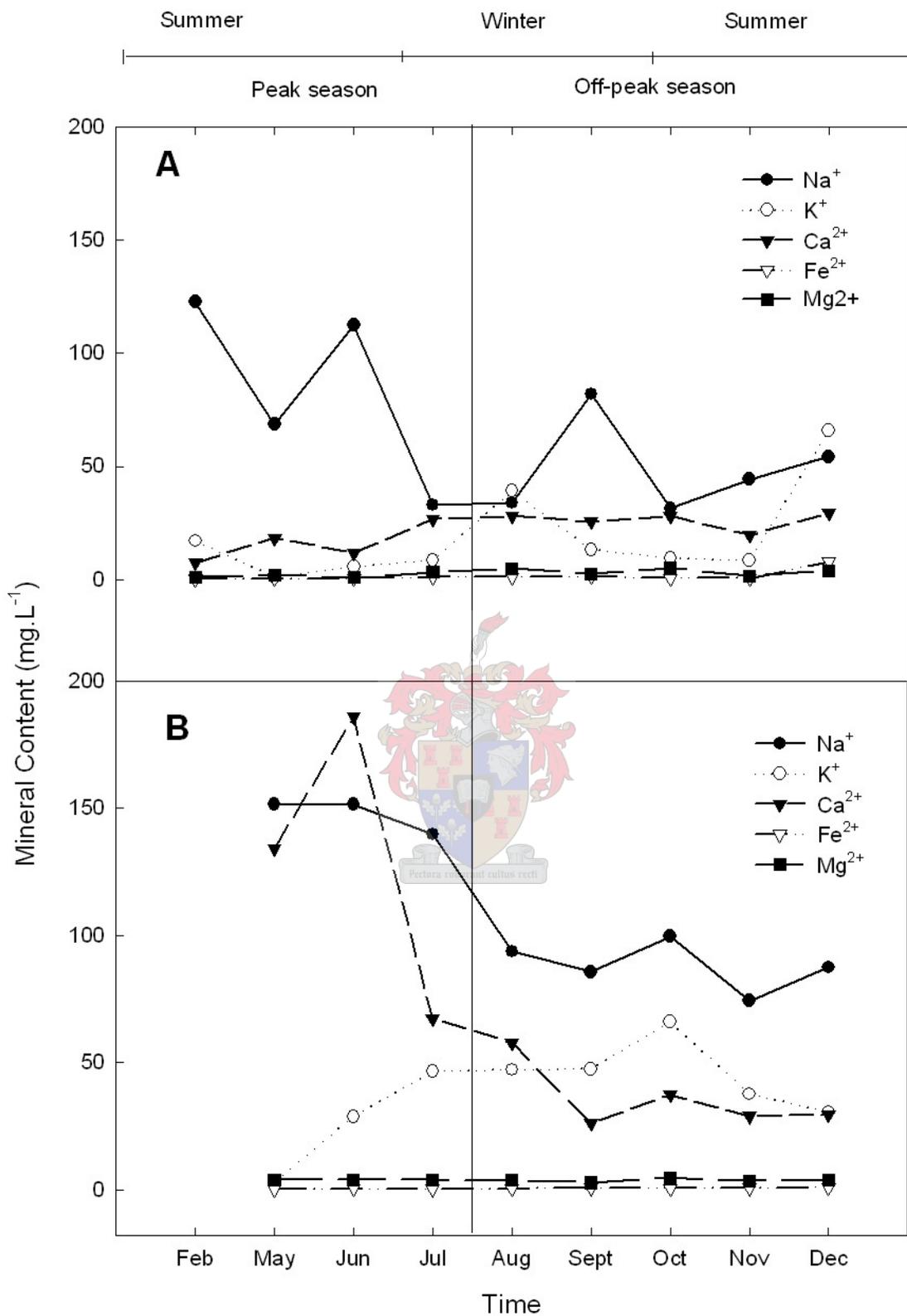


Figure 3.10 The mineral concentration of the incoming wastewater (A) and that of Pond A (B) during peak and off-peak season.

detrimental environmental impacts if it should leach into surrounding agricultural land where it may cause salinisation of groundwater (Droste, 1997; Mata-Alvarez, 2003).

The pond potassium-ion (K^+) content does not reflect the incoming wastewater K^+ content ($1.1 - 66 \text{ mg.L}^{-1}$) (Fig. 3.10 A). The pond (Fig. 3.10 B) has a higher K^+ content for most part of the year, than the incoming levels. An average K^+ concentration of 26 mg.L^{-1} was found in Pond A during the peak season and 45 mg.L^{-1} during the off-peak season. In general potassium helps as an antagonising agent to sodium toxicity (Droste, 1997). However, the concentrations obtained in Pond A were too low to have any significant effects on the performance of the anaerobic pond system.

The calcium (Ca^{2+}) content in Pond A (Fig. 3.10 B) was dependent on the lime (CaO), which was added to assist the buffer capacity of the anaerobic pond. It was assumed that the Ca^{2+} in the pond is derived from the CaO . This assumption is made because the trend of the Ca^{2+} curve in Fig. 3.10 B resembled the total monthly CaO usage-curve as shown in Fig. 3.6 A. It is also well known that the presence of calcium is beneficial to the digestion process.

The magnesium (Mg^{2+}) and iron (Fe^{2+}) contents for both the incoming wastewater and Pond A were the lowest found for all the minerals. This was expected and was correlated to the fact that processed fruit are not a natural source of these specific minerals. The specific concentrations of both Mg^{2+} and Fe^{2+} in Pond A were very low, and therefore it is believed that no significant inhibitory or stimulatory effects may be contributed to the presence of these two minerals (Mata-Alvarez, 2003).

The overall mineral content of Pond A (Fig. 3.10 B) was low and at a satisfactory level. In the literature it has been shown that minerals may have a stimulatory effect on the anaerobic digestion at low concentrations but may be inhibitory to the microbial populations if too high concentrations are present (Chen *et al.*, 1980; Bitton, 1999; Mata-Alvarez, 2003).

COD, total Kjeldahl nitrogen (TKN) and total phosphorous (P)

The average monthly COD concentration of the incoming wastewater was found to vary between $2\,900$ and $8\,550 \text{ mg.L}^{-1}$ throughout the year (Fig. 3.11). The data showed that the lower values were found during the off-peak season as a result of the decrease in the incoming wastewater COD concentration during the off-peak season when there was a decrease in fruit processing activity. The nitrogen and phosphorous concentrations, ranged from 3.0 to 15.0 mg.L^{-1} and 0.09 to 5.2 mg.L^{-1} for the peak and off-peak seasons. However,

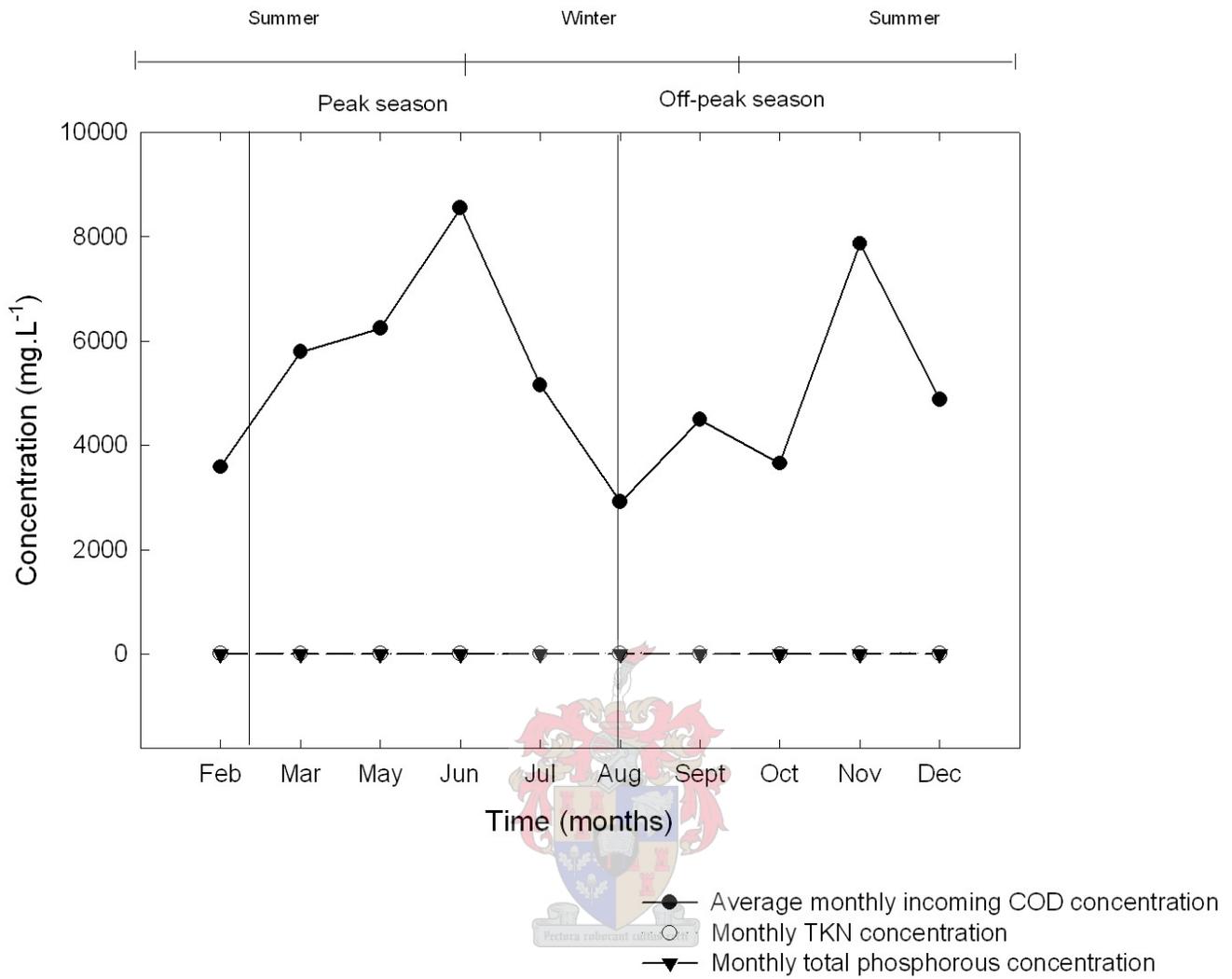


Figure 3.11 The average monthly COD concentration, total monthly Kjeldahl nitrogen and total monthly phosphorous concentration for the incoming wastewater of Pond A.

these values varied throughout the year regardless if it was peak or off-peak season (Fig. 3.11).

The C:N:P ratio (for this study, COD was taken as equivalent to the carbon value) efficient anaerobic digestion is generally recommended as 700:5:1 (Bitton, 1999; Mata-Alvarez, 2003). These authors also suggested that for optimal gas production the C:N ratio should at least be in the range of 25 – 30:1. In this study on Pond A the average incoming wastewater C:N:P ratio was 1000:2:0.5. The incoming wastewater C:N ratio however, was 1000:2, which would appear to vary slightly from the literature recommendation. The data shows that the incoming wastewater may have a slight deficiency regarding nitrogen. However, nutrient supplementation of the incoming wastewater is not recommended for the following reasons: The average nitrogen concentration in Pond A was found to be 3 400 mg.L⁻¹. This led to the assumption that nitrogen accumulation may occur in Pond A due to poor nutrient removal. It can also be assumed that nutrient removal, especially N and P occurs late in the pond treatment system, most probably in the maturation pond where the most suitable conditions prevail (Pearson, 1996).

Processing Profile

The processing profile for the year 2004 (Figs. 3.12 and 3.13) consisted of the total monthly fruit processed, total monthly water usage and total monthly wastewater generated. It was clear that these factors are inter-linked and that during the peak season the water usage in the processing plant is at its' highest and correspondingly the wastewater generated, is the highest. These processing and environmental conditions during the peak season are favourable for the operation of the wastewater treatment system and particularly the anaerobic Pond A. The favourable pond temperature during the peak season and the high COD loads (due to processing of an average of 15 000 tons of fruit a month) contribute to the pond activity, which leads to good COD degradation (91%) and subsequent biogas and CH₄ production. In contrast to the peak season, a lower mass (700 – 2 000 tons per month) of fruit is processed, during the off-peak season. These consist mostly of fruit from cold storage. Therefore, a much lower volume of wastewater is generated and this together with the non-favourable environmental conditions, create a more unfavourable environment for the anaerobic pond microbial consortium. The lower available COD loads (Figs. 3.3 and 3.11), but more importantly, the decrease in pond temperature to 10° - 15°C (Fig. 3.7) influences the microbial activity in such a way that there is a strong decline in activity as the microbial

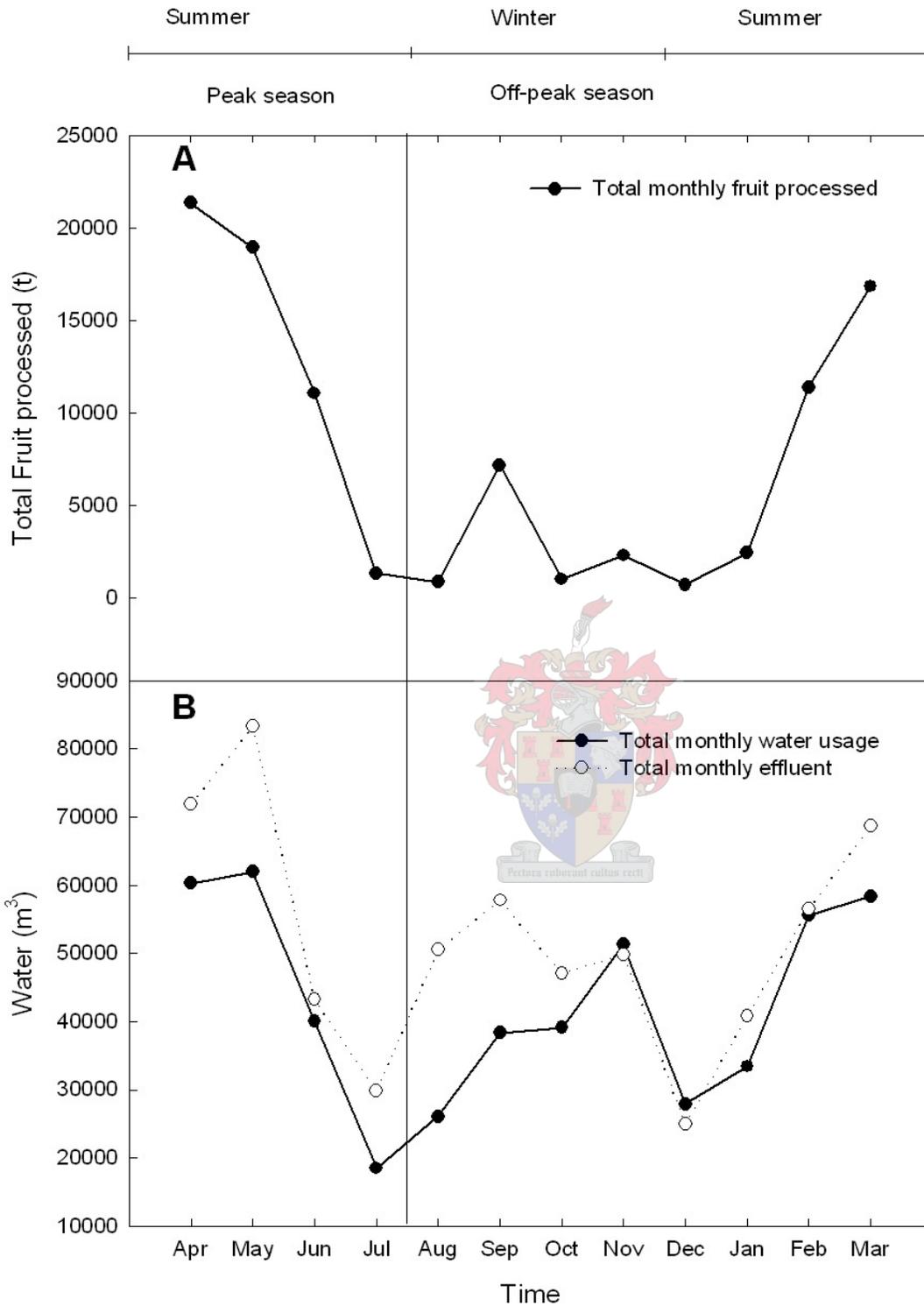


Figure 3.12 Total fruit processed (A), monthly water usage (B) and wastewater generation (B) during the peak and off-peak season during the year 2004.

