



**ADVANCEMENTS IN POULTRY SLAUGHTERHOUSE WASTEWATER
TREATMENT PLANT DESIGN**

by

**MAGGIE NTOMBIFUTHI BINGO
205173497**

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**Supervisor: Dr. Moses Basitere
Co-supervisor: Prof. Seteno Karabo O. Ntwampe**

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DECLARATION

I, **Maggie Ntombifuthi Bingo**, declare that the contents of this thesis represent my own unaided work, except where specifically acknowledged in the text, and that the thesis has not previously been submitted for academic examination towards any qualification. Furthermore, it represents my own opinions and not necessarily those of the Cape Peninsula University of Technology, or their sponsors.



Signed

13 September 2021

Date

ABSTRACT

The poultry industry is the largest agricultural sector in South Africa (SA), and it consumes large quantities of freshwater, which end up being discharged as high-strength poultry slaughterhouse wastewater (PSW). Generally, PSW is discharged into municipal wastewater drainage systems, which is detrimental for municipal treatment plants since a limited number of these wastewater treatment works (WWTWs) operate optimally. Furthermore, increasingly stringent standards for effluent discharge have placed an urgency on the development of advanced wastewater treatment technologies. Currently, extensive research has been done experimentally on lab-scale anaerobic and aerobic bioreactors; however, to upscale these reactors for industrial application, simulation and process modelling must be conducted to assess the feasibility of any proposed system.

This study initially investigated the development of a model to simulate the performance of lab-scale anaerobic digesters treating PSW to assist small-scale poultry product producers. The single-stage, two-stage, and three-stage anaerobic digestion (AD) models were assessed with regards to predicting the removal of organic matter, total suspended solids (TSS) and volatile suspended solids (VSS) in PSW. However, from the model, there was a minuscule increase in nutrients, ammonia (NH_3) and phosphate (PO_4^{3-}); thus, the model design required refinement. Thereafter, the performance of a lab-scale integrated multi-stage PSW treatment system consisting of an aerobic pre-treatment tank, an expanded granular sludge bed (EGSB) bioreactor coupled with submerged ultrafiltration (UF) membrane, with the objective being to assess the treatment efficiency of the individual treatment systems as well as that of the overall treatment system. The possibility of treating PSW to a water quality standard compliant with discharge by-laws or effluent discharge standards was investigated.

The PSW used in this study was collected in 25L containers from a poultry slaughterhouse located in the Western Cape (WC) Province, SA and stored in a refrigerator at less than 4°C until it was fed to the treatment plant. Ecoflush™, a hydrolysis agent, was added to the pre-treatment tank (25L) together with raw PSW, and the mixture was aerated for 24h using an adjustable air pump. The aerated mixture was then allowed to settle for a further 24h to reduce the mixture's dissolved oxygen (DO) because the following treatment process was anaerobic. The pre-treatment process was batch operated at room temperature ranging between 20–23°C, and on the third day, the resulting product was screened before being placed in a

feeding tank (25L) that was continuously stirred using a magnetic stirrer. The EGSB (2L), containing glass marbles as the underdrain system, was inoculated with anaerobic granular sludge from a full-scale UASB reactor treating brewery wastewater, untreated PSW and a milk solution. The EGSB was operated continuously at mesophilic temperatures (33–40°C) for 120 days. The membrane tank, which was also continually operated at ambient temperature (20–24°C), was inoculated with raw PSW, tap water and Ecoflush™.

In order to determine the reliability of the variations in the concentrations and removal efficiencies (REs) achieved, a visual outlier detection was implemented using boxplots. No outliers were detected for the membrane tank; therefore, data processing was only performed on the pre-treatment and EGSB processes. A correlation matrix using Heatmaps and density contours was applied to determine if there was a correlation between the REs investigated, and no correlations were observed. The pre-treatment process achieved REs of 44% chemical oxygen demand (COD), 66% fats, oil and grease (FOG) and 53% TSS, respectively, proving to be more effective at removing FOG due to the capability of the Ecoflush™ hydrolysing the hydrocarbon chains in FOG. The organic loading rate (OLR) for the EGSB feed ranged from 200 to 700mgCOD/L.h. The EGSB successfully removed on average 56%, 63%, and 73% of the COD, FOG, and TSS present in the feed. The submerged membrane had the best RE performance obtaining on average 88% COD, 64% FOG and 90% TSS removal with an OLR that fluctuated between 50 and 450mgCOD/L.h. The integrated multi-stage treatment plant achieved overall average REs of 98% COD, 97% FOG, and 99% TSS reducing the content of the treated water to 101mg/L COD, 8mg/L FOG and 7mg/L TSS, i.e. qualities which are comparable to inland surface water, rendering it safe for discharge into the City of Cape Town (CCT) WWTWs.

Keywords

Expanded granular sludge bed (EGSB); fats, oil and grease (FOG); poultry slaughterhouse wastewater (PSW); Submerged membrane bioreactor.

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DEDICATION

To my late grandparents

Timothy and Maggie Bingo,

and

my late aunt,

Mrs. Ruth Spiwe Simbayi

RESEARCH OUTPUTS

The following research outputs represent the contributions by the author of this thesis to the knowledge relevant to poultry slaughterhouse wastewater treatment:

The following International conference proceedings were published for research studies related to this thesis:

- I. **Bingo, M.N.**, Basitere, M. and Ntwampe, S.K.O. Poultry Slaughterhouse Wastewater Treatment Plant Design Advancements: Conference: 16th SOUTH AFRICA International Conference on Agricultural, Chemical, Biological and Environmental Science (ACBES-19), Johannesburg, South Africa, November 18-19, 2019. Pp. 289-294. ISBN 978-81-943403-0-0. DIO: 10.17758/EARES8.EAP1119145.
- II. **Maggie N. Bingo**, Seteno Karabo Obed Ntwampe, Lionel Neddy Aymar Ndeba-Nganongo and Moses Basitere, 2019. Trinal Simulator Stages for Modelling of Poultry Slaughterhouse Wastewater Nutrient Removal. 2019 Innovation Conference on Sustainable Wastewater Treatment and Resource Recovery Nov. 24-28, 2019, Shanghai, China. www.nrr2019.com.
- III. **M. N. Bingo**, M. Basitere, S. K. O. Ntwampe, and D. N. Dlamini, 2021. Trinal Simulator Stages for Modelling a Pilot Scale Poultry Slaughterhouse Wastewater Treatment Plant Using Sumo. 12th Eastern European Young Water Professionals Conference. 31 March -2 April 2021, Riga, Latvia, <http://iwa-ywp.eu/>.

The following DHET- accredited research article was submitted to the Journal of Water Process Engineering for research study related to this thesis:

- IV. **Bingo, M.N.**, Njoya, M., Basitere, M., Ntwampe, S.K.O. and Kaskote, E., 2021, Performance evaluation of an integrated multi-stage poultry slaughterhouse wastewater treatment system. Manuscript Number: JWPE-D-21-01655R1 (**Accepted 06 September 2021**).

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Safety, Risk, Reliability and Quality (Q1)

Water Science and Technology (Q1)

Biotechnology (Q2)

Process Chemistry and Technology (Q2)

Waste Management and Disposal (Q2)

LAYOUT OF THESIS

This thesis comprises of the following chapters:

- **Chapter 1** is an introductory chapter that provides background information about the Cape Town, SA, 2017 water crisis, poultry slaughterhouse water consumption and wastewater characterization, and motivation for the study. It includes the research problem, research questions, aims and objectives, significance of the research and delineation of the study.
- **Chapter 2** is a literature review of the poultry industry in SA, environmental issues associated with PSW, industrial waste discharge charge system (WDCS) and the treatment methods for PSW.
- **Chapter 3** is a brief background of the development of the Activated Sludge Models (ASMs), the basis of the ASMs, an explanation on simulation packages, and a description of the simulation model used in this study (Sumo).
- **Chapter 4** reports on the modelling using Sumo of a proposed design, i.e. the feasibility of a three-stage treatment system.
- **Chapter 5** describes the setup and operation of an integrated multi-stage PSW treatment system consisting of an aerobic pre-treatment tank, an EGSB bioreactor coupled with a submerged membrane. It specifies the operating conditions of each treatment stage, sampling methods, and analytical equipment used to analyse the feed and products streams. This chapter further discusses results relating to the performance of individual treatment stages and that of the overall system.
- **Chapter 6** provides the overall conclusion of this study with recommendations for further research.

All the references used in this study are listed in accordance with the guidelines for research theses for a CPUT master's degree qualification.

