

**FIRST-STAGE AND SINGLE-STAGE CONTINUOUSLY
STIRRED TANK ANAEROBIC DIGESTION OF SYNTHETIC
COMPLEX WASTEWATER AND PIGGERY WASTEWATER
(WITH EMPHASIS ON THERMOPHILIC TEMPERATURE)**

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DECLARATION

I declare that this thesis is my own account of my research work undertaken which has not been previously submitted for a degree at any tertiary educational institution

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ABSTRACT

Single-stage continuously stirred tank reactor (CSTR) is commonly used in the anaerobic treatment of animal manure slurry, municipal sewage sludge and concentrated wastewaters containing a high proportion of biodegradable particulate organic materials at relatively long hydraulic retention times (HRTs) of 12 to 24 days. It is also commonly used as a holding tank to equilibrate the big variations in wastewater flow or pollution strength as well as for pre-acidification of wastewater. Its simplicity, ease of operation, low capital and maintenance costs are appealing features that made it a natural choice of reactor configuration for the pilot-scale thermophilic first-stage acidogenic digester of a two-stage thermophilic-ambient anaerobic digestion system which is based at Roseworthy Campus of University of Adelaide, South Australia and operated by South Australian Research and Development Institute (SARDI).

As the first-stage acidogenic reactor plays a crucial role in the solubilisation of particulate organic matter in complex substrates to soluble organics and acidification to volatile fatty acids (VFAs) for enhancing pathogens destruction in wastewater treatment, the initial aim of this research study was to optimise the first-stage anaerobic CSTR to effectively convert particulate-containing complex organic wastewater to intermediate fermentation products for feed to the second-stage methane reactor. Pig feed pellets was used as the model substrate to prepare the complex synthetic wastewater to investigate the effects of temperature (37, 47 and 55°C) and pH (6, 7 and 8) on organics solubilisation and acidification in two sets of batch vial experiments while the effects of HRT (4- to 1-day) on organics conversion efficiency of the semi-continuous first-stage anaerobic CSTRs were investigated in two sets of experiments conducted at mesophilic (37°C) and thermophilic (55°C) conditions. Findings from the batch vial experiments with low organic strength (4 g/L TCOD) wastewater found mesophilic temperature at 37°C and pH 7-8 were optimum conditions for solubilisation (hydrolysis) and acidification than at thermophilic temperatures of 47°C and 55°C. Results from the semi-continuous CSTR anaerobic reactors confirmed that the mesophilic (37°C) reactor hydrolysed

and acidified significantly more particulate and soluble organic matter respectively than thermophilic (55°C) reactor, with 2-day HRT being the optimum for both the anaerobic acidogenic reactors. The lack of methane in the biogas which contained hydrogen and carbon dioxide confirmed that the methanogens present in the acidic reactor effluents were completely inhibited.

Following reports that the pilot-scale first-stage thermophilic (55°C) acidogenic reactors treating raw piggery wastewater was experiencing substantial loss of volatile fatty acids to methane formation at relatively short HRTs of 4 and 7-day, the complex synthetic wastewater was replaced with real piggery wastewater from Roseworthy Campus's piggery holding sump to allow meaningful lab-scale reactor experiments to be conducted in order to provide realistic information for the pilot-scale acidogenic reactor. Semi-continuous first-stage anaerobic reactor experiments were carried out to examine the influence of piggery influent concentrations with and without pH reduction on the fermentation behaviour of the thermophilic acidogenic reactor at a shorter HRT of 2-day. The 2-day HRT was found to be optimum in previous acidogenic experiments with the synthetic complex wastewater.

The studies on the acidogenic treatment of piggery wastewater at 2-day HRT revealed that irrespective of the feed concentrations or organic loading rates, first-stage anaerobic treatment of the piggery wastewater without pH intervention could not completely inhibit the syntrophic acetogenic and methanogenic microorganisms because of the wastewater's inherently high levels of alkalinity and ammonia-nitrogen which buffered the system against VFA souring. Some losses of total VFAs were observed at the highest TCOD feed concentration of 13 g TCOD/L and OLR of 6.5 g/L/d. The vast differences in the physico-chemical and microbiological characteristics of the raw piggery wastewater and synthetic complex wastewater, particularly with respect to their initial ammonia-nitrogen, soluble COD, volatile fatty acids (VFA), buffering capacity and anaerobic microorganisms, were the key determining factors for the contradictory outcomes in organics conversion performance of the thermophilic and mesophilic first-stage CSTRs.