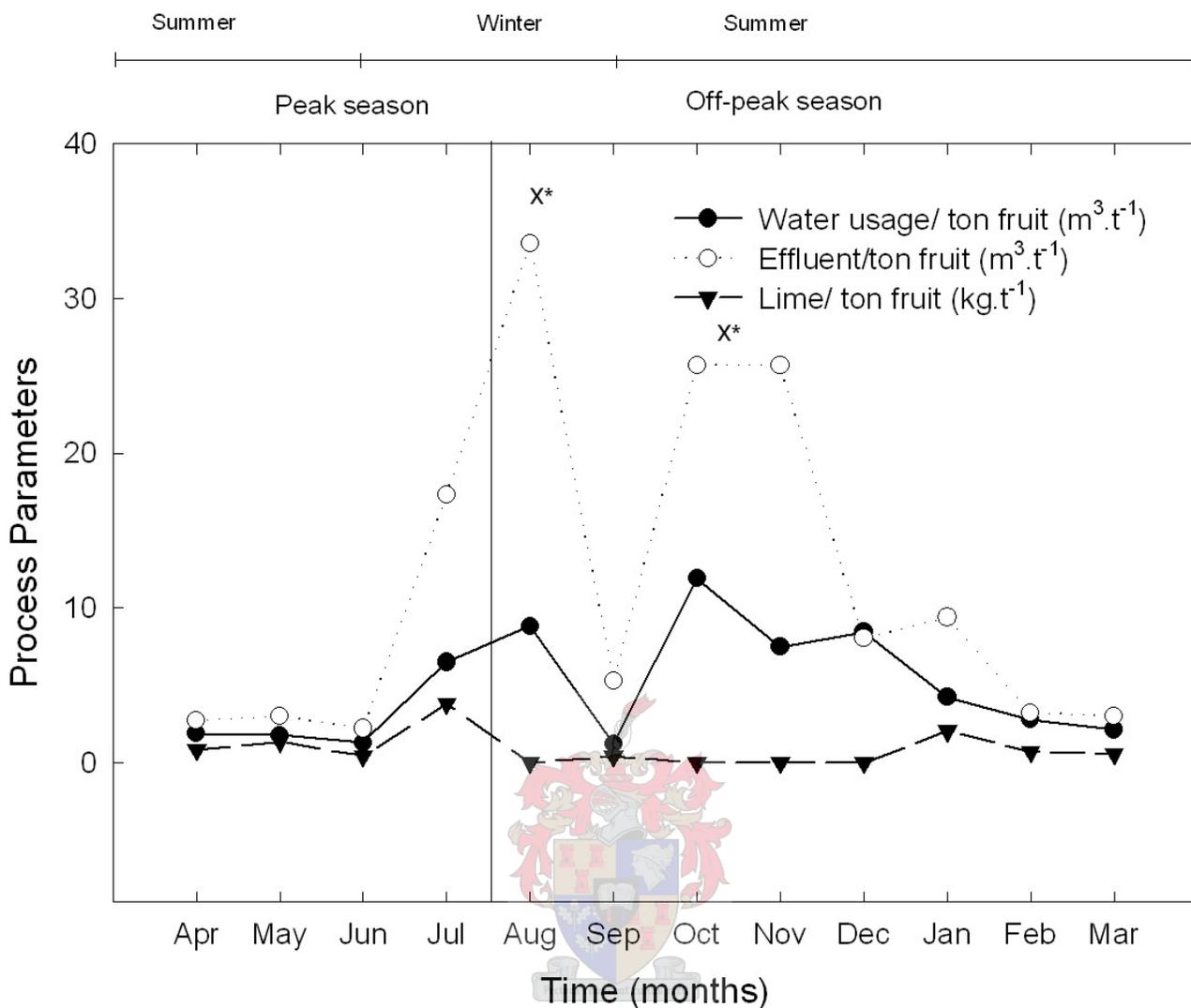


Figure 3.13 The processing profile, consisting of water usage ($m^3.t^{-1}$), wastewater generated ($m^3.t^{-1}$) and lime usage ($kg.t^{-1}$) per ton fruit processed, for both the peak and off-peak seasons during 2004. X* = High effluent volumes attributed to “at end of season” cleaning and maintenance (values taken from “Service&Utilities” report Muller 2004).

populations become dormant (Fig. 3.9). Thus much less COD is metabolised and converted to biogas and this negatively affects the degradation of organic material in the pond. However, this trend is reversed during the peak season when higher COD concentrations and more favourable pond temperatures of around 25°C (Fig. 3.7) are found. The microbial consortium then becomes active and degrades organic material and produces biogas (Taconi, 2004). This confirms that specific sets of environmental conditions, such as higher COD loads and higher or more optimum temperature ranges need to be maintained in order to achieve optimum microbial activity, which will subsequently lead to optimum wastewater treatment efficiencies and optimum energy recovery in the form of CH₄.

Conclusion

The system profile established with the monitoring of various process parameters clearly shows that the pond system is impacted by several processing factors, the seasonal nature of fruit processing and environmental factors. During the peak season high COD concentrations and higher temperatures (25° – 30°C) persist and this favours the anaerobic digestion process. Contrary, during the off-peak season lower COD concentrations and lower temperatures (below 15°C) are experienced, which are more unfavourable to the anaerobic treatment process.

Nevertheless, stable pH values, sufficient nutrient availability and alkalinity are maintained throughout the year. Furthermore, the data showed that the pond treatment system is self-maintaining and does not require nutrient supplementation. The data also showed that the treatment process is stable during the off-peak season and that the system has the ability to recover easily and operate at its optimum at the onset of the peak season.

It was however clear from the study that the most limiting factor during the off-peak season appeared to be the pond temperature. It is therefore recommended that investigations should be done to try and improve heat retention in Pond A during the off-peak season so as to maintain treatment efficiencies and improve biogas production. Thus, during this study it was found that the pond system used to treat the fruit factory wastewater was sufficient for its current application as a wastewater treatment system.

Future work is also needed to assess the conditions that exist during the off-peak season in order to overcome the limitations that exist due to environmental and processing influences and to improve biogas production. This is needed to make biogas recovery viable throughout the year and not only during the peak season when favourable digestion

conditions do exist. Therefore, the next step would be to evaluate the anaerobic ponds' sludge activity for methane generation potential under the influence of environmental factors such as decreased temperatures.

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CHAPTER 4

ASSESSMENT OF SLUDGE ACTIVITY AND BIOGAS POTENTIAL FROM AN ANAEROBIC POND TREATING FRUIT-PROCESSING WASTEWATER

Summary

The sludge activity of an anaerobic pond treating fruit-processing wastewater was evaluated to determine the effect of the apple-processing peak and off-peak season on the specific activity of the acidogenic and methanogenic populations within the sludge. An activity test using four different test media was used during the activity assays. Sludge samples were taken at four different sampling positions across the sludge bed. The sludge was also subjected to a biogas production study, which simulated pond conditions on laboratory scale. This was done in order to evaluate the biogas production potential of the anaerobic pond during actual conditions experienced by the pond throughout the year.

The results of the activity tests were expressed as the cumulative biogas volume and composition over 25 h. The cumulative biogas volume and total CH₄ composition showed little or no difference between the four sludge samples. The biogas formation rate (S_B) and methane production rate (S_M) were determined during the biogas formation study. A significant difference was found between the activity of the microbial populations during the peak and off-peak seasons. The overall trend in S_B and S_M values showed an increased activity during peak season and a decreased activity during off-peak season. During the biogas formation tests after an incubation period of 72 hours, the best biogas productions and COD reductions were achieved at 25°C incubation. From the theoretical CH₄ calculations and estimates based on the results obtained during the biogas formation tests as well as the Ponds' true COD reduction values, it was clear that CH₄ recovery from the anaerobic pond is definitely a worthwhile consideration.

Introduction

In an anaerobic pond system organic waste may be converted to carbon dioxide (CO₂), methane (CH₄) other gaseous end-products, organic acids and cell biomass during the anaerobic degradation process. The CH₄ recovered from pond systems is normally of a good quality and not only represents energy recovery but also prevents the release of CH₄ into the environment (Pearson, 1996; Pescod, 1996). Methane recovery from anaerobic ponds can significantly contribute to bio-energy (Chynoweth *et al.*, 2001). Furthermore in terms of pollution control, carbon conversion efficiencies in pond systems have been reported to range from 75 to 85% when operating at optimal conditions (Drysdale, 1981; Pearson, 1996; Pescod 1996; Bitton, 1999).

There are several environmental factors that influence the operation of the anaerobic digestion process and these include chemical oxygen demands (COD), pH, alkalinity, temperature, hydraulic retention times, nutrient availability and the presence of volatile fatty acids. These factors will ultimately influence the biogas production, which in turn affects the treatment efficiency of the pond system. Therefore, the rate of biogas production may be considered to be one of the most important indicators of operational performance as CH₄ production is the last step during the anaerobic conversion process (Petrozzi *et al.*, 1992). The specific activity, quality and quantity of the biomass involved influence the biogas production rate and thus, activity testing of the biomass is necessary to get an indication of the effectiveness of the wastewater treatment process.

Activity has been defined as the substrate dependent biogas or methane production rate per unit mass of volatile solids of the biomass (O'Kennedy, 2000). This generally involves the measurement of biogas produced from a biomass system (continuous or batch) containing a specific substrate (Sørensen & Ahring, 1993; Lamb, 1995). Subsequently, the total activity of a given sludge sample can be measured to give an indication of overall activity. The individual activity value gives information with regards to the basic stages in the digestion process and can be used to reveal the dynamics between the different bacterial species (Soto *et al.*, 1993).

Activity testing of biomass has been shown to be of value when monitoring the activity of an individual population in methanogenic environments, characterisation of the microbial composition and optimising the start-up of a new digester (Dolfing & Bloemen, 1985). It

also assists when studying the potential toxicity of certain substances on the biomass (Soto *et al.*, 1993). For energy recovery purposes the most important characteristic of activity testing is that the specific CH₄ yield can be determined by means of preliminary methanogenic activity tests (Zábranská *et al.*, 1994).

The aim of this study was to determine the effect of the fruit processing seasons (peak and off-peak season) on the specific activity of the acidogenic and methanogenic populations in the sludge of an anaerobic pond system. The biogas production potential at various temperatures was also assessed to determine the viability of CH₄ recovery.

Material and methods

Sampling

Sludge samples were obtained from an anaerobic pond (Pond A) of a local apple juice manufacturer situated in Grabouw, South Africa. The specific anaerobic pond had a volume of 6 600 m³ and a depth of 4 m. The sludge samples (1 L) were taken monthly (weather permitting) and were collected with a submersible pump from the anaerobic pond. Thus a total of 11 sampling sets were successfully taken over the course of the year. The samples were taken at four sampling positions: at 30 m; 26 m; 24 m and 14 m from the side of the pond and at a depth of 3 m from the surface. The four positions were colour coded as green, red, yellow and blue to ensure identification.

The sludge samples (taken monthly) were subjected to a series of direct activity assays. These assays were done to determine the microbial activity over the course of the year in terms of cumulative biogas volumes and also the biogas and methane production rates (S_B and S_M). The direct activity assays involved four different test media: control (BTM); glucose (GTM); fructose (FTM); and acetic acid (ATM), to determine the activity of each individual microbial population (Table 4.1).

Biogas formation studies were used to simulate the actual conditions in the pond, such as hydraulic retention time (HRT), temperature and sludge:wastewater ratio. This was done to quantify the potential biogas production of the sludge under controlled but simulated (in terms of temperature, pH, COD concentration) conditions.

Table 4.1 Composition of the control activity assay (Basic Test Medium - BTM) (O’Kennedy, 2000).

Compound*	Concentration (g.L ⁻¹)
Glucose	2.0
K ₂ HPO ₄	1.0
KH ₂ PO ₄	2.6
Urea	1.1
NH ₄ Cl	1.0
Na ₂ S	0.1
MgCl ₂ .6H ₂ O	0.1
Yeast Extract	0.2
pH	7.1

* All chemicals used were obtained from Merck and were of analytical grade.

Direct activity assays

The activity assays were done to determine the microbial activity in terms of cumulative biogas volumes as well as biogas and CH₄ production rates (S_B and S_M). Activity assays were done using the activity method of O'Kennedy (2000). Study trials were performed and monitoring of process parameters (as discussed in Chapter 3) occurred during the off-peak season (day 1 – 53). Peak season started on day 53, however, sampling problems were experienced and it was only possible to start with the activity assays after day 140.

As part of the preparation for the activity assays, activation of the monthly sludge samples from each of the four sampling positions were performed in an activation medium, which consisted of glucose, KHPO₄ and urea (NH₄CO). The sludge samples and the activity media were mixed (ratio of 1:2) and incubated at 35°C for 24 h. The activation medium with the added sludge was decanted every 24 h and replenished. After a 48 h activation period, the samples were centrifuged at 5000 × g for 10 min. This was done to ensure that a fairly water-free sludge sample was obtained. Four different test media were used during the assays and included a control (Basic test medium - BTM) (Table 4.1), glucose (GTM), fructose (FTM) and an acetic acid test medium (ATM) (Table 4.2).

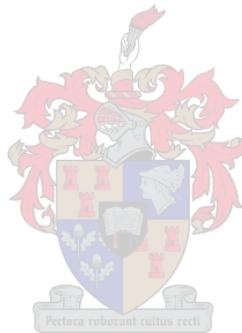
The BTM was used to measure the general baseline activity of the sludge, while the GTM and FTM media were used to measure the specific glucose and fructose utilisation activity of the acidogens (Table 4.2). The ATM medium was used to measure the specific activity of the acetoclastic methanogens (Table 4.2).