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LIST OF ABBREVIATIONS

Abbreviation	Description
ABR	Anaerobic baffled reactor
ACBES-19	Agricultural, Chemical, Biological and Environmental Sciences – 2019
AD	Anaerobic digestion
AL	Aerated lagoon
ANFIS	Adaptive neuro-fuzzy inference system
AOP	Advanced oxidation processes
AS	Activated sludge
As	Arsenic
ASM	Activated sludge model
BOD	Biological oxygen demand (mg/L)
BioERG	Bioresource Engineering Research Group
CCT	City of Cape Town
Cd	Cadmium
CH ₃ CH ₂ COOH	Propionate
CH ₃ COO ⁻	Acetate
CH ₃ COOH	Acetic acid
CH ₄	Methane
CHP	Combined heat and power
C ₄ H ₇ O ₂ ⁻	Butyrate
Cl ⁻	Chloride
CO	Carbon monoxide
CO ₂	Carbon dioxide
COD	Chemical oxygen demand (mg/L)
CPUT	Cape Peninsula University of Technology
Cr ⁶⁺	Chromium
Cu	Copper
DAF	Dissolved air floatation
DAFF	Department of Agriculture, Forestry and Fisheries
DEA	Department of Environmental Affairs
DEADP	Department of Environmental Affairs and Development Planning
DO	Dissolved oxygen (mg/L)
DOI	Digital object identifier
DWAF	Department of Water Affairs and Forestry

DWS	Department of Water and Sanitation
EC	Electrical conductivity (mS/m)
EGSB	Expanded granular sludge bed
EIP	Environmental Integrity Project
FOG	Fats, oil and grease (mg/L)
H ₂	Hydrogen
HDPE	High-density polyethylene
Hg	Mercury
H ₂ O	Water
H ₂ O ₂	Hydrogen peroxide
HRT	Hydraulic retention time (h, days)
H ₂ S	Hydrogen sulphide
HUASB	Hybrid up-flow anaerobic sludge blanket
HUASB-AL	Hybrid up-flow anaerobic sludge blanket aerated lagoon
IMF	Integrated membrane filtration
IPCC	Intergovernmental Panel on Climate Change
K ₂ O	Potassium
MBR	Membrane bioreactor
MF	Microfiltration
MWWW	Municipal wastewater works
N	Nitrogen
Na ⁺	Sodium
NH ₄ ⁺	Ammonium
NH ₃	Ammonia
Ni	Nickel
NO ₂	Nitrite
NO ₃ ⁻	Nitrate
NWA	National Water Act
O ₂	Oxygen
O ₃	Ozone
OLR	Organic loading rate (kg/m ³ .days; gCOD/L.day; gtCOD/L.day)
ORP	Oxidation-reduction potential (mV)
P	Phosphorus
Pb	Lead
PET	Polyester
PES	Polyether-sulfone
PO ₄ ³⁻	Orthophosphate

PSW	Poultry slaughterhouse wastewater
PVC	Polyvinyl chloride
RE	Removal efficiency (%)
RO	Reverse Osmosis
RSM	Response surface methodology
RTI	Research, Technology and Innovation
SAB	South African Brewery
SDIW	Soft drink industry wastewater
SSND	Single-stage nitrification-denitrification
SA	South Africa
SAPA	South African Poultry Association
SGBR	Static granular bed reactor
SO ₄ ²⁻	Sulfate
SRT	Solid retention time (h, days)
SS	Suspended solids (mg/L)
SumoSlang	Sumo simulation language
SWW	Slaughterhouse wastewater
tCOD	Total chemical oxygen demand (mg/L)
TDO	Total dissolved oxygen (mg/L)
TDS	Total dissolved solids (mg/L)
TKN	Total Kjeldahl Nitrogen (mg/L)
TN	Total nitrogen (mg/L)
TP	Total phosphorus (mg/L)
TSS	Total suspended solids (mg/L)
UASB	Up-flow anaerobic sludge blanket
UF	Ultrafiltration
ufMM	Ultrafiltration membrane module
USA	United States of America
UST	Ultrasound Technology
UV	Ultraviolet
VFA	Volatile fatty acids (mg/L)
VSS	Volatile suspended solids (mg/L)
V _{up}	Upflow velocity (m/h)
WC	Western Cape
WCG	Western Cape Government
WDCS	Waste Discharge Charge System
WDL	Waste Discharge Levy

WMC	Waste Mitigation Charge
WWTP	Wastewater treatment plant
WWTW	Wastewater treatment work
3D CFD	Three-dimensional computational fluid dynamics

GLOSSARY/BASIC TERMS AND CONCEPTS

Aerobic digestion – is based on the activity of aerobic bacteria that rapidly consumes the organic matter and produce single-cell proteins, water and carbon dioxide (Kosseva, 2020).

Anaerobic digestion – is a complex transformation process incorporating a series of interdependent biochemical reactions that take place in the absence of oxygen, or through the metabolic pathways of anaerobic microorganisms (Kosseva, 2020).

Biochemical process – is a chemical process that occurs using living organisms, involving biomolecules (Kte'pi, 2011).

Biological oxygen demand (BOD) – is the amount of dissolved oxygen needed for microbial oxidation of soluble, biodegradable matter in an aqueous environment (Singleton and Sainsbury, 2006).

Biodegradable – is a material's ability to decompose after interaction with biological agents (Goswami and O'Haire, 2016).

Catchment – is the area from which any rainfall will drain into watercourses through the surface flow to common points (Department of Environmental Affairs, 2014).

Chemical oxygen demand (COD) – is a measurement of the capacity to consume oxygen during the decomposition of organic matter and the oxidation of inorganic chemicals such as ammonia and nitrate (Abdulsyukor *et al.*, 2021).

Day Zero – is the day municipal taps run dry (Booyesen *et al.*, 2019).

Eutrophication – is the enrichment of water by nutrients, especially nitrogen and/or phosphorus and organic matter, causing increased growth of algae and higher forms of plant life to produce an unacceptable deviation in structure, function and stability of organisms present in the water and to the quality of water concerned, compared to reference conditions (Andersen *et al.*, 2006).

Expanded granular sludge bed (EGSB) reactor – is a modification of an up-flow anaerobic sludge blanket (UASB) that was primarily developed to improve substrate–biomass contact within the treatment by expanding the sludge bed and intensifying hydraulic mixing, and has an up-flow configuration (Mchugh *et al.*, 2003).

Granular sludge – is a well-balanced micro-ecosystem that include all bacterial species necessary for the degradation of the organic pollutants present in the wastewater to which it is exposed. It has specific properties that make it very suitable for upflow wastewater treatment systems (Alphenaar, 1994).

Heterocyclic compounds – are cyclic compounds with the ring containing carbon and other elements, the component being oxygen, nitrogen and sulfur (Siddiquee, 2014).

Heterotrophic microorganisms – are biocatalysts for the biotransformation of organic compounds (Nakamura, 2007).

Hydraulic retention time (HRT) – is the average number of days that a feed stays in a digester and is related to digester capacity (Gould, 2015).

Organic loading rate (OLR) – is the application of soluble and particulate organic matter, and is typically expressed on an area basis as the mass of BOD per unit area per unit time (Washington State Department of Health, 2002).

Pre-treatment – is the initial step in waste management that occurs after waste generation, and a key part of any decommissioning programme and involves a variety of processes applied to liquid and solid wastes (Ojovan and Lee, 2014).

Total suspended solids (TSS) – is all the solid particles in wastewater, effluent, or a natural body of water that will not pass through a filter of a given size; components may include silt, decaying plant and animal matter, industrial wastes, and sewage (Cleveland and Morris, 2015).

Up-flow anaerobic sludge blanket (UASB) reactor – is a suspended-growth reactor that maintains a very high microbial biomass concentration by promoting granulation (Khanal *et al.*, 2017).

CHAPTER ONE

Introduction

1.1. Background of the research problem

Water is a necessity in everyday life, but it has become difficult to continually meet the ever-increasing world water needs over the years due to the exponential growth of the population, economic growth and global warming (Le Page, 2018). Over ten years ago, the Intergovernmental Panel on Climate Change (IPCC), together with other organisations, reported on the developments of global warming, and how it would result in the reduction of precipitation over Northern Africa and the south-western region of South Africa (SA) by the end of the 21st century, which could cause droughts, floods, heatwaves, and temperature changes (Richman & Leslie, 2018). This report was meant to serve as a cautionary report to major cities worldwide that depended heavily on rainfall as a water source. According to the City of Cape Town (CCT) (2018), a huge percentage of Cape Town's water supply emanates from water that has evaporated off the Atlantic Ocean, which rises from the sea's surface to form clouds and ultimately falls in the form of rain. A portion of this rain is captured by fourteen dams that have a combined volume of almost 900 000 000 000 litres. Six large dams (Theewaterskloof, Voëlvlei, Berg River, Wemmershoek and the Steenbras Upper and Lower dams) provide the majority of this capacity (Sinclair-Smith and Winter, 2018). Therefore, it became apparent that additional water sources and management were a requirement, however at the end of the 2014 wet season, all of Cape Town's six dams were filled, and therefore the urgency of the matter was not anticipated (Richman and Leslie, 2018).