Although the study on pH reduction of the piggery wastewater to pH 5.5 found the approach was successful in suppressing the activities of syntrophic consortia of acetogenic and methanogenic microbial populations while stimulating the acidogenic

bacteria, the operational inconvenience from foaming-related spillages and the anticipated need to re-adjust the acidic effluent pH to neutral for feed to the second-stage reactor far out-weighed the small gains in the increased hydrolysis and acidification of the piggery influent organic matter.

The observations that around 30% of the organics still remained as insoluble particulate form in the treated effluent and more than 60% of the organic carbon compounds in the raw piggery wastewater was already in soluble and acidified forms coupled with its high buffering capacity which protects the anaerobic system against failure from VFA souring, it was decided that single-stage thermophilic anaerobic digestion at longer HRT of 10- and 15-day might be more cost-effective for enhancing the solubilisation of the particulate organics and organic carbon conversion to methane in the undiluted piggery wastewater. Semi-continuous thermophilic CSTR experiments at 55°C were carried out to examine the extent of organic carbon conversion at 10- and 15-day HRT. Mesophilic CSTR experiment at 37°C was also carried out to compare its organics conversion performance with the thermophilic reactor at 15-day HRT.

The results show that while increasing the HRT of the thermophilic anaerobic CSTRs from 2- to 10- and 15-day saw a gradual increase in specific methane yields, the methane yield at the longer HRT of 15-day was considered low (26% of total COD fed) based on the COD material balance of the digested effluents. Around 30% of the organic matter still remained as non-biodegradable particulate organics while propionate (19%) and unidentified non-VFA soluble organic matter (17%) formed the two largest groups of unconverted soluble organics in the digested piggery effluent. The build-up of propionate at higher HRT of 10- and 15-day which correlated positively with increased free ammonia concentration implied that the syntrophic propionate-oxidising bacteria and hydrogenotrophic microorganisms were under increased stress. At 15-day HRT, although anaerobic thermophilic digestion at 55°C had significantly higher specific methane yield than mesophilic digestion at 37°C, the chemical quality of thermophilic digested effluent was poor with regards to its higher levels of free ammonia, propionate, total VFA and soluble COD compared to the mesophilic effluent. However, thermophilic digestion is universally recognised for its higher pathogens destruction efficiency than mesophilic digestion.

Five sets of thermophilic (55°C) batch vial experiments were conducted to investigate the single effect of pH reduction, chemical (zeolite, humic acid) and biological (piggery biomass, municipal biomass) supplements as well as the combined effects of pH reduction and chemical or biological supplements in enhancing methane production from thermophilic piggery effluent. Reduction of the piggery effluent pH from 8.1 to 6.5 alone and zeolite treatment (10 to 20 g/L) with or without pH reduction of the piggery effluent to pH 6.5 were found to be effective strategies for enhancing methane production yet not elevating the effluent COD level compared to its initial level.

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LIST OF ABBREVIATIONS

APHA	American Public Health Association
atm	atmosphere
CSTR	Continuously Stirred Tank Reactor
CO ₂	Carbon dioxide
d	day
FISH	Fluorescence <i>In Situ</i> Hybridisation
GC	Gas Chromatography
g	gram
HRT	Hydraulic Retention Time
hr	hour
H ₂	Hydrogen
IBS	Integrated BioSystem
kg	kilogram
kJ	kiloJoule
L	Litre
mL	millilitre
µL	microlitre
mM	millimolar
mg	milligram
M	Molar
N	Normality
ng	nanogram
CH ₄	Methane
OLR	Organic Loading Rate
PCR	Polymerase Chain Reaction
rRNA	ribosomal RiboNucleic Acid
rDNA	ribosomal DeoxyRibonucleic Acid
stp	standard temperature and pressure
ΔG°	standard Gibbs free energy
SRB	Sulphate Reducing Bacteria

SRT	Solid Retention Time
SCOD	Soluble Chemical Oxygen Demand
SARDI	South Australian Research and Development Institute
TPAD	Temperature-Phased Anaerobic Digestion
TVFAs	Total Volatile Fatty Acids
TSS	Total Volatile Solids
TS	Total Solids
T-RFLP	Terminal-Restriction Fragment Length Polymorphism
TCOD	Total Chemical Oxygen Demand
VSS	Volatile Suspended Solids
VS	Volatile Solids