For the direct activity assays sludge inoculums (3 g) (all tests in duplicate) from Pond A were placed in 20 mL vials, to which 13 mL of the relevant test medium was added, leaving 6 mL headspace. The vials were then sealed with butyl septa and capped with aluminium caps and incubated at 35°C, after which the biogas volume was measured using a free moving gas-tight 10 mL syringe with a 12-gauge needle. The needle was inserted through the butyl septa and the plunger was allowed to move freely until equilibrium had been reached. The volume of biogas was measured after 5, 10 and 25 h.

The biogas composition (%CH₄ and %CO₂) was determined using gas chromatography. A gas sample (1.0 mL) was injected into a Fisons GC equipped with a thermal conductivity detector and 2.0 m x 3.0 mm i.d. column packed with Porapak Q (Waters Ass. Inc, Milford, MA), 80/100 mesh. The oven temperature was set at 55°C and

Table 4.2 Composition of the different test media used to determine activity of specific microbial groups (O’Kennedy, 2000).

Test medium	Activity Indicator Groups
Only BTM*	Control
GTM = BTM + 2 g.L ⁻¹ Glucose	Acidogens
FTM = BTM + 2 g.L ⁻¹ Fructose	Acidogens
ATM = BTM + 1g.L ⁻¹ Acetic acid	Acetoclastic Methanogens



* The Basic Test Medium was used as basic component for each medium – composition given in Table 4.1.

helium was used as carrier gas at a flow rate of $30 \text{ mL}\cdot\text{min}^{-1}$ (Lamb, 1995; Ronquest, 1999).

Biogas formation test

This test was designed to simulate, under lab-conditions, the environmental conditions experienced during the off-peak season of the apple processing plants' Pond A (see Chapter 3 of this thesis). This was done in order to determine especially the influence of the non-optimal lower temperatures on biogas production. In the previous section the determination of the direct activity gave an indication of the biogas and CH_4 production rates of the individual microbial populations in the sludge under optimum laboratory conditions. Therefore, with the biogas formation test, it should be possible to compare the optimum activity and biogas production potential with the actual biogas production by the microbial populations that had been exposed to non-optimum conditions.

The biogas formation test was carried out in 500 mL Schott bottles where the bottle caps had been modified to accommodate a glass tube (Fig. 4.1). The tube was sealed in the cap with silicone to ensure that a sealed system was established. A latex tube was fitted to the glass tube and secured with a cable tie and also sealed with silicone. The open end of the tube was sealed by a goose-neck-tie and secured with a cable tie.

Each test bottle was filled with 100 mL sludge and 300 mL of substrate. The substrate was prepared from wastewater collected at the fruit processing factory (Associated Fruit Processors Pty Ltd.) and supplemented with apple concentrate (± 70 °Brix) to give a COD concentration of about $5\,000 \text{ mg}\cdot\text{L}^{-1}$. When required the pH of the substrate was adjusted to between 6.2 and 6.4 with industrial lime. This was done to ensure that the average pond pH (Chapter 3 of this thesis) was simulated during the biogas formation tests. The bottles were tightly sealed with the modified cap and the contents mixed and the bottles incubated at 10° , 18° and 25°C . A free moving gas-tight 10 mL syringe with a 12-gauge needle was inserted into the latex tubing to measure the gas volume. The plunger was allowed to move freely until equilibrium had been reached. The biogas was measured after 24, 48 and 72 h, in order to determine when the most biogas was produced. A sample of the measured biogas was used to determine the biogas composition ($\%\text{CH}_4$) with gas chromatography. The COD reduction (%) was measured by

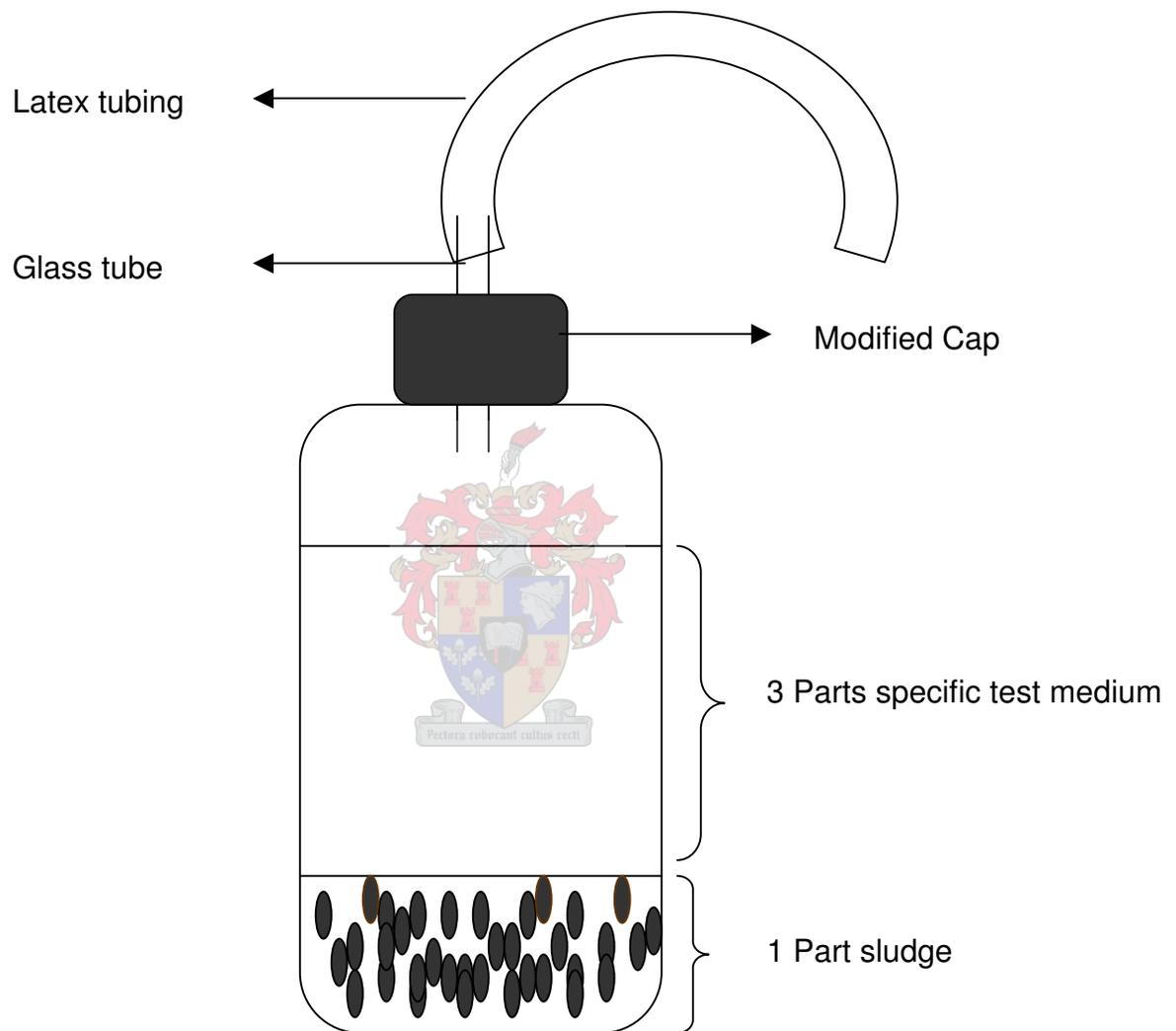


Figure 4.1 Schematic illustration of the experimental setup used in the biogas production tests.

determining the COD concentration of the substrate before incubation and after 72 h of incubation. All the tests were carried out in duplicate.

CH₄ potential of anaerobic Pond A

The Biochemical Methane Potential (BMP) for the anaerobic pond was determined using the following Q_m equation recommended by Droste (1997):

$$Q_m = Q(S_{T0} - S_{Te})M = QEMS_{T0}$$

where

Q_m = the quantity of methane produced per unit time

Q = the influent flow rate of incoming feed

S_{T0} = the total influent COD (suspended + solid)

S_{Te} = the total effluent COD (suspended + solid)

E = an efficiency factor (dimensionless, ranging from 0 to 1)

M = the volume of CH₄ produced per unit of COD removed

The BMP is generally used to determine the maximum CH₄ production potential from a wastewater (Droste, 1997). The rate of CH₄ production is related to the flow rate and COD substrate removal. The flow rate in this study for Pond A was taken as 3 000 m³.d⁻¹ (determined in Chapter 3 of this thesis), while the COD reduction value was that which was obtained in the biogas formation tests.

Results and discussion

Pond Temperature

To clarify some of the figures that will be used to illustrate data, a figure taken from Chapter 3 of this thesis has been included here. This figure (Fig. 4.2) shows Pond A's temperature profile for the year 2004.

Direct activity assays

In this study direct activity of the sludge samples from Pond A was taken as substrate dependent biogas or methane production in a standardised biomass system containing

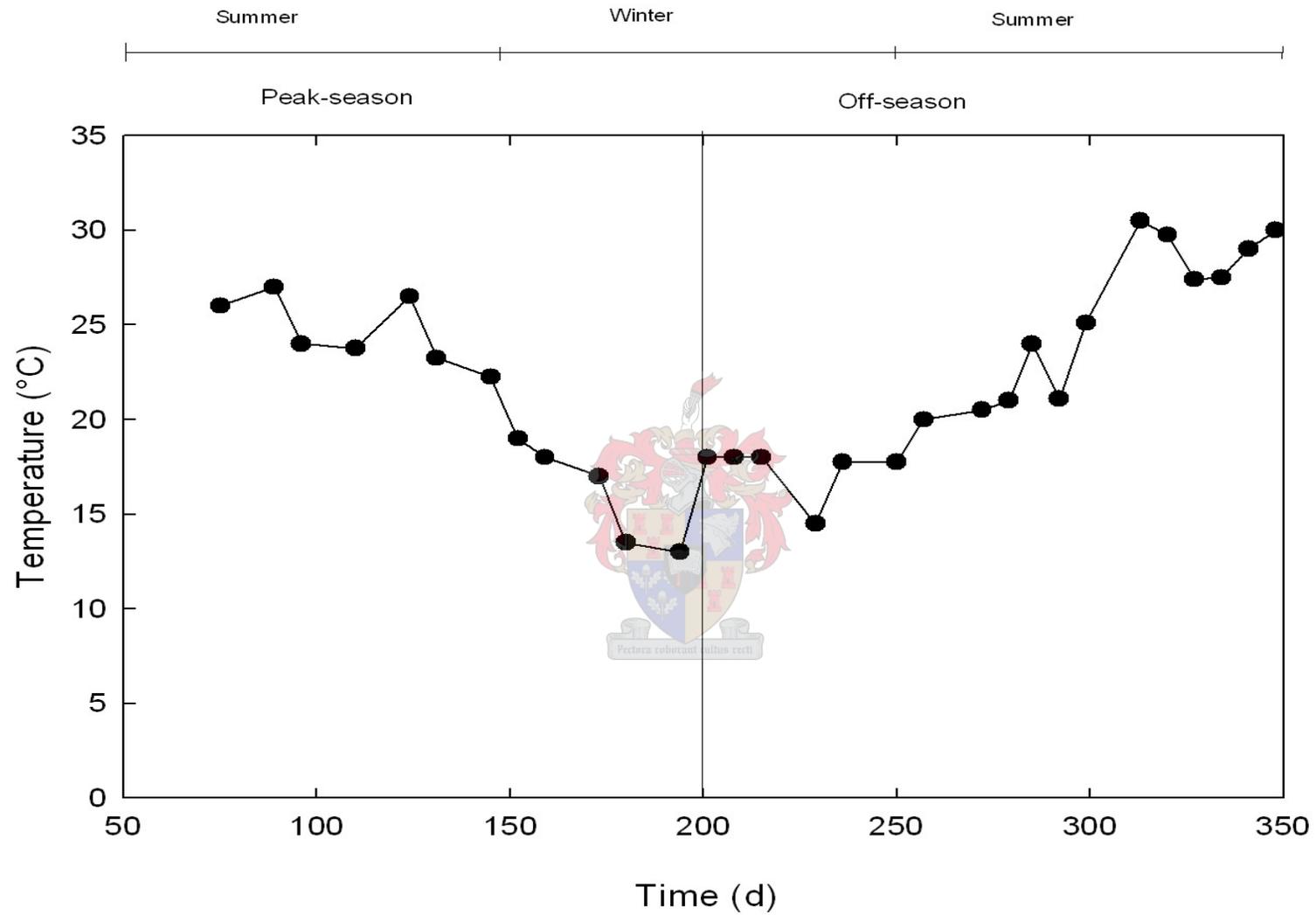


Figure 4.2 The weekly average temperature variation in Pond A during the peak and off-peak seasons.

specific substrates. The cumulative biogas and cumulative CH₄ volumes were used to indicate microbial activity as well as relate decreased activity to decreased CH₄ production.

The cumulative biogas production (after 25 h) obtained during the direct activity assays, is shown in Fig. 4.3. The activity assays indicated that the activity of the sludge was influenced by the peak season (days 50 – 173) and the off-peak season (days 200 – 271) as well as temperature changes throughout the year (discussed in Chapter 3).

Cumulative biogas volume after 25 h incubation – The cumulative biogas volume was used to indicate the total biogas produced after 25 h. This was used to compare the biogas production activity of the sludge samples.

In Fig. 4.3 A, using the monthly sludge samples from the four sampling positions, it can be seen that the biogas productions for the four sampling positions were very similar. When using the BTM medium, higher cumulative biogas volumes were obtained during the peak season (Table 4.3).

When using the GTM medium (Fig. 4.3 B), the cumulative biogas volumes obtained for the peak season were, as expected, higher than these obtained for the BTM and ranged between 8.3 – 13.6 mL, while a range of 6.3 – 11.2 mL was obtained for the off-peak season (Table 4.3).

The cumulative biogas volume attained when using the FTM medium (Fig. 4.3 C) were similar to the values for the GTM and ranged between 7.5 – 12.3 mL for the peak season and 6.7– 10.9 mL for the off-peak season. When using the ATM medium, the data in Fig. 4.3 D indicate a range of 4.8 – 9.2 mL for the peak season and 4.0 – 6.7 mL for the off-peak season (Table 4.3).

Table 4.3 Summary of cumulative biogas volume ranges.