In 2017 the Western Cape (WC) Province went through the worst drought ever since 1904. As a result, from November 2016 to February 2018, the CCT gradually enforced six levels of water restrictions on a population of approximately 3.8 million to control usage and avoid "day zero," whereby the supply capacity would fall below the critical level of 13.5%. Domestic consumers were forced to reduce their household water usage from 540L to 280L per day (Booyesen *et al.*, 2019; Richman & Leslie, 2018). By January 2018, the levels of the dams were below 25% capacity. Had these storage levels dropped even further, Capetonians would have had their water consumption restricted to 25-litres-per-person-day, which they would have had to collect from about 200 collection points. Fortunately, rainfall in February, April and May 2018 moved day zero from April 2018 to August 2018 and has been completely avoided to date. However, the continuing threat of a future "day zero" remains (Le Page, 2018; Richman and Leslie, 2018). Building additional dams will prove to be extremely expensive and possibly harmful to the environment; therefore, a tactful alteration in the use and preservation of water resources to promote efficient water usage is obligatory and in agreement with the National Water Act (NWA) (Act 36 of 1998) "*which emphasizes effective management of our water resources*" (Department of Water Affairs and Forestry (DWAF), 2004). Currently, SA's water

consumption is around 16 billion m³/year (GreenCape, 2018), and Figure 1 indicates how sectors in SA contribute to this consumption of water. Agriculture is responsible for 62% of water consumption in the country; therefore, any percentage reduction in consumption of this sector would assist in the conservation of water resources a great deal (Department of Water and Sanitation (DWS), 2015).

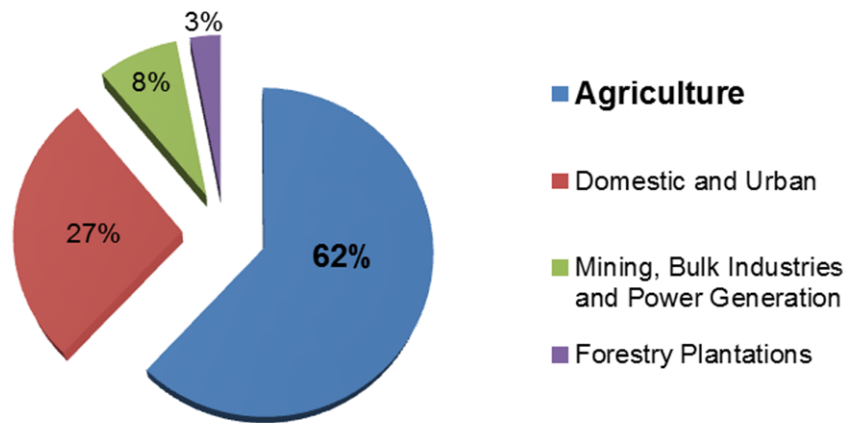


Figure 1.1: Water consumption of Sectors in SA (DWS, 2015)

The poultry industry is water-intensive, with 90% of its water consumption discharged as wastewater due to it being used for cleaning and washing carcasses and meat products, as well as the sanitation and disinfection of equipment and all product processing areas of the slaughterhouse (Molapo *et al.*, 2009; Department of Environmental Affairs and Development Planning (DEA & DP), 2015). On average, water consumption per bird ranges between 15–20L (Molapo, 2009). The wastewater from poultry slaughterhouses is typically categorised by high levels of organic matter, namely chemical oxygen demand (COD) and biological oxygen demand (BOD), as well as pathogens, suspended solids (SS), and fats, oil, and grease (FOG) (Meiramkulova *et al.*, 2020), as indicated in Table 1.1. The presence of organic matter results from blood, fats, urine, and faeces (Al Smadi *et al.*, 2019). The primary nutrients in poultry slaughterhouse wastewater (PSW) are total phosphorus (TP) and total nitrogen (TN). Orthophosphate (PO_4^{3-}) is the most prevalent form of phosphorus (P), and its presence is due to the cleaning and sanitizing of detergents (Bustillo-Lecompte & Mehrvar, 2017; Yaakob *et al.*, 2018). Nitrogen (N) is available in organic form as ammonia (NH_4^+ , $\text{NH}_3 - \text{N}$) and inorganic form as nitrite (NO_2^-) and nitrate (NO_3^-), which is a steady form of N found in water and occurs because of the natural decaying process of biological matter. A high COD value indicates the

presence of high chemical reactions between organic substances, while a high value of BOD indicates the availability of high microbial loads (Yaakob *et al.*, 2018). The presence of chemical elements in PSW is due to the cleaning and sanitizing detergents used for cleaning purposes, and pharmaceuticals resulting from veterinary purposes (Bustillo-Lecompte & Mehrvar, 2017). However, wastewater characteristics vary depending on the type of bird being slaughtered, the number of birds being processed, the water consumed per bird, and the type of process being implemented (Basitere *et al.*, 2017; Al Smadi *et al.*, 2019). The characteristics of the PSW influences the selection of the treatment process to be used (Bustillo-Lecompte & Mehrvar, 2015).

Table 1.1: Typical characteristics of wastewater from a poultry slaughterhouse in the WC, CCT discharge by-laws and DWS standards of effluent discharge (Council of the City of Cape Town, 2014; DWS, 2017; Williams, 2017)

Parameter	Poultry Slaughterhouse Wastewater	CCT Discharge By-Laws	Standards for Effluent Discharge
	Range	Not to exceed	General Limits
pH	6.5 – 8.0	12.0	5.5 – 9.5
Alkalinity (mg/L)	0 – 489	-	-
Total COD [tCOD] (mg/L)	2 133 – 9 695	5000	75
Soluble COD (mg/L)	595 – 1 526	-	-
BOD (mg/L)	1 100 – 2 750	-	-
Total Kjeldahl Nitrogen [TKN] (mg/L)	77 – 352	-	-
NH ₄ ⁺ - N (mg/L)	29 – 51	-	6
PO ₄ ³⁻ - P (mg/L)	8 – 27	25	10
FOG (mg/L)	131 – 684	400	2.5
Total dissolved oxygen [TDO] (mg/L)	372 – 936	-	-
Total Suspended Solids [TSS] (mg/L)	315 – 4 992	1000	25
Volatile Suspended Solids [VSS] (mg/L)	275 – 1 200	-	-
Soluble proteins (mg/L)	0 – 368	-	-
Volatile Fatty Acids [VFA] (mg/L)	96 – 235	-	-
NO ₃ ⁻ - N (mg/L)	0 – 2.903	-	15

A portion of the treated PSW is re-used for washing trucks or floors (Molapo, 2009), while most of it is generally disposed into streams, rivers and lakes, municipal wastewater treatment plants (WWTPs), and spraying it on grass or cropland (Environmental Integrity Project (EIP), 2018). The disposal of usually untreated slaughterhouse wastewater is detrimental to the health and safety of the environment due to the high content of biodegradable organic matter and nutrients (Bustillo-Lecompte & Mehrvar, 2017). When this wastewater is discharged into

channels and municipal treatment systems, it may result in algae growth, dissolved oxygen (DO) depletion, and consequently, resulting in the death of fish and other marine life, and the spreading of water-borne diseases (EIP, 2018; Yaakob *et al.*, 2018). When sprayed on land, this wastewater can lead to soil and land pollution (Molapo, 2009). Additionally, the progressively stricter standards for effluent discharge worldwide have put an urgency on the advancement of wastewater treatment technologies (Bustillo-Lecompte & Mehrvar, 2015). In SA, the treatment requirements are determined by the quantified discharge limitations indicated by the Water Services by-laws as per the Municipal Systems Act, and these by-laws differ from one province to the next (Molapo, 2009). The government has levied a water resource management charge for sectors such as irrigated agriculture, mining, and forestry to reduce water consumption (Letsoalo *et al.*, 2007). The DWS (2007) developed the national pricing strategy, including the Waste Discharge Charge System (WDCS), founded on the polluter pays principle. This charging system was proposed to create a financial incentive for polluters who reduce their waste and use water resources in an optimum manner. However, slaughterhouses are fined when their wastewater does not meet the required effluent discharge standards specified by municipalities. As indicated by Table 1.1 substantial treatment is necessary to meet the required discharge standards for wastewater treatment effluent.

Cape Town's most recent water crisis, together with the global impact of climate change, has led to growing interest and investment from science and policy researchers to ensure water security (Green *et al.*, 2015). This is further emphasised in the Master Plan developed by the DWS in 2018, highlighting the importance of safeguarding the maintainable use of water resources and adequate water availability for current and future requirements.

1.2. Statement of the research problem

Slaughterhouse waste has been acknowledged as the most challenging food waste type to manage in the Western Cape due to its harmful nature. Between 2015 and 2016, poultry wastewater was 43% of the total slaughterhouse waste generated in the WC, as indicated by Figure 1.2 (Western Cape Government (WCG), 2016).

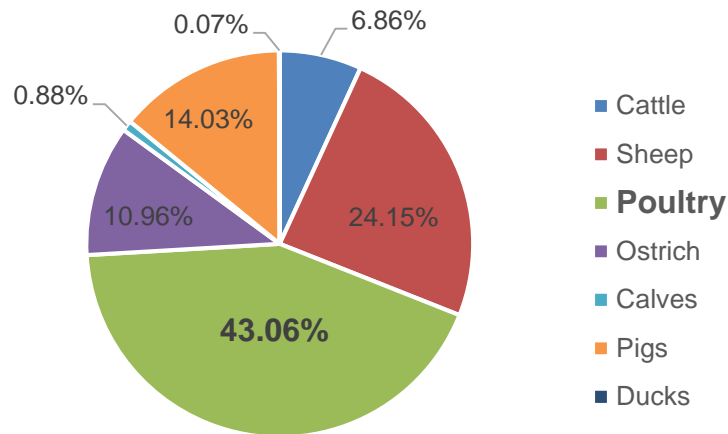


Figure 1.2: Slaughterhouse waste in the WC during 2015/2016 (WCG, 2016)

Widespread research has been done experimentally on lab-scale PSW bioreactors, while few studies have been reported using mathematical modelling to simulate this treatment process. Mathematical modelling is a simplified representation of reality intended to understand the processes being investigated and is a recognised tool in engineering practice to assist with plant design, development, operation, control and research purposes through focused simulations (Gazsó *et al.*, 2017). There are various mathematical models and software possibilities available; however, Gazsó *et al.* (2017) recommend Sumo©, a full-featured wastewater treatment process simulator software developed by Dynamita (Environmental Services, Sigale, Provence-Alpes-Côte d'Azur) for environmental models, to be ideal for investigating key elements of treatment systems and detecting anaerobic digester system reactions for long-term variations of the feed load. Sumo is further recommended by Kolovos *et al.* (2016) since it has a user-friendly interface and open-source process codes, equipping the user with a better understanding of the calculations behind the model. To top it all, it is highly cost-effective in that it has a once-off licensing fee, unlike other software that has an annual licensing fee. Therefore, this current study investigated the possibility of predicting the performance of a treatment process treating PSW using SUMO software.

1.3. Research questions

- Can the Sumo Simulator successfully simulate the performance of lab-scale anaerobic digesters treating PSW regarding the removal of COD, FOG, TSS, BOD, NH₃ and PO₄³⁻?

- Based on the simulation results, does the quality of the treatment plant's effluent meet the standards for effluent discharge as stipulated by CCT by-laws?
- Can the proposed integrated multi-stage treatment system, consisting of an aerobic pre-treatment tank, an expanded granular sludge bed (EGSB) coupled with a submerged membrane tank, successfully treat PSW with the effluent meeting the DWS standards for effluent discharge?
- What is the performance of each treatment stage with regards to COD, FOG, and TSS removal?
- What is the performance of the overall treatment system with regards to COD, FOG and TSS removal?

1.4. Aim and objectives

This research aimed first to investigate the development of a basic model to simulate the performance of lab-scale anaerobic digesters treating PSW using the Sumo19 Simulator software. Thereafter, the performance of a three-stage PSW treatment system was investigated. The obtained effluent results were compared to the CCT discharge by-laws and the DWS standards of effluent discharge to determine whether the treated PSW would be suitable to discharge into the CCT WWTWs or on-site re-use. In realising this aim, the primary removal efficiency parameters, COD, FOG and TSS, were evaluated.

1.5. Significance of the research

If this study is successful, it could assist poultry producers to reduce their usage of potable water and promoting wastewater management by reducing the number of contaminants discharged into waterways, those associated with land application, and thus safeguarding the ecosystem. This would be in agreement with the National Water Act 36 of 1998 and falls within the Cape Peninsula University Technology (CPUT) Research, Technology and Innovation (RTI) cluster on Bioeconomy and Environmental sustainability. This research project is further aligned with the 2030 Sustainable Development Goals on clean water and sanitation, which focuses on the assurance and contribution to the availability and sustainable management of water and wastewater globally.

1.6. Delineation of the research

This research study will not focus on the following:

- Cost evaluation of the system or scale-up studies.

- The production of biogas.
- The submerged membrane fouling effects.
- The kinetic growth of microscopic organisms in the anaerobic treatment stage.

CHAPTER TWO

Literature review

Part of this chapter was published as a conference proceeding in 2019 as:

Bingo, M.N., Basitere, M. & Ntwampe, S.K.O: Poultry Slaughterhouse Wastewater Treatment Plant Design Advancements: Conference: **16th SOUTH AFRICA International Conference on Agricultural, Chemical, Biological and Environmental Science (ACBES-19)**, Johannesburg, South Africa, November 18-19, 2019. Pp. 289-294.

DOI: [10.17758/EARES8.EAP1119145](https://doi.org/10.17758/EARES8.EAP1119145)

2.1. The poultry industry in South Africa

During the past few decades, the consumption of poultry meat has increased in many countries around the world, and this is due to poultry having a rapid growth rate (Barbut, 2002), poultry meat forming part of a balanced diet for it is a valuable source of protein (Roberts, 2017), and being inexpensive for a majority of the low-income families in developing countries (Tan *et al.*, 2018). In SA, the poultry industry is the largest sector within the agricultural sector regarding the production value. According to the Department of Agriculture Forestry and Fisheries (DAFF) (2017), the poultry industry generated a gross value of R38.6 billion from 2015 to 2016, which made up 15.6% of the total gross value of the agricultural produces. The DAFF (2017) further points out that the total poultry meat production consists of 93.6% broiler meat and 6.4% which is made up of mature chicken slaughter, small-scale broiler, geese, turkey, duck and guinea fowl meat products. Figure 2.1 depicts the distribution of broilers in SA during the year 2016. The provinces that produced the largest percentage of broiler meat were North West, Western Cape and Northern Cape, and Mpumalanga.

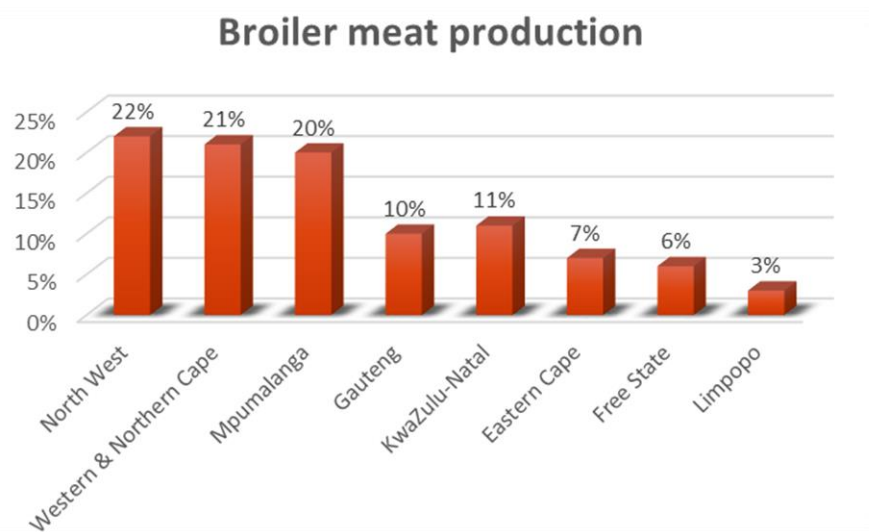


Figure 2.1: Distribution of broilers in SA during 2016 (DAFF,2017)

In 2017 a total of 927.1 million broilers were slaughtered (South African Poultry Association (SAPA), 2017), and as outlined herein, the average water consumption per bird ranges between 15–20L. As a result, the total water consumption of broiler slaughterhouses in 2017 can be calculated to have ranged between 13.9–18.5 million m³, and this gives evidence that slaughterhouses are large consumers of freshwater. The majority of the water consumption is used for cleaning the reception and slaughter areas, scalding and de-feathering, evisceration,

chilling, general washing and by-product processing. Nevertheless, water consumption varies depending on the size of the slaughterhouse facility, the number of birds being slaughtered, and the process and type of technology used at the facility (DEA & DP, 2015). Smaller facilities typically have a higher water usage, for they function on a stop-start basis and do not have the necessary workforce to handle the demanding water management practices. Thus, not much attention is given to these practices (WCG, 2015), which leads to large volumes of high-strength slaughterhouse wastewater consisting of high organic matter containing N and P (Basitere *et al.*, 2016). To make matters worse, the composition and concentration of the wastewater fluctuate depending on the processes being carried out (Marcos *et al.*, 2017). If not managed properly, this can harm the environment due to wastewater disposal and discharge (Harvey *et al.*, 2017).