Media	Peak Season	Off-peak season
BTM	3.9 – 7.9	3.5 – 5.6
GTM	8.3 – 13.6	6.3 – 11.2
FTM	7.5 – 12.3	6.7– 10.9
ATM	4.8 – 9.2	4.0 – 6.7

In general, the cumulative biogas ranges (Table 4.3) obtained with the four test media over the year showed only minor differences between the four sampling positions

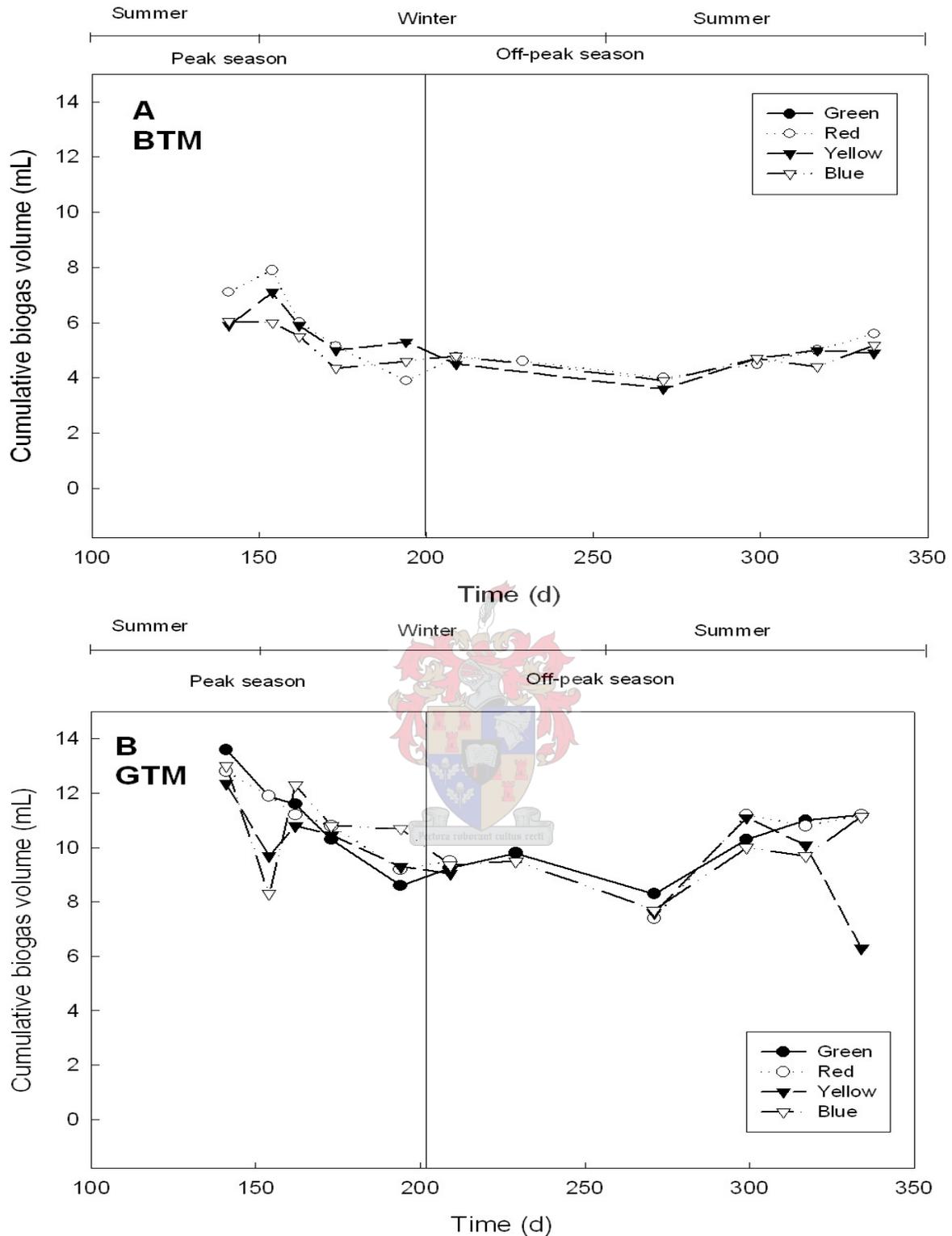


Figure 4.3. The cumulative biogas production after 25 h incubation at 35°C of the monthly sludge obtained from the four sampling positions of Pond A throughout 2004. Cumulative biogas volume was measured in four media: BTM (A), GTM (B), FTM (C) and the ATM media (D).

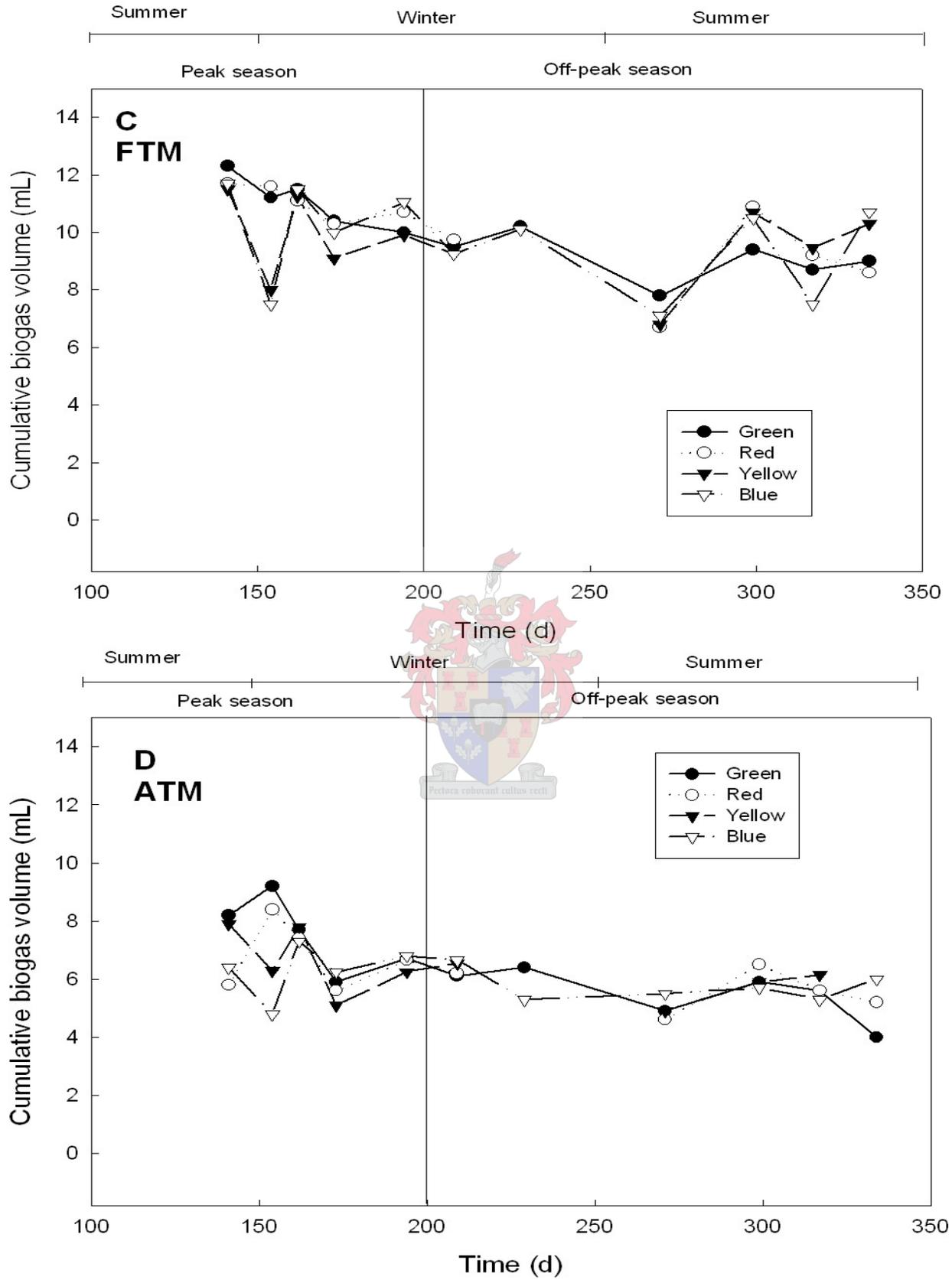


Figure 4.3 continued

(green, red, yellow and blue) were similar. Furthermore, the general trend in the data in all the graphs indicated a higher cumulative biogas volume during the peak season and a decline during the off-peak season. This was expected and was ascribed to the impact on the microbial populations by a combination of decreases in wastewater COD and lower winter temperatures (as shown in Chapter 3 and in Fig. 4.2) during the first part of the off-peak season. These factors probably impacted the microbial activity and metabolic status of the microbial populations. The results also showed that higher cumulative biogas volumes were achieved with the GTM (Fig. 4.3 B and Table 4.3), followed by the FTM medium (Fig. 4.3 C and Table 4.3). This was attributed to the metabolic activity of the microbial population, especially the acidogens, which might have been metabolically more conditioned to utilise simple sugars.

Cumulative CH₄ volume after 25 h incubation – The cumulative CH₄ volumes of the monthly sludge samples from the four sampling positions were used to determine the cumulative CH₄ produced after 25 h. This was also used to correlate CH₄ production activities of the monthly sludge samples over the peak and off-peak seasons.

The results obtained with each of the four test media showed few major differences between the four sampling positions (green, red, yellow and blue). As with the cumulative biogas volume, the general trend in all the graphs indicated higher cumulative CH₄ volumes during the peak season and a decline during the off-peak season. This was expected, and was ascribed to the decrease in COD concentrations and lower temperatures (as seen in Chapter 3) during the winter months and off-peak season (Fig. 4.2), which resulted in a decrease in microbial activity and total biogas production.

Both the data in Figs. 4.4 B and C showed a wider cumulative CH₄ volume range during the peak season with a rapid decline in the cumulative CH₄ volume towards the off-peak season (winter months) (from days 173 to 210). In all four cases (Fig. 4.4 A, B, C & D) after day 300 the variation in cumulative CH₄ volume became too large to compare data sets by eye. This variation could not be explained or be ascribed to the impact of one or even more environmental factors. The lowest CH₄ production was found (Fig. 4.4 B) during the colder winter months suggesting that temperature was the most important environmental factor that impacted the CH₄ production. This is important as it corroborates what is stated in the literature (Bryant & McInerney, 1981) confirms that the

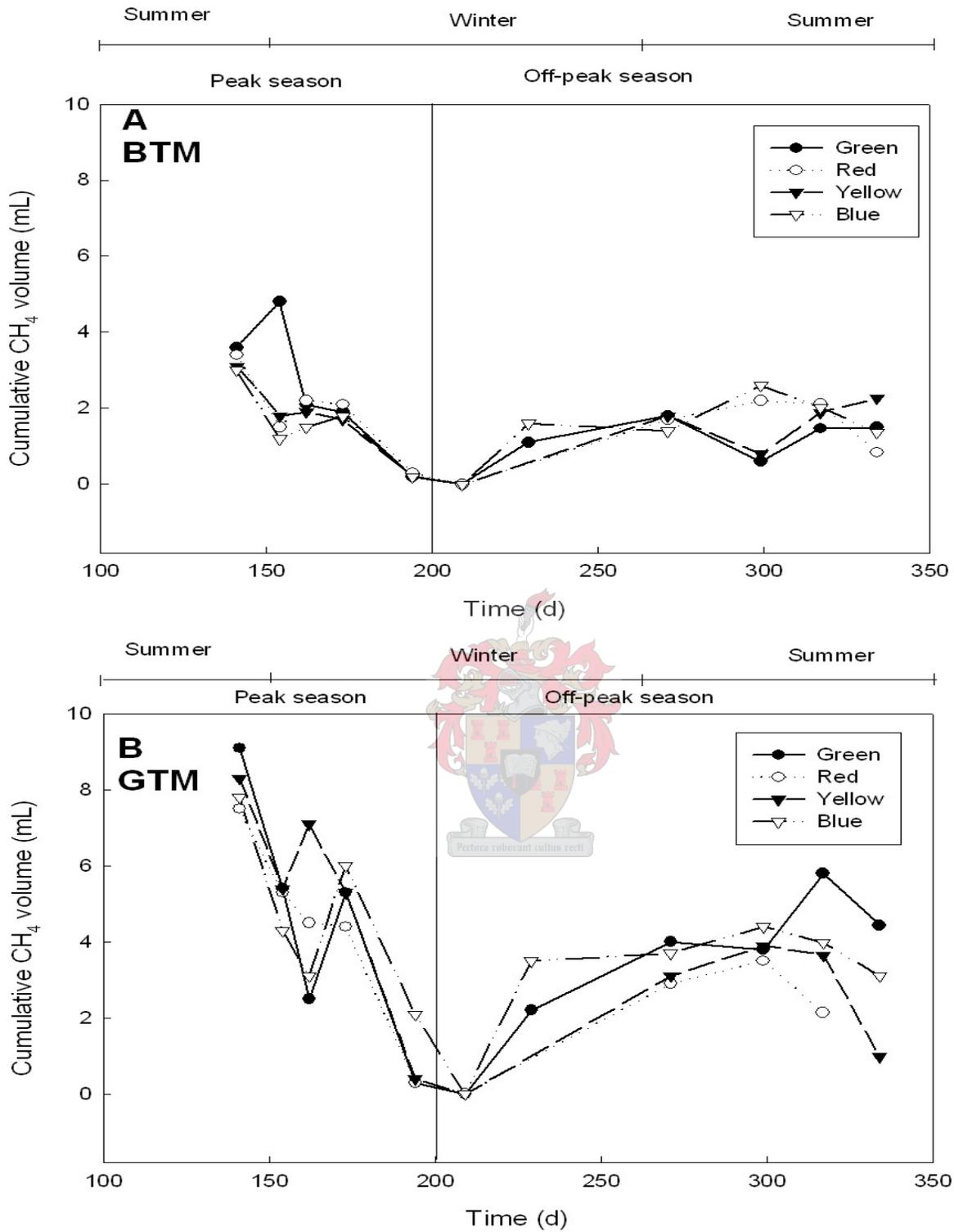


Figure 4.4 The cumulative CH₄ production after 25 h incubation at 35°C of the monthly sludge obtained from the four sampling positions of Pond A throughout 2004. Cumulative CH₄ volume was measured in four media: BTM (A), GTM (B) and FTM (C) and the ATM media (D).