2.2. Environmental issues associated with poultry slaughterhouse wastewater

PSW is commonly disposed into streams, rivers and lakes, municipal wastewater treatment works WWTWs, and by spraying it on grass or cropland (EIP, 2018). Unfortunately, PSW is considered highly polluted due to the presence of BOD, COD, TSS, blood and nutrients from the slaughtering of birds and cleaning of the facilities (Yaakob *et al.*, 2018). This wastewater can lead to water, soil and air pollution if not treated before disposal (Molapo, 2009). When PSW is discharged into water bodies, the biodegradable organic compounds may effect a reduction of the DO present in surface water resulting in the death of aquatic life and bad odour (Gerber *et al.*, 2007; Yaakob *et al.*, 2018). Macronutrients might cause eutrophication (Figure 2.2), which is the enrichment of water by N, P and organic matter, triggering an amplified growth of algae and higher forms of plant life. This destabilizes the organisms present in the receiving water and thus affects the quality of the water and ecosystem, causing the spread of waterborne disease (Andersen *et al.*, 2006; Yaakob *et al.*, 2018). Therefore, it is a requirement that industrial effluent undergo treatment before being discharged into WWTWs to avoid damaging equipment, which could result in the ineffectiveness of the WWTWs (Department of Environmental Affairs (DEA), 2014). According to the DWS (2018), only 56% of SA's municipal WWTWs are in working condition.

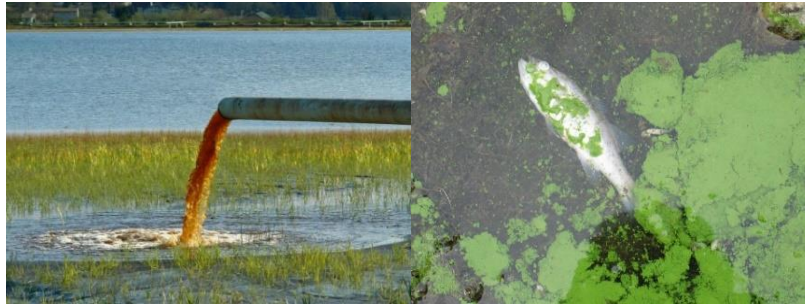


Figure 2.2: Discharging wastewater into water bodies results in eutrophication (Scannone, 2016).

When spraying PSW on croplands, it should be considered that N has been identified as a pollutant for soils because its accumulation in the soil can become toxic for plants (Gerber *et al.*, 2007). Governments worldwide have implemented legislative regulations to preserve water resources and minimize pollution (Letsoalo *et al.*, 2007).

2.3. Industrial effluent waste discharge charge system

In SA, the National Water Act (NWA) (Act 36 of 1998) and the Water Services Act (WSA) (Act 108 of 1997) govern the regulations of water, wastewater management practices and industrial discharge standards (Basitere, 2017). The NWA provides the standards for purifying wastewater before discharge, whereas the WSA stipulates the water supply and effluent discharge guidelines and tariffs for the area. Even though most of the abattoirs pre-treat their wastewater prior to discharging it into municipal sewers, abattoirs generally struggle to meet the municipal by-law discharge standards regarding SS and FOG (Pocock & Joubert, 2017). These industrial discharge standards vary from province to province because they are stipulated by the local municipality (Molapo, 2009). Therefore, the poultry slaughterhouse located in the WC must comply with the wastewater and industrial effluent discharge standards set out by the CCT municipality (Basitere, 2017), and they are listed in Table 1.1. To ensure sustainable water use in SA, the DWA has developed the Waste Discharge Charge System (WDCS) that forms part of the National Pricing Strategy, established under the NWA (Act 36 of 1998). The WDCS is intended to encourage the reduction of waste and the conservation of water resources by financially incentivising the waste dischargers who reduce their waste and optimally use water resources, and penalising them if their wastewater does not meet the effluent discharge standards as stipulated by the municipalities. Therefore, the WDCS is founded on the polluter pays principle (Pegram *et al.*, 2014). This charging system applies to surface water and groundwater resources and consists of two different water use charges: 1) the Waste Mitigation Charge (WMC), which is intended to recover the costs of measures undertaken to alleviate the impacts of discharging waste into catchment water resources, and

2) the Waste Discharge Levy (WDL) that incentivizes dischargers who adopt processes which reduce their waste discharge load (DWAf, 2007).

The WDCS will be imposed on water quality variables based on the type of water discharge source, the nature of the water being discharged and the cost-effectiveness of monitoring different variables. Some of these water quality variables are listed below (DWS, 2015):

- Nutrients: nitrate (NO_3^-), phosphate (PO_4^{3-}) and ammonium (NH_4^+)
- Salinity: total dissolved solids (TDS), electrical conductivity (EC), chloride (Cl^-), sodium (Na^+) and sulfate (SO_4^{2-})
- Heavy metals: arsenic (As), cadmium (Cd), chromium (Cr^{6+}), copper (Cu), mercury (Hg), lead (Pb), nickel (Ni) and zinc (Zn)
- Organic material: chemical oxygen demand (COD), biological oxygen demand (BOD)

A great deal of time, energy, and resources focus on integrated wastewater management including water conservation. This involves adopting the latest technologies to reduce freshwater consumption and increase reuse practices to achieve zero effluent discharges (Agricultural Sector Education and Training Authority (AgriSETA), 2018).

2.4. Treatment methods for poultry slaughterhouse wastewater

Slaughterhouse wastewater (SWW) treatment methods are similar to municipal WWTWs and consist of the following step: 1) pre-treatment, 2) primary, 3) secondary, and 4) tertiary treatment systems (Mbulawa, 2017). These methods are considered highly complex systems in that they consist of physico-chemical, biological and biochemical processes, and these treatment plants are considered dynamic due to significant changes in influent wastewater flowrate and composition (Gazsó *et al.*, 2017). Thus, the treatment system for PSW requires a combination of processes that will treat and disinfect the wastewater on-site (Bustillo-Lecompte & Mehrvar, 2017). The selection of the treatment process not only depends on the characteristics of the wastewater but also on compliance with regulations and the technology available (Bustillo-Lecompte & Mehrvar, 2015). Their treatment performance depends on several factors such as influent characteristics of the wastewater (such as substrates composition, macro and micronutrients, toxic compounds), operational conditions (organic loading rate (OLR), pH, hydraulic and sludge retention times (HRT and SRT), temperature variations, biomass concentration and doses of chemicals (Yetilmezsoy *et al.*, 2015).

2.4.1. Pre-treatment methods

The pre-treatment options typically practised in poultry slaughterhouses are screening, settling, catch basins, and flotation systems. The screening process is usually the first, simplest and most inexpensive form of wastewater treatment that recovers offal materials (feathers, meat particles, bones, protein, and FOG) generated during the poultry slaughtering process, which are valuable by-products for the poultry rendering industry (Mbulawa, 2017; Pocock & Joubert, 2017). It has been reported that screening can remove up to 60% of the solids and 30% of the BOD provided the SS are reduced by 50–70%; however, overloading of the screen or under-sizing of screen gaps can lead to mechanical failures and blanking. The solids removed by the screening process are dewatered and compacted to minimize moisture content and volume, and the product is treated as solid waste (Bustillo-Lecompte & Mehrvar, 2015; Mbulawa, 2017). There is a wide range of screens available such as stationery or incline screens, rotary cylindrical screens, brushed screens and vibrating screens; therefore, the particle size of the solids to be removed determines the size and type of the screen to be used (Pocock & Joubert, 2017). See Figure 2.3 as an example.



Figure 2.3: Rotary cylindrical screen used at a poultry slaughterhouse in Virginia, United States of America (USA) (Burrows and Reidy, 2018).

Once the coarse solids have been removed, the wastewater still consists of fine SS and FOG, and both catch basins and settling tanks are capable of removing these materials from the wastewater through gravity. Particles that are denser than water will sink to the bottom, and the resulting sludge is removed using a scraper, whereas fine solids and FOG rise to the water's surface and are removed using a skimmer (Mbulawa, 2017). These treatment systems have been reported to achieve a 30% BOD and 70% soluble solids REs (Bustillo-Lecompte & Mehrvar, 2015). The scum that is skimmed off at the surface can generate an income as it is used as animal feed or processed as raw material in manufacturing soaps and cosmetics. The

sludge removed from the bottom is treated further, and the resulting clearer water leaves the system from the top of the tank for further treatment (Pocock & Joubert, 2017). See Figure 2.4.

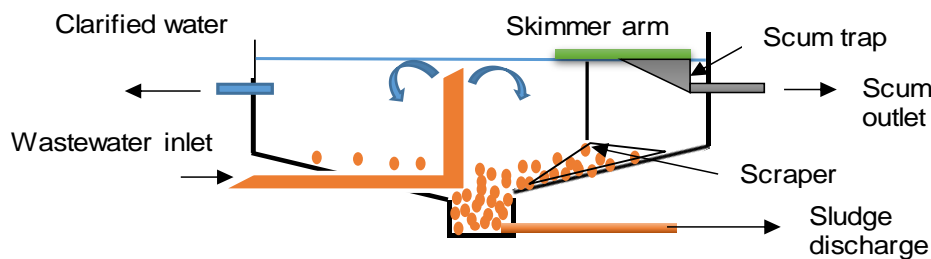


Figure 2.4: Schematic illustration of a settling tank.

2.4.2. Primary treatment process

In meat processing plants, the physico-chemical treatment is a primary treatment process that further separates the solids from liquids by removing TSS and FOG from the wastewater (Basitere *et al.*, 2017), reducing COD and BOD content (Mbulawa, 2017). Dissolved air floatation (DAF) systems are commonly used as a primary treatment process for PSW, even though there are systems that can obtain similar results at a low-cost (Mittal, 2006). In the DAF system, a portion of the treated wastewater is recycled from a point downstream of the DAF, retained for a few minutes in a pressure vessel for mixing and air saturation to occur, and then injected through the bottom of the DAF unit. Once the pressure drops, causing the air to come out of solution, fine bubbles form and carry light solids and FOG to the surface, where the scum is skimmed off (Banks & Wang, 2006). The solid materials are either discharged into sewer systems or onto agricultural land (Pocock and Joubert, 2017b). In order to enhance the performance of the DAF, coagulants (e.g. aluminium sulfate and ferric chloride) and/or flocculants (e.g. polymers) are mixed with the pre-treated wastewater before being fed into the DAF unit in order to promote protein clattering, precipitation and fat flotation. The REs of COD and BOD generally range from 30–90% and 70–90%, respectively, and large amounts of nutrients are removed. However, the disadvantages of the DAF are systematic malfunctioning and insufficient TSS separation (Bustillo-Lecompte & Mehrvar, 2015). Figure 2.4 gives a layout of the DAF system.

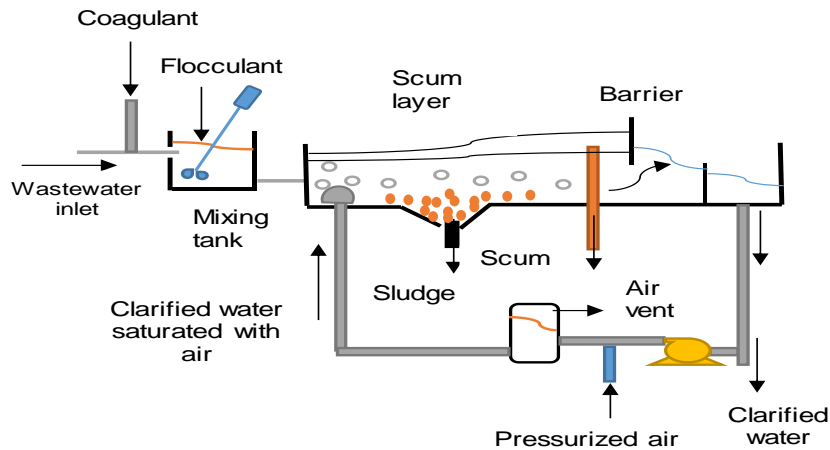


Figure 2.5: Schematic diagram of a DAF clarifier unit in operation.

2.4.3. Secondary treatment systems

The biological treatment system is applied as a secondary treatment process, and its main purpose is to reduce the concentration of organic compounds employing microorganisms (Basitere *et al.*, 2017). Biological treatment processes are classified under two categories, namely anaerobic and aerobic treatment systems. The ideal biological treatment for PSW is anaerobic treatment because of its advantage in successfully treating slaughterhouse wastewater with less complex equipment necessities, high COD removal, and biogas production, which has the prospect of offsetting the energy cost of running the slaughterhouse treatment systems (Al Smadi *et al.*, 2019; Bustillo-Lecompte & Mehrvar, 2017). Even though anaerobic treatment is ideal for PSW, anaerobically treated wastewater contains solubilized organic matters and nutrients, and these can be successfully treated using aerobic processes. As a result, aerobic treatment, which operates at rates higher than anaerobic treatment methods, is applied as post-treatment for anaerobic effluent (Bustillo-Lecompte & Mehrvar, 2017). The anaerobic and aerobic treatment systems are discussed further in the following sections.

2.4.3.1. Anaerobic treatment

The primary driver for anaerobic digestion (AD) is manure and/or sludge (Williams, 2017). A cluster of microorganisms catabolizes the AD of complex polymers in PSW through a biochemical process that consists of four stages: hydrolysis, acidogenesis, acetogenesis and methanogenesis. Hydrolysis is considered the first stage and involves the degradation of complex polymers (i.e. carbohydrates, proteins and fats) into sugar, amino acids and long-chain fatty acids (Manyi-Loh *et al.*, 2013). Due to the formation of non-desirable volatile fatty

acids (VFA), hydrolysis is considered to be the rate-limiting step for organic substrates degradation (Yuan and Zhu, 2016) because the increased production of VFA lowers the pH causing the process to stop. Therefore, the pH must be maintained at around 7.0–7.2 (Irshad *et al.*, 2016). The hydrolysis products are then converted into CO₂, hydrogen (H₂), NH₃, H₂S, alcohols and VFAs (i.e. short-chain fatty acids, namely acetate (CH₃COO⁻), propionate (CH₃CH₂COOH) and butyrate (C₄H₇O₂⁻)) by fermentative or acidogenic bacteria, and this second stage is called acidogenesis (Basitere, 2017; Williams, 2017). During this third stage, acetogens convert organic acids and alcohols into more CH₃COO⁻, CO₂ and H₂ to maximize the production of methane (CH₄) (Manyi-Loh *et al.*, 2013). Williams (2017) mentions that there is a possibility of degrading CH₃COO⁻ when there are sulfate-degrading organisms present and that the formation of CH₃COO⁻ can be hindered by the accumulation of H₂ generation during the acetogenesis stage. Fortunately, methane-forming bacteria consume the H₂ to generate CH₄. The fourth and final stage of AD is methanogenesis where CH₄ is produced by either converting acetic acid (CH₃COOH) molecules using acetrophic methanogens or reducing CO₂ by hydrogenotrophic methanogens (Manyi-Loh *et al.*, 2013). The AD biochemical process stages are summarised in Figure 2.6.

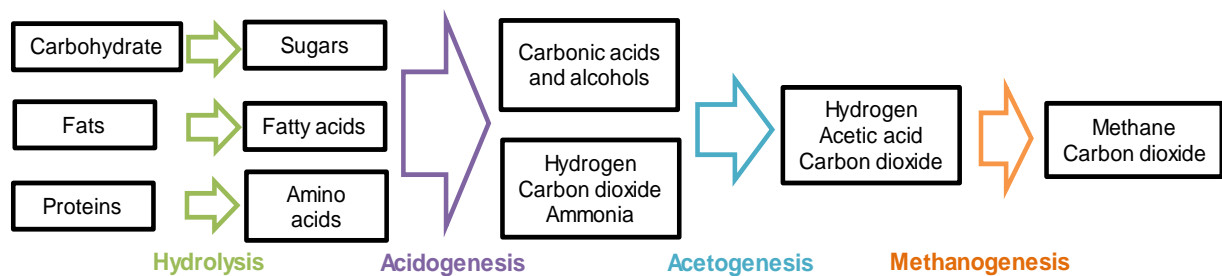


Figure 2.6: The four process stages of AD.

The biogas produced by the AD process contains 50–75% CH₄, 25–45% CO₂, and traces of carbon monoxide (CO), H₂S, NH₃, oxygen (O₂) and water vapour (Manyi-Loh *et al.*, 2013). Biogas can be utilised as combustible fuel in combined heat and power (CHP) gas engines or processed further to produce natural gas-quality biomethane. The remaining material after AD can be used as fertilizer for the bio-augmentation of agricultural soil (Williams, 2017). The advantages of anaerobic systems also include low sludge production and minimal energy requirements with the potential for nutrients and biogas recovery (Bustillo-Lecompte & Mehrvar, 2015). There is a wide variety of anaerobic treatment processes available, lagoons, anaerobic baffled reactor (ABR), up-flow anaerobic sludge blanket (UASB) reactor, static granular bed reactor (SGBR), and expanded granular sludge bed (EGSB) reactor, to name a

few. Each process has distinctive treatment advantages and operational restrictions (Basitere *et al.*, 2017).