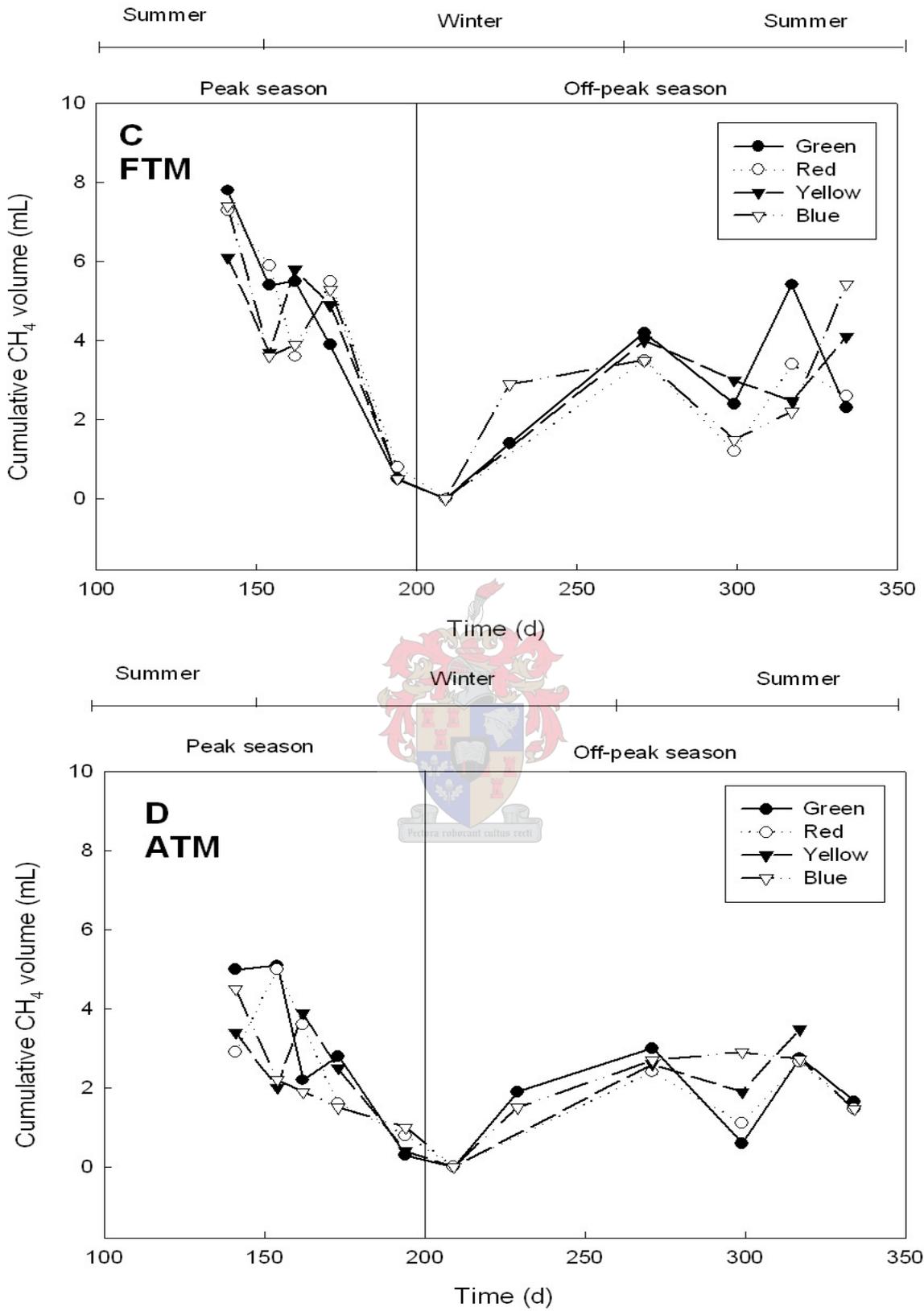


Figure 4.4 continued.

microbial species responsible for CH₄ production are more sensitive to changes in environmental parameters, such as decreases in temperature. This negative impact on CH₄ formation during the colder winter months is a factor that must be taken into consideration when the biogas potential of Pond A is to be calculated.

In Fig. 4.4 A when using the BTM, it was found that the cumulative CH₄ volumes obtained for the peak season were, as expected, lower than the total biogas produced. It was also found that the CH₄ during the peak season was higher than during the off-peak season. With the GTM and FTM (Figs. 4.4 B and C), the cumulative CH₄ volumes obtained for the peak season were much higher followed by a drastic drop during the winter months (days 173 - 210) and then by about day 225, just after the winter as the temperatures started increasing (as shown in Chapter 3), a steady increase was found.

The data in Fig. 4.4 D (ATM medium), gave the same production profile as found for the other media but the data clearly shows the CH₄ production, with a range of 0.3 – 5.1 mL for the peak season and 0.6 – 3.5 mL was lower for the off-peak season.

Biogas production rate (S_B) – The biogas production rates (S_B) were used to assess the activity of the individual microbial populations present in the sludge samples throughout the peak and off-processing seasons. Thus the S_B of a given sludge sample can also be used to compare overall activity for all media per time period (5, 10 and 25 h).

The S_B measured after 5 h incubation (Fig. 4.5 A) in the BTM was found to be higher (0.1 – 0.2 mL.h⁻¹)(see Table 4.4) during the peak season and much lower during the winter months of the off-peak season. There was however a steady increase in the S_B value after day 250. This was, as previously ascribed, a direct result of the increases in Pond temperatures after the winter months.

The S_B values with the GTM (Fig. 4.5 A and Table 4.4) ranged from 0.25 – 0.36 mL.h⁻¹ during the peak season and 0.16 – 0.24 mL.h⁻¹ during the off-peak season, with the same increase after day 250 as the ponds temperature increased. For the FTM (Fig. 4.5 A), the S_B values ranged from 0.17 – 0.31 mL.h⁻¹ for the peak season, 0.15 – 0.26 mL.h⁻¹ for the off-peak season, with the same increase after day 250. When using the ATM, the S_B values were lower than for the other media and ranged between 0.09 – 0.23 mL.h⁻¹ during the peak season and 0.09 – 0.14 mL.h⁻¹ during the off-peak season.

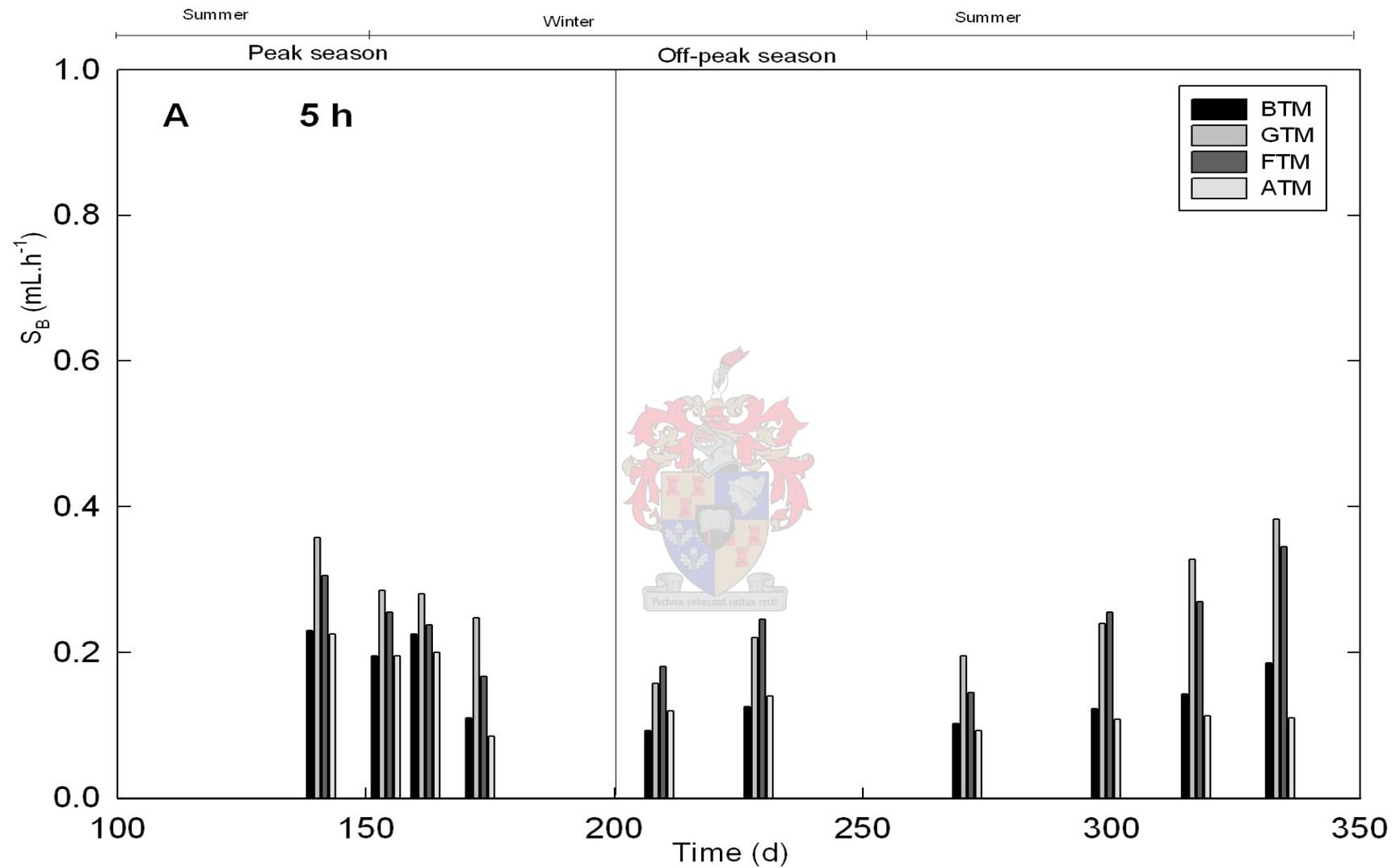


Figure 4.5 Biogas production rates (S_B) during 2004 after 5 h (A), 10 h (B) and 25 h (C) incubation during the peak and off-peak season.

The S_B values obtained after 10 h incubation (Fig. 4.5 B and Table 4.4) with the BTM ranged between 0.28 – 0.35 mL.h⁻¹ during the peak season and 0.14 – 0.24 mL.h⁻¹ during the off-peak season. The GTM S_B values ranged between 0.45 – 0.84 mL.h⁻¹ during the peak season and 0.34 – 0.64 mL.h⁻¹ during the off-peak season. For the FTM, the S_B values obtained ranged between 0.47 – 0.7 mL.h⁻¹ for the peak season, 0.33 – 0.56 mL.h⁻¹ for the off-peak season. When using ATM, the S_B values ranged between 0.28 – 0.44 mL.h⁻¹ during the peak season and 0.2 – 0.31 mL.h⁻¹ during the off-peak season. In the case of the 10 h incubation the distinctive impact of the increase in temperature after day 250 was not as clear as found with the 5 h incubation samples. This suggests that the longer incubation of 10 h period probably facilitated the activation of the metabolic pathways of the “dormant” winter sludges. This contributed to the higher biogas production rate (Fig. 4.5 B) due to the methanogenic populations having more time to metabolise specific carbon compounds.

Table 4.4 Summary of S_B value ranges.

Media	Peak Season	Off-peak season
BTM	0.1 – 0.2	0.093 – 0.185
GTM	0.25 – 0.36	0.16 – 0.24
FTM	0.17 – 0.31	0.15 – 0.26
ATM	0.09 – 0.23	0.09 – 0.14

The S_B values after 25 h of incubation (Fig. 4.5 C), for all the test media were found to be generally lower than those after 10 h incubation. No clear differences were found to correlate the peak and off-peak seasons with increases in temperatures after the winter periods. This activity stabilisation and overall decrease in activity can possibly be ascribed to exhaustion of the carbon source or possibly another growth factor. As part of the assay, the final pH was also measured and for all the samples the final pH after 25 h was never below 6.8. Thus, it was concluded that acidity was not the cause of the decrease in activity but rather the exhaustion of the carbon source.

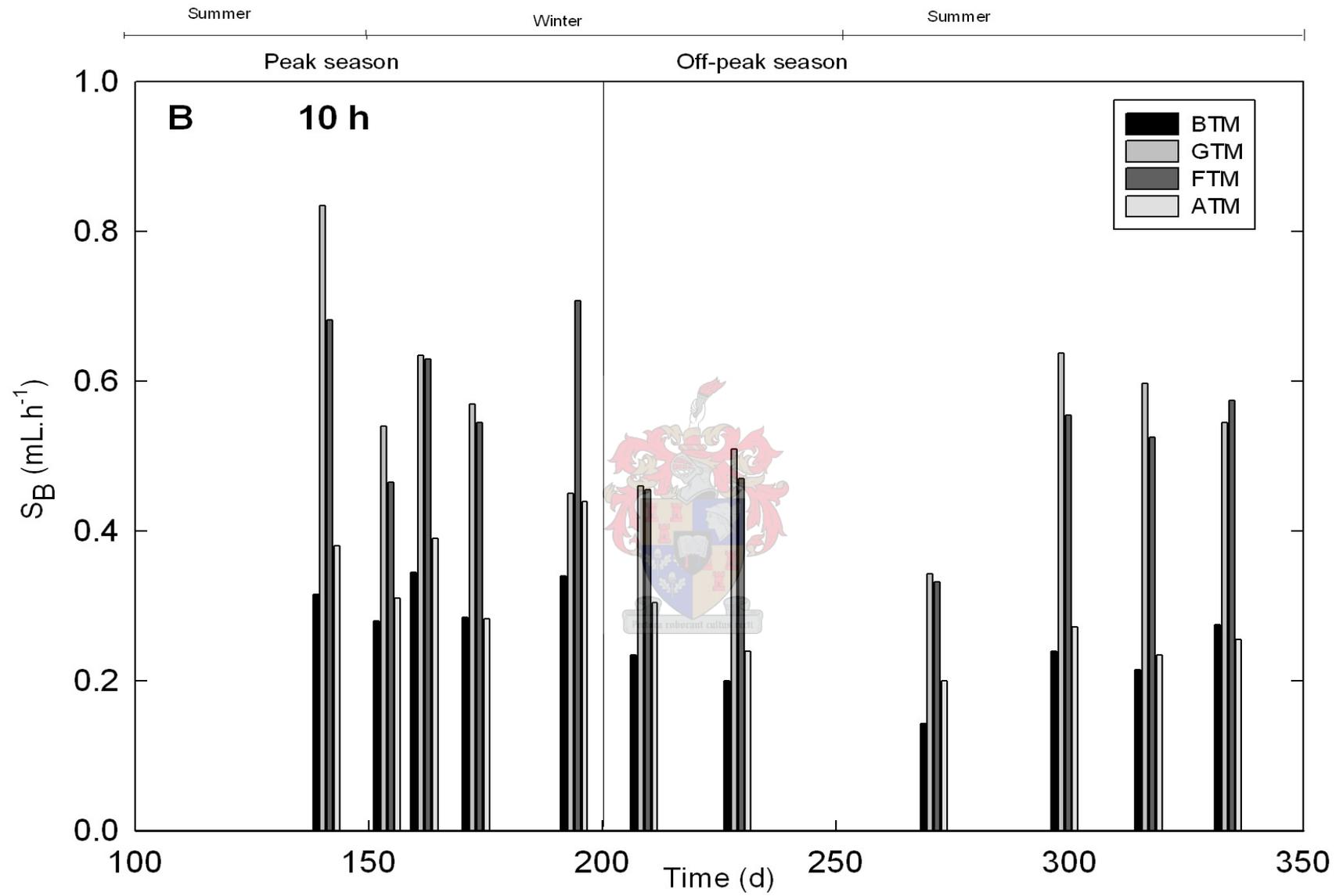


Figure 4.5 continued.

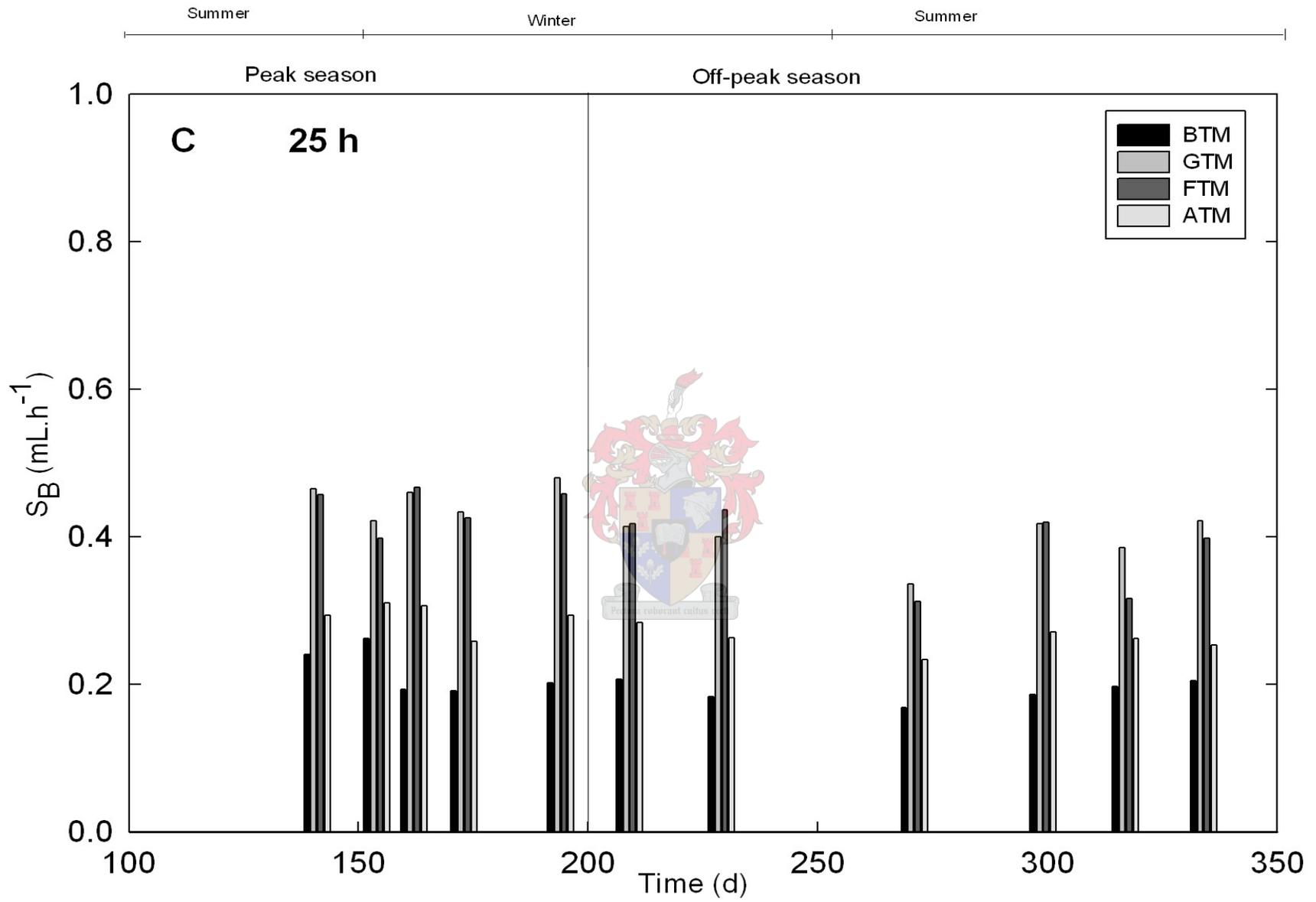


Figure 4.5 continued

Methane production rate (S_M)- The S_M rates were used to specifically evaluate the activity rates of the methane producing populations. The S_M rates after 5 h incubation (Fig. 4.6 A) for all the assay media were very low. This was expected since it is known that the methanogens are slow growers (long t_d values) taking much longer to become metabolically active. It is also known that the methanogens are very sensitive to exposure to unfavourable environmental conditions like the sampling process where contact with oxygen could have been possible leading to a decrease in activity. These 5 h results will thus not be discussed further.

The S_M values after 10 h (Fig. 4.6 B) in the BTM ranged between 0.02 – 0.13 mL.h⁻¹ during the peak season and during the off-peak season decreased to between 0.01 – 0.02 mL.h⁻¹. There was however in most cases an increase (as found with the S_B) in the S_M values after day 250 (0.02 – 0.03 mL.h⁻¹). The S_M values for the GTM ranged between 0.1 – 0.45 mL.h⁻¹ during the peak season and 0.05 – 0.34 mL.h⁻¹ during the off-peak season, while an increase in S_M was observed after day 250 (0.13 – 0.2 mL.h⁻¹). For FTM, the S_M values ranged between 0.06 – 0.38 mL.h⁻¹ for the peak season, 0.06 – 0.26 mL.h⁻¹ for the off-peak season, while increasing after day 275 (0.2 – 0.3 mL.h⁻¹). When using ATM, the S_M values ranged between 0.016 – 0.17 mL.h⁻¹ during the peak season and 0.01 – 0.06 mL.h⁻¹ during the off-peak season.

With the 10 h incubation assays (Fig. 4.6 B), higher values were found for the S_M values, especially during the processing season and then after day 250 the higher values probably was as a result of the higher pond temperature. It was also evident that some activity tests performed during the off-peak season resulted in very low S_M values (days 229 and 270). This suggests that the methanogenic population was barely active, which was probably as a result of the lower pond temperatures combined with lower COD values in the feed substrate (Chapter 3) and consequently very little CH₄ production occurred. Once again it appears as if environmental temperatures could have had a negative impact on the methanogens as the methanogenic activity increased again after day 250 with the increases in the pond temperature after the winter month period as shown in Fig. 4.2. It is interesting to note that the COD values of the inlet feed of the pond after day 250 were as low as they were during the winter months.

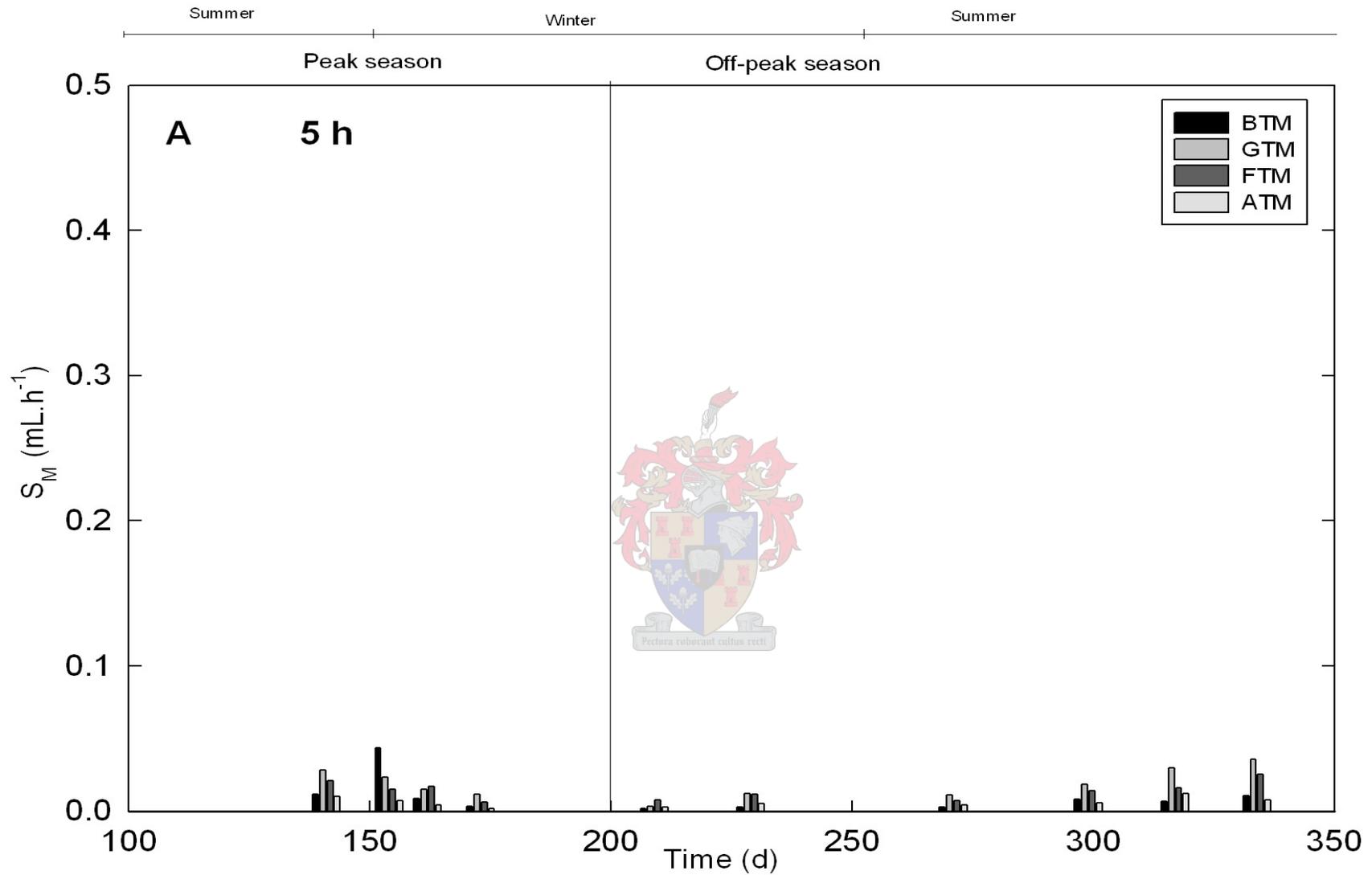


Figure 4.6 The methane production rates (S_M) during peak-and off-peak season after 5 h (A), 10 h (B) and 25 h (C) incubation.

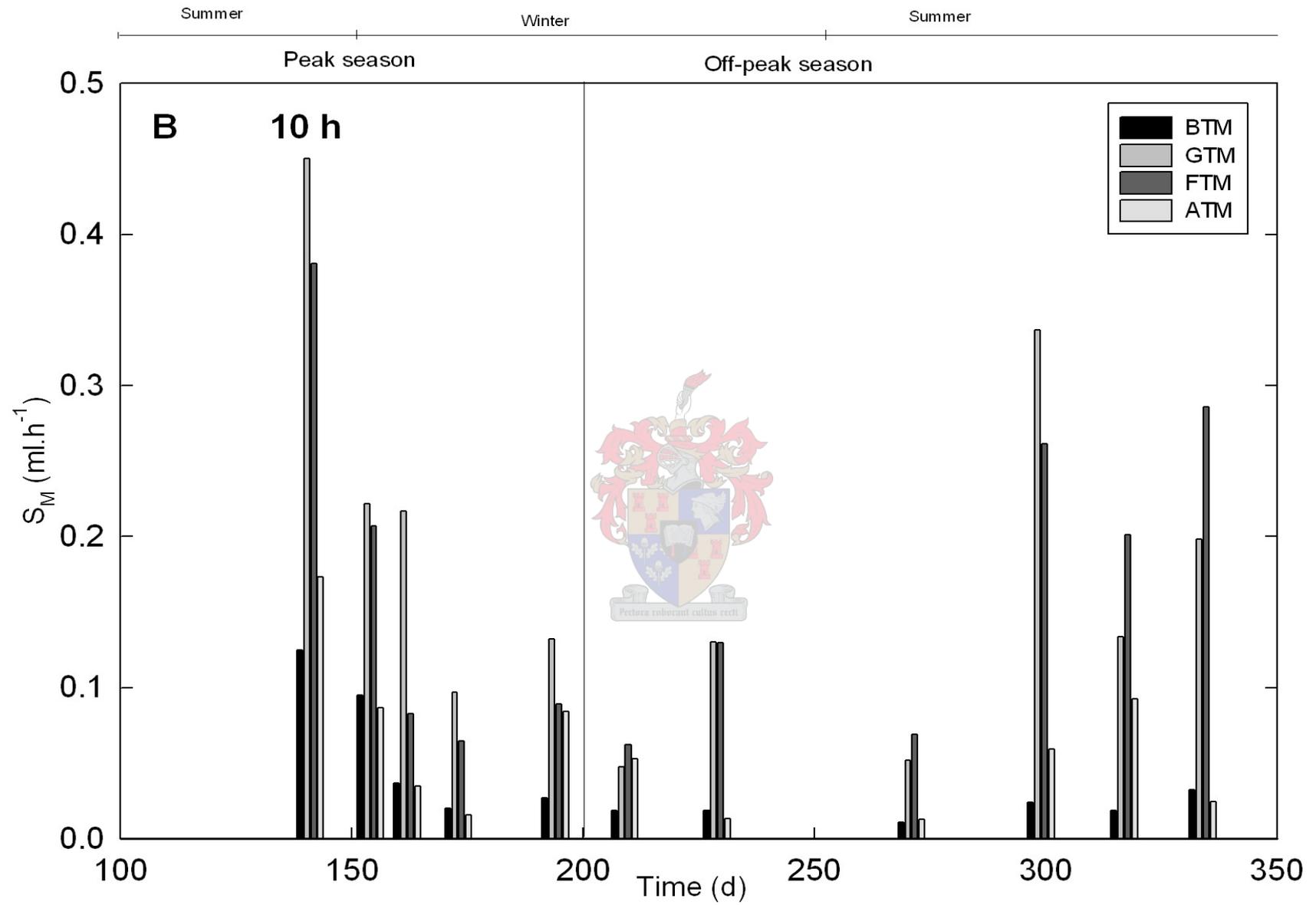


Figure 4.6 continue

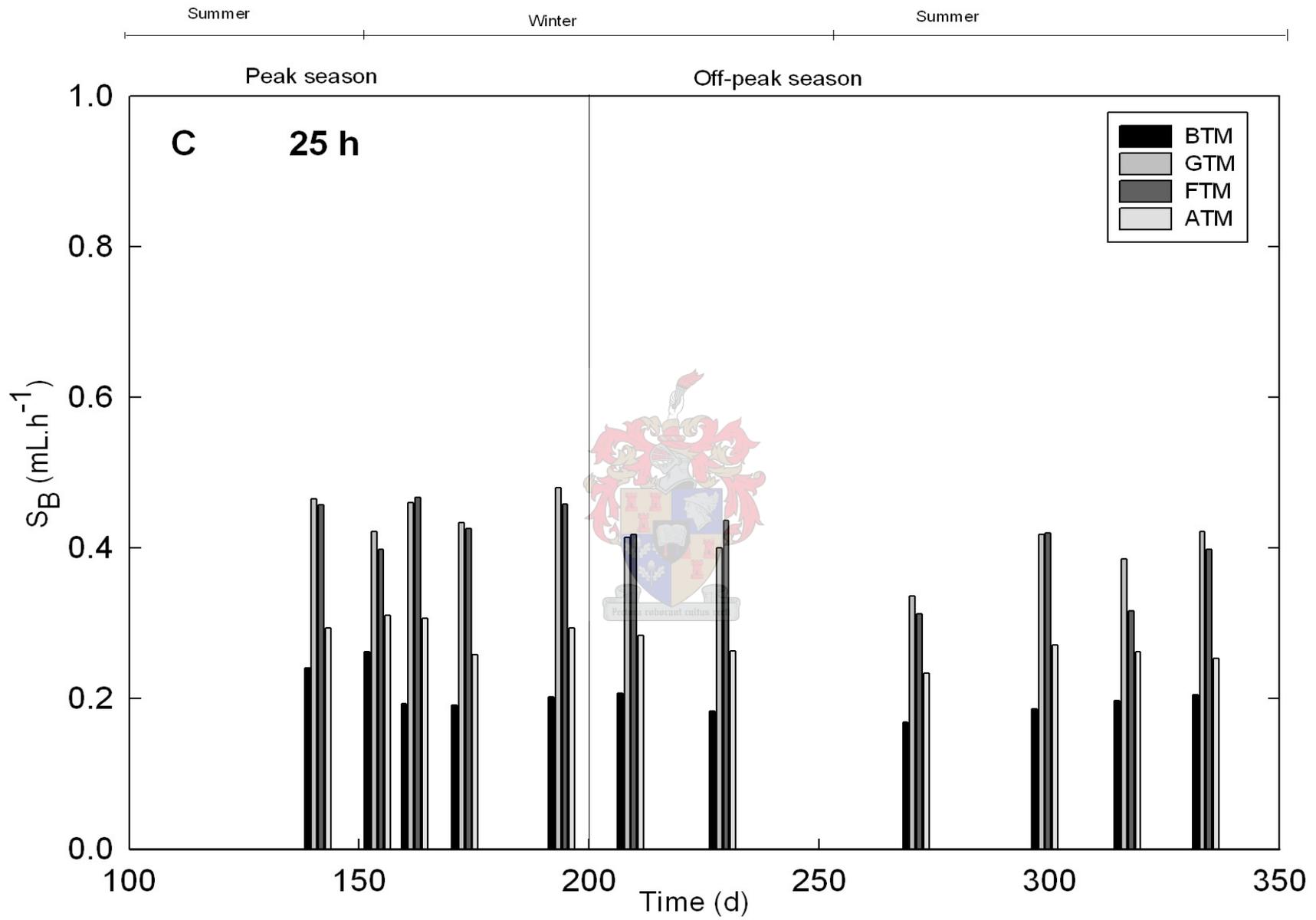


Figure 4.6 continued

The S_M after 25 h incubation (Fig. 4.6 C) when using BTM ranged between 0.09 – 0.17 mL.h⁻¹ during the peak season remained fairly stable throughout the off-peak season. The S_M values for the GTM ranged between 0.12 – 0.38 mL.h⁻¹ during the peak season and 0.14 – 0.21 mL.h⁻¹ during the off-peak season. For the FTM, the S_M values ranged between 0.11 – 0.34 mL.h⁻¹ for the peak season and 0.05 – 0.26 mL.h⁻¹ for the off-peak season. When using ATM, the S_M values ranged between 0.13 – 0.26 mL.h⁻¹ during the peak season and 0.09 – 0.18 mL.h⁻¹ during the off-peak season.