Lagoons are easily constructed and operated; however, they require ample space, and it is difficult to capture the biogas produced (Al Smadi *et al.*, 2019). In ABRs, there is an increased contact time between the wastewater and active biomass due to the successions of compartments and baffles where the influent flows under and over from the inlet to outlet, resulting in higher biodegradation occurrence (Bustillo-Lecompte & Mehrvar, 2015). Al Smadi *et al.* (2019) assessed the performance of a 10.45L ABR treating SWW at two different temperatures for 152 days. When the ABR was operated at temperatures ranging from 15–23°C for the first 103 days, it achieved an average COD and TSS RE of 70% and 33%, respectively. During the remaining 49 days the reactor was operated at 40°C, and this resulted in the REs increasing to 90% for the COD and 44% for TSS.

The UASB reactor is one of the most common high-rate AD reactors used for treating wastewater; unfortunately, substantial pre-treatment of the wastewater is required before being fed into the reactor because of its sensitivity to fats and other organic solids (Al Smadi *et al.*, 2019). The EGSB reactor is a variation of the UASB reactor and is widely used due to its increased organic loading rates (OLRs), gas production, efficient removal of soluble pollutants, and improved mixing inside the reactor (Yetilmezsoy *et al.*, 2015). Williams (2017) conducted a study where a lab-scale EGSB anaerobic digester treating PSW was successfully operated for 172 days. With an average HRT of 49.8h and OLR of 3gCOD/L.day, the EGSB achieved an overall RE of 69% tCOD, 98% TSS and 92% FOG. The SGBR is also founded on the design of the UASB but with a down-flow configuration meant to ease the separation of wastewater, solids, and biogas. These technologies have been extensively applied in high-strength wastewater treatment (Yang *et al.*, 2015). In a study conducted by Rinquest *et al.* (2019), the performance of a lab-scale PSW treatment system was evaluated. The treatment system consisted of an SGBR, a single-stage nitrification-denitrification (SSND) bioreactor and an ultrafiltration membrane module (ufMM). The average tCOD, TSS, BOD and FOG REs obtained by the SGBR were 80%, 95%, 89% and 80%, respectively, over 138 days. In this study, the hydraulic retention times (HRTs) ranged from 24 to 96h and OLRs from 0.73 to 12.49gCOD/L.day. Figure 2.6 is an illustration of a UASB bioreactor.

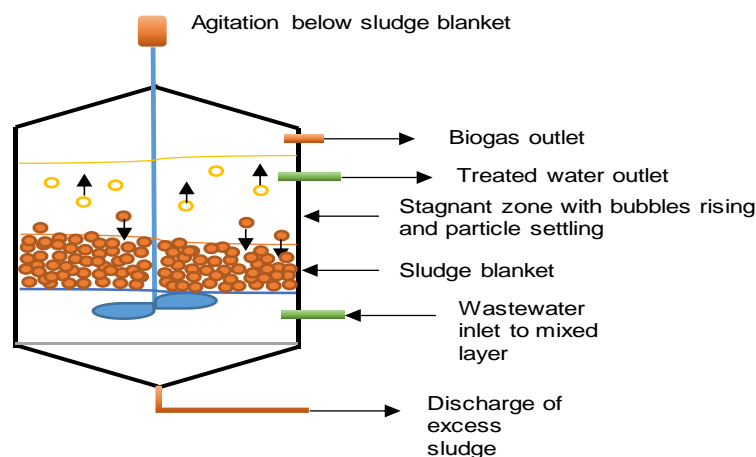


Figure 2.7: Schematic diagram of a UASB bioreactor.

2.4.3.2. Aerobic treatment

Aerobically treated effluent still does not meet the required discharge standards due to the presence of solubilized organic matter, nutrients and pathogens, which can be successfully treated by aerobic treatment methods (Bustillo-Lecompte & Mehrvar, 2017). Aerobic treatment has a high energy requirement due to aeration and high sludge production (Gomes *et al.*, 2018). In aerobic systems, organic substrates are degraded by microorganisms that need free DO; therefore, O_2 is administered by either mechanical aerators or compressors with diffusers or by passing the effluent down a trickling filter to come in contact with atmospheric O_2 (Molapo, 2009). The aeration of the reactor is effected by the concentration of the DO, HRT, OLR, pH, temperature and toxic substances (Irshad *et al.*, 2016). Aerobic digestion is a two-step process; in the first step, a culture of heterotrophic microorganisms consumes and removes a portion of the organic matter present in wastewater to produce new microorganisms, thus increasing biomass. The residual organic matter is directed into metabolic energy and oxidized to form CO_2 , H_2O and soluble inert material, providing energy for production and life support functions. Once the external source of organic matter is depleted, the microorganisms will commence endogenous respiration where cellular material is oxidized to fulfil the life support energy requirements, which is the second step of aerobic digestion. If this process is allowed to continue for a prolonged period, the total amount of biomass will be substantially high (Roš and Zupančič, 2002). To sum it up, in aerobic treatment, carbohydrates present in the wastewater are oxidised to form CO_2 . In contrast, nitrogenous wastes form NO_3^- and SO_4^{2-} (Molapo, 2009), and according to Gallert and Winter (2005), the degradation of sulphur-containing amino acids or heterocyclic compounds leads to the formation of NH_3 and hydrogen sulphide (H_2S). There is a variety of aerobic treatment methods available such as aerobic lagoons, activated sludge (AS) processes (extended

aeration, oxidation ditches, and sequencing batch reactors), and trickling filters (Pocock & Joubert, 2017).

Aerobic lagoons are large, shallow ponds where the sunlight, algae, bacteria and O₂ interact to treat wastewater (Bustillo-Lecompte and Mehrvar, 2015). This process takes between 2-6 days for complete treatment, and can achieve up to 90% reduction of BOD (Irshad *et al.*, 2016). A study on the RE of a UASB and a hybrid UASB (HUASB), both coupled with an aerated lagoon (AL), was conducted by Daud (2016). To do so, three reactors were installed, namely HUASB-AL (R1), HUASB-AL (R2) and UASB-AL (R3). The temperature of both R1 and R3 were maintained at 26±3 °C, while that of R2 was operated at 50±5 °C. The average REs achieved by each arrangement were 90–95% COD, 90–93% BOD, 46–86% TSS, 29–49% NH₃ – N and 82–84% TP.

In the AS process, screened, pre-settled effluent is mixed with small quantities of biologically active sludge in small amounts before being agitated in the presence of oxygen in an aeration tank (Irshad *et al.*, 2016). This process is meant to remove soluble and insoluble organic matter and transform this material into a flocculent microbial suspension that settles in a clarifier (Bustillo-Lecompte and Mehrvar, 2015). Treated wastewater is displaced by incoming effluent into clarifying tanks where some of the settled sludge is recycled to the aerated basin to maintain the microbial culture in peak conditions, as depicted in Figure 2.8. At the same time, the rest is disposed into landfill sites or spread over agricultural land after further treatment. At this stage, the treated wastewater can be discharged into water bodies (Pocock & Joubert, 2017). The AS processes are considered cost-effective treatment methods for SWW, which uses either the application of the adsorption or oxidation of organic matter. Nevertheless, these systems have been reported to produce inadequate settling flocs when treating SWW due to fats, and low DO levels and require extended aeration to minimize sludge production (Bustillo-Lecompte & Mehrvar, 2015). Alfonso-Muniozguren *et al.* (2018) conducted a study on a lab-scale wastewater treatment plant treating abattoir wastewater. This treatment plant consisted of an AS process, with a 24h HRT and a 13 day SRT, followed by filtration and ozonation systems. The average REs obtained by this treatment plant for COD, BOD, TSS and P were 93%, 98%, 99% and 98%, respectively, resulting in the effluent meeting the discharge standards without requiring further treatment. Trickling filters consist of tanks containing porous media with a high surface volume ratio, where wastewater is fed from the top of the tank. The organic matter and N₂ remaining in the influent encourage bacteria growth on the media (Molapo, 2009). The maximum RE of BOD attained by this treatment

process is 90%, and it further removes the residual SS; however, the disadvantages of these systems include blocking, high capital costs and large area requirements and treating wastewater using anaerobic lagoon together with trickling filters results in 74%, 73% and 69% REs of BOD, COD and FOG (Irshad *et al.*, 2016). Tanikawa *et al.* (2016) evaluated the performance of a lab-scale treatment system consisting of an ABR and an aerobic trickling filter, treating lipid-rich wastewater. The overall COD RE obtained by this treatment system was 98%. A typical AS wastewater treatment system is illustrated in Figure 2.8.