After 25 h incubation, the S_M values (Fig. 4.6 C) for the GTM and FTM did not vary much from the values obtained after the 10 h incubation. However, there was a definite increase in the overall S_M for the BTM and ATM from 10 h (Fig. 4.6 B) to 25 h. This increase was probably due to the longer incubation time allowing for a more complete conversion by the methanogens of the substrates to CH₄, and therefore resulting in higher S_M values. This is also a factor that must be taken into consideration when calculating the biogas production potential of Pond A.

Biogas formation test

The biogas formation test was designed to simulate, under lab-conditions, an anaerobic pond system treating apple factory wastewater. From the data obtained in the direct activity tests, the reaction of the acidogenic and methanogenic populations present in the sludge samples from Pond A to optimal conditions can be concluded. However, it is not clear how the general microbial consortium of the sludge would perform under the actual conditions experienced by the pond throughout the year. This information is necessary to extrapolate the data to obtain the biogas production potential, or more importantly, the CH₄ production potential for this specific anaerobic pond (Pond A).

The data of the biogas formation tests (after 72 h) are presented in Fig. 4.7 and specifically in terms of total biogas production, % COD reduction and % CH₄ content. The biogas formation test (at 25°C) resulted in a total biogas production of 41.0 mL with 42.0% CH₄ content and 73% COD reduction over a period of 72 hours (Fig. 4.7). The total biogas production obtained at 18°C incubation was 14 mL with 37% CH₄ and 70% COD reduction. At 10°C only 2.5 mL biogas was produced with a 9% CH₄ content and 59% COD reduction. Thus, the highest incubation temperature (25°C) resulted in the most biogas being produced, followed by 18°C and with 10°C resulting in the lowest biogas volume.

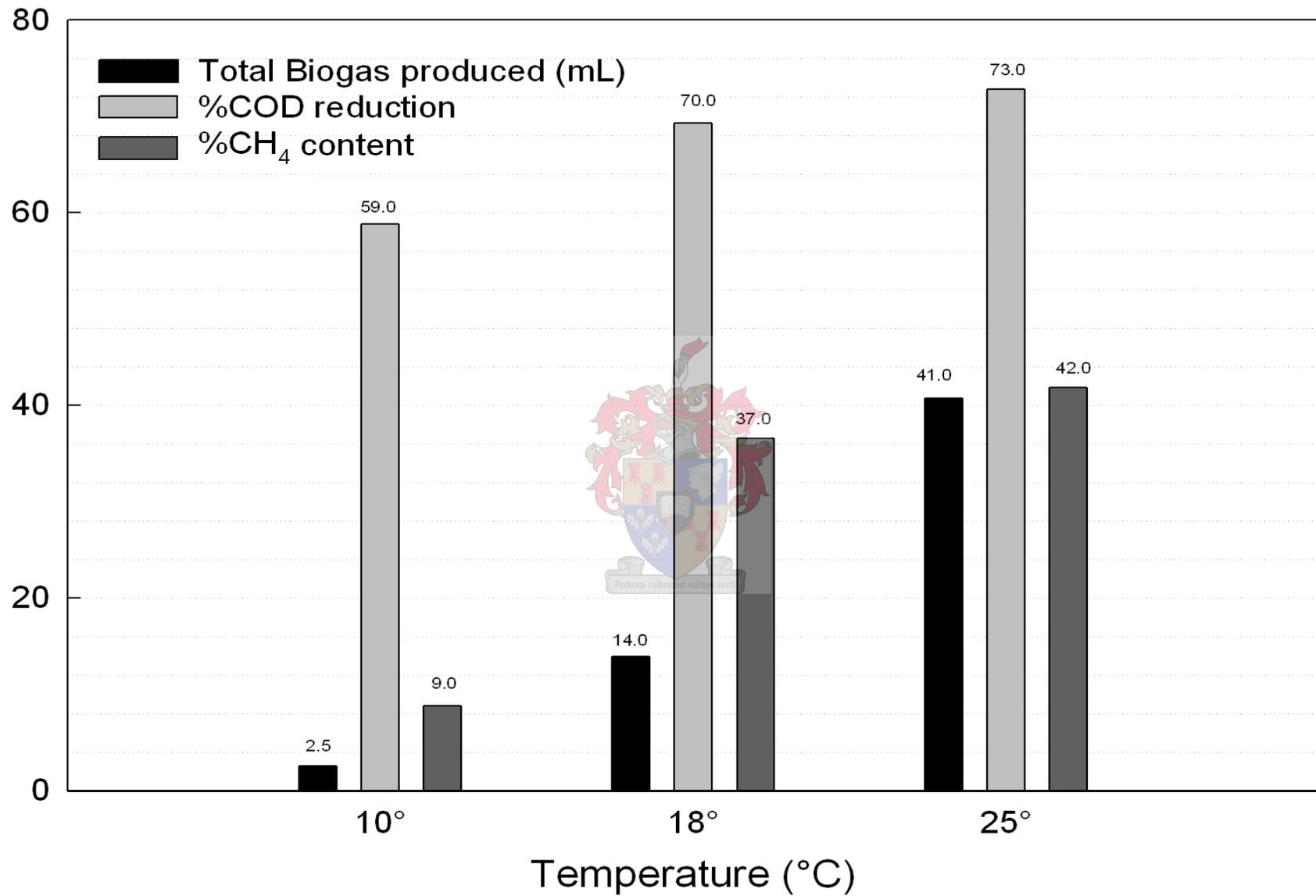


Figure 4.7 The total biogas produced, %COD reduction and %CH₄ content at the 10°, 18° and 25°C after 72 h incubation.

The biogas formation tests indicated that microbial activity and therefore biogas production was dependent on especially favourable temperature conditions. This confirms that temperature is an important operational factor that influences microbial activity and subsequently influences the treatment efficiencies (COD reduction), biogas volume and the CH₄ content. The lower efficiency found at 10°C was probably as a result of the mesophilic character of the microbial populations in the sludge. It has been reported that at low temperatures, which may occur in anaerobic ponds during winter months, very little CH₄ production occurs (Bryant & McInerney, 1981; Patel & Madamwar, 1984; Bitton, 1999) and this was confirmed in this study.

For the tests carried out at 25°C and 18°C, the activity trends of the results for the different temperatures (Fig. 4.8) were similar. The most biogas was produced during the first 24 h. This is probably due to the higher COD level at the start. Therefore, if the substrate contains easily degradable compounds the microbial populations digest the most organic material during the first 24 h with subsequent increase in biogas production. In this study (Fig. 4.8) the biogas production decreased after 48 h. This was ascribed to the depletion of easily degradable carbon sources and of other nutrients. It may also be due to the fact that some microorganisms had started dying off. After the completed period of 72 h, it was found that the biogas production started to increase, but only slightly. This may be ascribed to possible shifts in population dynamics and kinetics or inhibition as a result of a specific carbon source being neutralised (O'Kennedy, 2000) or that the cellular content of the dead cells was being reused by the active microbes to produce a little biogas.

CH₄ production potential of anaerobic Pond A

The BMPs for Pond A were determined (Table 4.5) using the data from the different processing seasons during the 2004 year. The calculations were based on the known flow rate for Pond A and the average COD reduction at 25°C (Table 4.5) as obtained in the biogas formation tests. From the biogas formation test values it is possible to estimate the theoretical maximum CH₄ production potential as well as the actual minimum CH₄ volume produced. This was done using 15% of the total pond volume (Fig. 4.9), the actual COD reduction as well as the actual CH₄ content obtained during the biogas formation tests at the three different temperatures. Due to the sloping construction design of Pond A only

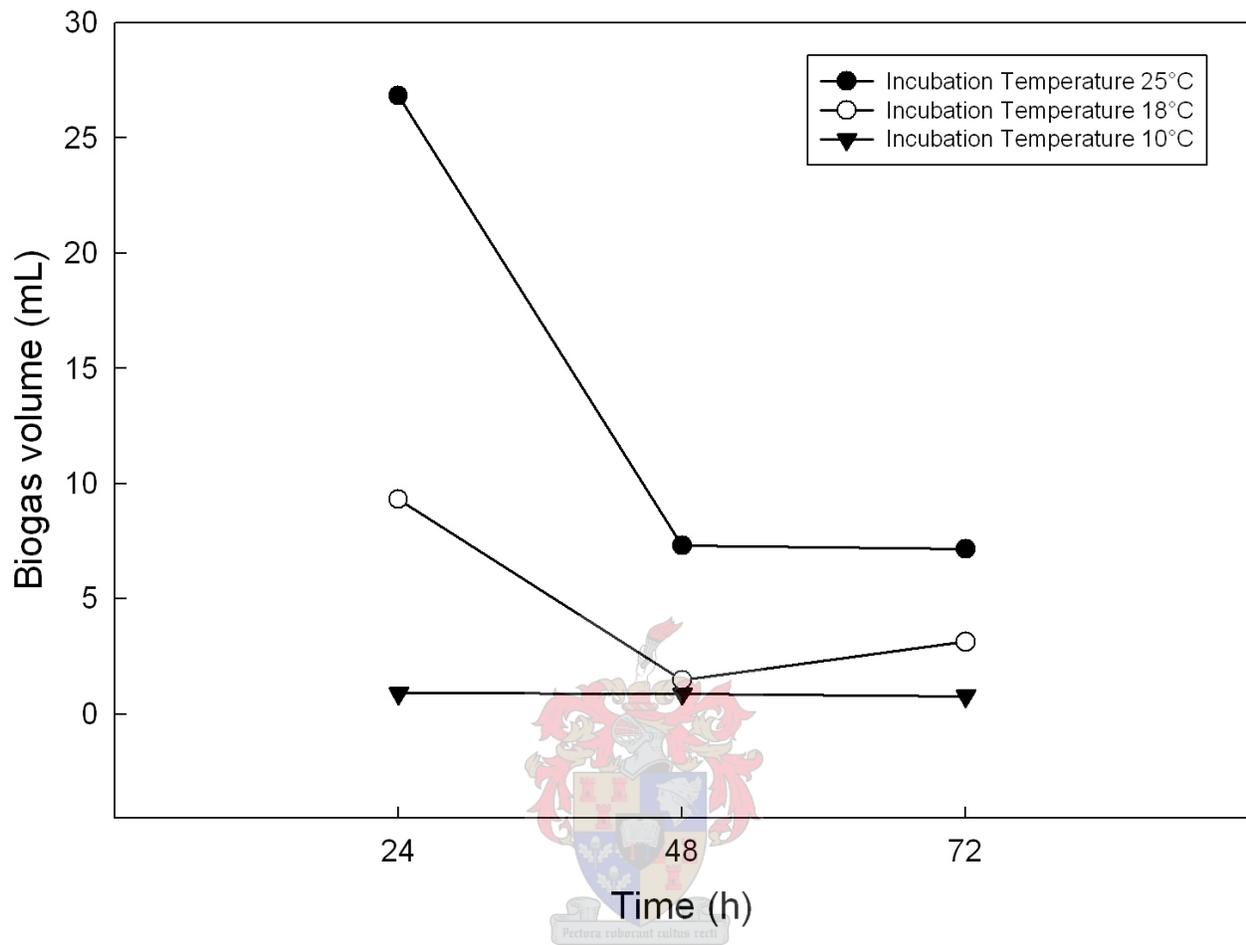
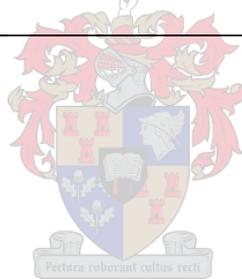


Figure 4.8 The total biogas volume obtained during the biogas formation test at 10°, 18° and 25°C.

Table 4.5 Parameters used in the BMP calculations of Pond A (Droste, 1997).

Flow Rate (m ³ .d ⁻¹)	COD reduction (%)	Max. CH ₄ yield (m ³ CH ₄ .kg ⁻¹ COD)	Average incoming COD during the 2004 year (kg.m ⁻³)	BMP (m ³ .d ⁻¹)
3 000	73.0	0.35	5.0	3 832



For clarification purposes:

$$\mathbf{BMP = QEMS_{T0} = 3832}$$

Where:

$$\mathbf{Q = 3\ 000}$$

$$\mathbf{E = 0.73}$$

$$\mathbf{M = 0.35}$$

$$\mathbf{S_{T0} = 5.0}$$

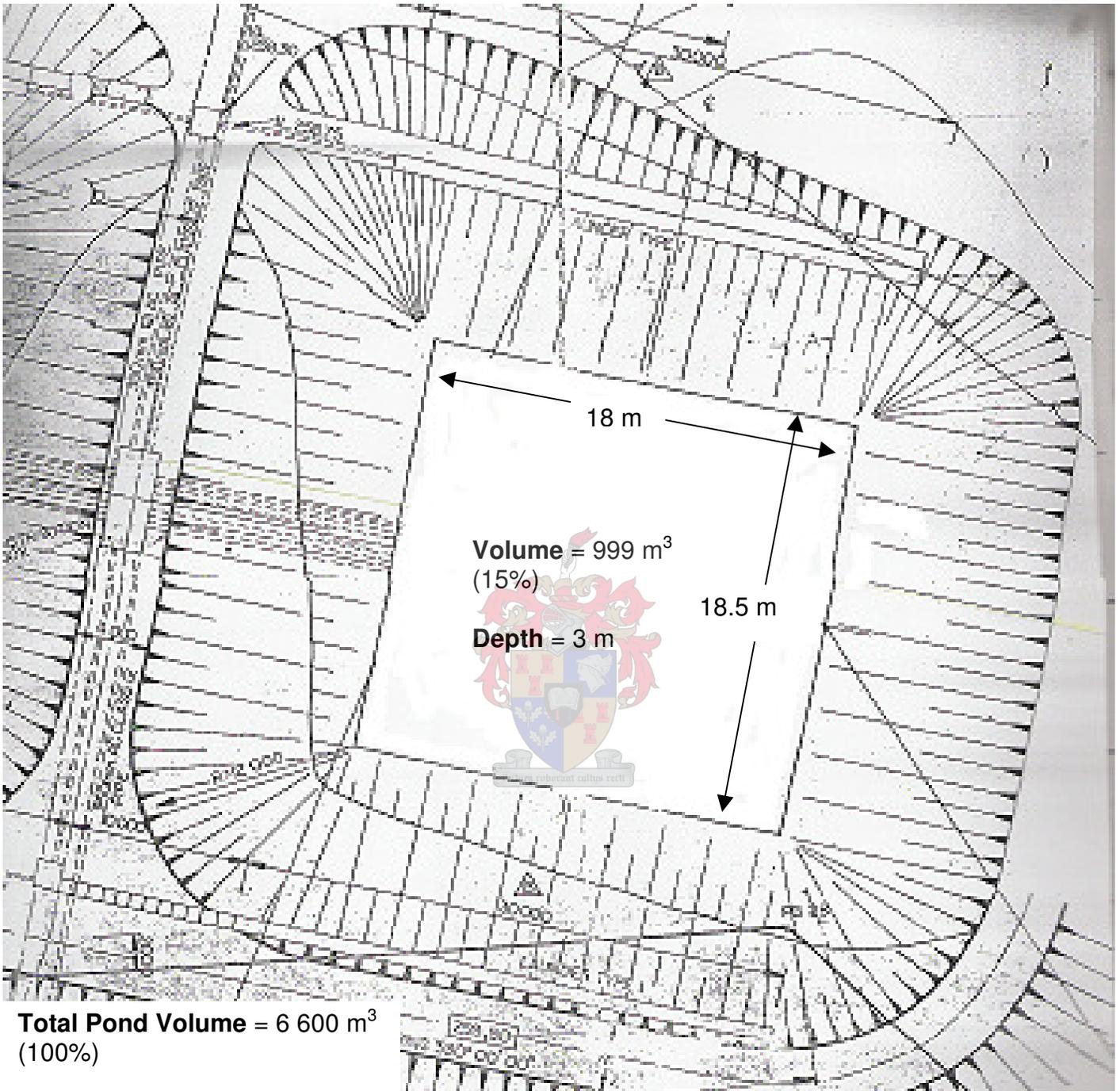


Figure 4.9 Schematic representation of Pond A illustrating the 15% volume calculations.

the central section, as illustrated in Fig. 4.9 had a uniform depth and thus (15%) was used in the BMP calculations.

The calculations for the BMP were based on the maximum theoretical CH_4 yield of $0.35 \text{ m}^3 \text{ CH}_4 \cdot \text{kg}^{-1} \text{ COD}_{\text{removed}}$ as stipulated by Droste (1997). For the BMP calculations a pond flow of $3\,000 \text{ m}^3 \cdot \text{d}^{-1}$, an average COD reduction of 73%, an average COD concentration of $5 \text{ kg} \cdot \text{m}^{-3}$ (as determined in Chapter 3) and the maximum CH_4 yield of $0.35 \text{ m}^3 \text{ CH}_4 \cdot \text{kg}^{-1} \text{ COD}$ of Droste (1997) was used. The BMP was calculated to be $3\,832 \text{ m}^3 \cdot \text{d}^{-1}$ (Table 4.5).

In Droste's (1997) calculations it was assumed that all COD was converted to CH_4 , but in this study, as shown in Fig. 4.7, it was clear that this did not happen. Thus, for further calculations in this study, the COD removal and CH_4 values as given in Fig. 4.7, will be used. Based on the temperatures, the period ranges for the pond were established using data obtained in Chapter 3 as given in Table 4.4 and are as follows: for the October to April time period higher temperatures were found for the pond ($\geq 25^\circ\text{C}$), followed by a low temperature period (June to August) with a temperature range of $< 20^\circ\text{C}$ to $> 15^\circ\text{C}$ and the May and September periods with temperatures of $< 25^\circ\text{C}$ and $> 10^\circ\text{C}$.

The determined CH_4 volumes (Table 4.6), based on results obtained from the biogas formation tests and average temperatures showed that for the time period October to April (212 days), the average temperature was 25°C and for the June to August period (92 days), the average temperature was 18°C . An average pond feed COD concentration of $5\,000 \text{ mg} \cdot \text{L}^{-1}$ ($5 \text{ kg} \cdot \text{m}^{-3}$) was used during the calculations. Assuming that an average of 73% COD reduction occurs and that 100% of the biogas formed is CH_4 , the maximum CH_4 production estimated for October to April period would be $340\,000 \text{ m}^3$ with an energy value of $23\,000\,000 \text{ MJ}$. However, results from the biogas formation tests indicated that at 25°C , 73% COD reduction does occur, but only 42% is converted to CH_4 , which gives only $20\,600 \text{ m}^3 \text{ CH}_4$ ($1\,380\,000 \text{ MJ}$). This was subsequently taken as the minimum actual volume of CH_4 to be produced by the Pond A system for the October to April period.

For the time period June to August at a temperature of 18°C , the estimated maximum CH_4 was $106\,000 \text{ m}^3$ ($7\,110\,000 \text{ MJ}$), whilst the minimum estimated actual CH_4 was 2033 m^3 ($136\,000 \text{ MJ}$).

Lastly, for May and September (61 days), the maximum estimated CH_4 was $133\,400 \text{ m}^3$ ($8\,940\,000 \text{ MJ}$) and minimum estimated actual CH_4 was 8090 m^3 ($542\,000 \text{ MJ}$).

Table 4.6 Impact of different temperature ranges on the estimated CH₄ production during the 2004 year. Only 15% of the total pond (Pond A) volume was used in the calculations.

Temperature Range (°C)	Calculated Average Temperature (°C)	Time Period (Months)	Total calculated CH ₄ (m ³)	*Energy from CH ₄ (MJ)
≥ 25°C	25°	Oct – Apr (212 days)	Max. 340 000 Min. 20 600	23 000 000 1 380 000
< 20°C > 15°C	18°	Jun – Aug (92 days)	Max. 106 000 Min. 2033	7 110 000 136 000
< 25°C > 10°C	20°	May and Sept (61 days)	Max. 133 400 Min. 8090	8 940 000 542 000

* Energy value calculated using a typical heating equivalent of 50 MJ.Kg⁻¹ CH₄ at 1 atm and a density of 0.674 kg.m⁻³.

Discussion and conclusions

The biogas (S_B) and the methane production rates (S_M) were used to assess the activity of the specific microbial populations throughout the processing seasons. The cumulative biogas and CH_4 volumes were used to indicate microbial activity and to relate decreased activity to the presence of unfavourable pond environmental conditions. The activity tests used in this study confirmed that the peak and off-peak seasons and subsequent air temperature changes strongly influenced the microbial activity of the sludge. The data obtained also showed that the cumulative biogas performed on sludge from the four sampling positions (green, red, yellow and blue) showed that there appeared to be a homogenous distribution of the microbial populations throughout the sludge bed in the pond. This suggests that sufficient pond mixing takes place ensuring that the microbial activity is distributed throughout the pond and therefore that efficient anaerobic digestion occurs.

The influence of the peak seasons on the bacterial activity was found to be very pronounced especially during peak season when higher substrate COD concentrations and favourable temperatures were present. The data from the study clearly showed that the decrease in activity during the off-peak season was mainly due to lower pond temperatures. Similarly, the gradual increases in activity after day 275 were ascribed to higher air and pond temperatures.

In this study the biogas formation test was utilised to simulate the biogas formation in an anaerobic pond system treating apple factory wastewater and it was found that the microbial activity and therefore biogas production was dependent on especially favourable temperature conditions. This again confirms that temperature is an important operational factor that influences microbial activity and subsequently the treatment efficiency (COD reduction), biogas values and CH_4 content. It was concluded that at $25^\circ C$ and $5\ 000\ mg.L^{-1}$ COD, the biogas production (40 mL), CH_4 content (41%) and %COD removal (73%) was reasonable considering the conditions.

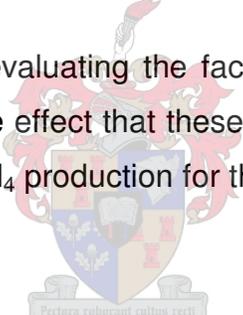
The above calculated CH_4 estimations are valuable and suggest that CH_4 recovery from the Pond A system will be a worthwhile consideration for most part of the year, and especially during the peak seasons. These estimated CH_4 values should be considered as the minimum obtainable biogas and in fact would be much higher in reality due to the fact

that only 15% of the total pond volume was used in these calculations. Furthermore, it should be noted that the biogas formation tests were conducted on laboratory scale and that the large scale Pond A system may reveal different results to that obtained in these calculations. These calculations however represent under-estimations, since the actual average COD reduction of Pond A for the peak season as measured each day at the pond outlet was 91%, whereas a 73% reduction was used in the calculations of this study.

In conclusion, from the average COD reduction of Pond A, the theoretical CH₄ calculations and estimates based on the results obtained during the biogas formation tests, it is clear that CH₄ recovery from the anaerobic pond would definitely be a worthwhile consideration. If it were assumed that the estimated CH₄ volumes (based on 15% of the pond volume) obtained could be applied as an energy source, the minimum yearly savings in coal usage would amount to about R 665 000. For this calculation the average cost of coal was taken as R 600 per ton (D. Koorts, Appletiser SA, South Africa, personal communication, 2004).

This study was valuable in evaluating the factors such as pond conditions, pond activity and air temperatures and the effect that these have on biogas production potential as well as, and more importantly, CH₄ production for the purposes of energy recovery.

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CHAPTER 5

GENERAL DISCUSSION AND CONCLUSION

The Western Cape Province is known for its extensive deciduous fruit industry and is home to a variety of fruit processing plants (Hurndall, 2005). During fruit processing large volumes of wastewater are generated. The recent implementation of water restrictions (Anon., 1998) has emphasised the importance of waste minimisation and sustainable environmental growth. Therefore the treatment and disposal of the polluted wastewaters has become a cause of some concern to both fruit processors and controlling bodies responsible for waste management (Water Research Commission, 1993). One treatment option, on-site anaerobic treatment of wastewater, has become very popular due to its economical and environmental advantages.

In this study an anaerobic pond system, treating apple-processing wastewater, was studied to establish a characteristic operational profile. The profile was then used to determine and evaluate the treatment efficiencies and to pinpoint environmental limitations throughout the peak and off-processing seasons. The various process parameters used to establish the operational profile clearly showed that the system was impacted by several processing and environmental factors as well as the seasonal nature of fruit processing. It was clear that the high COD concentrations (5 000 – 16 500 mg.L⁻¹) and higher temperature (30°C, Chapter 3) was favourable to the anaerobic treatment process during the peak season. In contrast, the lower COD concentrations (500 mg.L⁻¹) and lower temperature ranges (10° - 18°C) during the off-peak season proved to be unfavourable to the digestion process.

It was also found that the alkalinity profile was as a direct consequence of the lime dosing intervals. The alkalinity of the pond during peak and off-peak season was much lower than specified in the literature for optimum operating conditions for anaerobic digestion (Chen *et al.*, 1980; Bitton, 1999). However, the alkalinity levels in Pond A appear to be acceptable for the digestion taking place in this particular anaerobic pond. This conclusion was also corroborated by the stable pH profile found for Pond A. It was thus concluded that stable pH values, sufficient nutrient availability and acceptable alkalinity levels are maintained throughout the year. From the data obtained it was also concluded

that the pond treatment system is self-maintaining and does not require nutrient supplementation.

The treatment process was also found to be stable during the off-peak season and that the system has the ability to recover rapidly and reach efficient operational levels by the onset of the peak season. The most limiting factor however, was clearly found to be the lower pond temperatures experienced during the winter months and off-peak season.

In this study (Chapter 4) it was also found that the influence of the season on the bacterial activity of the sludge was very pronounced especially during peak season when higher substrate COD concentrations and favourable temperatures were present. The decrease in microbial activity was clearly shown during the off-peak season and ascribed to the lower pond temperatures. This was confirmed by applying the biogas formation test to simulate the biogas formation as found in the anaerobic pond system. It was found that the microbial activity and therefore biogas production was dependent on especially favourable temperature conditions. This again confirmed that temperature is an important operational factor that influences microbial activity and subsequently the treatment efficiency (COD reduction), biogas values and CH₄ content.

From the data obtained in this study it was concluded that at 25°C and 5 000 mg.L⁻¹ COD, the biogas production, CH₄ content and %COD reduction was reasonable considering the conditions. However, in the literature it has been stated that the CH₄ content should be between 55 - 65% at 35°C in order to allow justifiable energy recovery (Droste, 1997). When considering the data from the 18°C incubation, the results were not drastically lower than those found at 25°C. Though, at 10°C it was clear that the biogas production and CH₄ content decreased considerably although the %COD reduction of 59% was still acceptable. It thus appears that when an anaerobic pond system is subjected to environmental changes such as decreased air temperature, it would still be fairly efficient in terms of COD reduction, but the CH₄ production would probably not be acceptable for energy recovery purposes.

During the simulation studies it was found that at incubation temperatures of 25°C and 18°C the most biogas was generated during the first 24 h. This is of value when considering energy recovery and suggests that an HRT of 3 days would be sufficient as longer HRTs would probably not lead to an increase in biogas production. This however, should be investigated and confirmed. It may also be assumed that the increased

temperatures during the summer months (around 32 °C) would lead to more biogas and a higher CH₄ content.

The characteristic operational profile established by monitoring various process parameters indicated that the anaerobic treatment process was stable, efficient and self-maintaining. The trends in the profile also suggested that the microbial population of the anaerobic pond were susceptible to changes in environmental conditions. Nevertheless, the system profile as well as microbial activity indicated easy microbial recovery during the peak season. Furthermore, it was estimated, based only on 15% of the ponds' volume, that the CH₄ production potential during the peak season could contribute to a minimum cost saving of about R 700 000 when used in conjunction with coal (when the average cost of coal is R 600 per ton - D. Koorts, Appletiser SA, South Africa, personal communication, 2004) as fuel for boilers. Further theoretical calculations for the total Pond A volume (100%) indicated that the average yearly CH₄ production potential would result in over 1 000 000 m³ (40 000 000 MJ). This can contribute to a cost saving of about R 1 200 000 per year, which would otherwise have been used to purchase coal. This however emphasises the potential of energy recovery from the anaerobic pond and may only be achieved when the pond system is optimised. To further optimise the CH₄ production potential it is recommended that the anaerobic pond (Pond A) is covered. This would improve heat-retention during the winter months and off-peak seasons and consequently increase the digestion efficiency. This will ultimately improve the overall and or downstream wastewater treatment processes and most importantly optimise CH₄ recovery in Pond A.

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