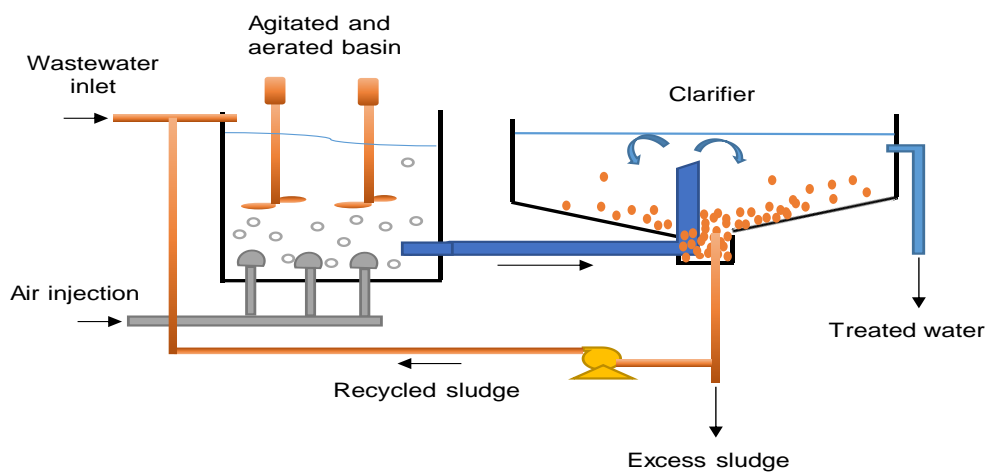


Figure 2.8: An illustration of an AS wastewater treatment system.

2.4.4. Tertiary treatment methods

Suppose the purpose of the treatment process is to recycle the treated water within the slaughterhouse for poultry meat processing. In that case, tertiary treatment processes can be implemented (Muro *et al.*, 2012) to remove suspended or dissolved substances that could not be entirely removed by the secondary treatment processes (Kang *et al.*, 2007). Tertiary treatment processes, also known as the polishing stage (Williams, 2017) that are utilized for treating PSW include advanced oxidation processes (AOPs) and membrane filtration systems such as membrane bioreactors (MBRs), and these processes are considered to be highly expensive (Molapo, 2009). The AOPs are founded on physico-chemical processes that produce powerful oxidative species, which enables the oxidation and degradation of organic matter and the deactivation of pathogens without adding additional chemicals to the system that might result in the formation of harmful by-products. Additional advantages of these systems include low treatment time and high reaction rates. AOPs comprise gamma radiation, ozonation, ultrasound technology (UST), ultraviolet-radiation and hydrogen peroxide

(UV/H₂O₂), ultraviolet-ozone (UV/O₃), and photo-catalysis (Bustillo-Lecompte & Mehrvar, 2017; Litter & Quici, 2010).

Membrane filtration is a pressure-driven process that uses semi-permeable membranes to separate small particles, such as pathogens, from wastewater and is considered to be ideal for disinfection and water polishing to a point where the treated water meets the standards of potable water for they are exceptional at removing residual N and P. A few membrane processes are run in semi-batch cycles for the rate at which the wastewater is added is the same as the rate at which the permeate is withdrawn. Even though membrane processes have a lower carbon footprint when compared to conventional filters, which utilize chemicals, the cost of the membrane life cycle is exorbitant. Membrane filtration does not require temperature monitoring; however, the presence of FOG may result in the formation of thick biofouling layers on the surface of the membrane, thus decreasing the permeation rate of the wastewater (Rinquest, 2017, Williams, 2017, Baker *et al.*, 2020). The two most frequently applied membrane filtrations for wastewater treatment are microfiltration (MF) and ultrafiltration (UF) due to the affordability of the membrane life cycle cost (Rinquest, 2017). For the past 20 years, UF has been extensively used in the food processing industry, for it is capable of providing a barrier for the removal of pathogens which could be critical for recycling PSW (Sardari *et al.*, 2018). In a study conducted by Basitere *et al.* (2017), the performance of a bench-scale treatment plant consisting of a mesophilic SGBR coupled with a UF membrane treating PSW was evaluated. The average REs obtained by the UF membrane were 64% COD, 88% TSS and 48% FOG. Sheldon and Erdogan (2016) evaluated the treatment of soft drink industry wastewater (SDIW) using a multi-stage anaerobic/aerobic MBR and determined that the MBR could remove up to 87% of the COD present in SDIW.

Nonetheless, to upscale these treatment systems for industrial purposes, process modelling needs to be explored to efficiently evaluate the recommended treatment systems' viability.

CHAPTER THREE

Mathematical modelling

3.1. Introduction

Developing a descriptive mathematical model may assist in optimizing and controlling a specific environmental process more effectively because it permits the role and effect of essential parameters to be analysed. The worth of modelling is determined by the accuracy and reliability of available experimental data, the type of wastewater being treated, and the biochemical reactions involved (Orhon and Çokör, 1997; Yetilmezsoy *et al.*, 2015). Martinez *et al.* (2014) stipulate that the effectiveness of anaerobic reactors is affected by flow patterns of the reactor, loading rates, presence of toxic compounds, mass transfer in the biofilm and kinetic effects. Even though various models have been developed for anaerobic treatment, each kinetic model is only appropriate for specific cases and processes (Coskun *et al.*, 2012).

There is a wide variety of wastewater treatment modelling packages available, with most being based around the Activated Sludge Model (ASM) (Nutt *et al.*, 2004). A Task Group on mathematical modelling for the design and operation of activated sludge processes was established in 1983 by the International Association on Water Pollution Research and Control (IWAPRC), known as the International Water Association (IWA). The purpose of the Task Group was to construct a common platform that would be used to develop future models for nitrogen-removal activated sludge processes, which resulted in the creation of the Activated Sludge Model (ASM) No. 1 in 1987 (Melcer *et al.*, 2003). Ndeba-Nganongo *et al.* (2018) described the ASM1 as a systematic mathematical model founded on COD as a common unit used in wastewater characterization and depicts the main process describing biochemical and physico-chemical processes involved in the decomposition of complex organic matter into biogas and inert by-products. This model considers the oxidation of carbon, nitrification and denitrification and describes the fractionation of the mixed liquor based on 13 components: particulate and soluble substrates (Kuş and Kara, 2020). The ASM1 was more than a model for it also provided guidelines for wastewater characterization and development of computer codes and a set of default values that have proven to give realistic model results with minimal parameter changes. In 1995 the ASM2, which included the removal of nitrogen and biological phosphorus because of the growing popularity of biological phosphorus removal and the increasing understanding of the process' basic phenomena, was published. The ASM2 was expanded to ASM2d to include denitrifying phosphorus-accumulating organisms (PAOs) in 1999. The ASM3, which was based on the developments in the understanding of the AS processes, was developed to be a new modelling platform for the next generation of ASMs (Henze *et al.*, 2000a). As highlighted by Kuş and Kara (2020), the parameters used for characterizing the organic carbon content in wastewater are BOD and COD in all these models.

3.2. The basis of the IWA Activated Sludge Models

To begin the process of modelling activated sludge systems, the Task Group addressed systems that integrated CO₂, nitrification and denitrification phenomena. To provide realistic prediction, the first step was to identify the essential processes occurring within the system, where the term “process” is used to define a “micro” event (i.e. cell growth or maintenance) rather than a series of “macro” operations (i.e. an AS process) (Melcer *et al.*, 2003). The following step was to characterize the kinetics and stoichiometry of those processes and then integrate process rate expressions into material balance equations portraying the system’s physical configuration (Grady *et al.*, 1986). As mentioned by Henze *et al.* (2000) the Task Group decided to base the matrix format on the work done by Peterson (1965) because it communicated the most amount of information and to use the notation recommended by Grau *et al.* (1982).

To introduce the matrix format and notation, consider a situation where heterotrophic bacteria grow through a soluble substrate for carbon and energy in an aerobic atmosphere. The increase of biomass by cell growth and decrease by decay are the two fundamental processes occurring. Also occurring is the utilization of oxygen and removal of substrate; however, these are not considered fundamental, for they result from the growth and decay of biomass and join them through the system stoichiometry (Grady *et al.*, 1986). The model of such a situation must consider the concentrations of the three components, namely biomass, substrate and DO (Henze *et al.*, 2000). The matrix, including the outcome of these three components in the two fundamental processes, is displayed in Table 3.1. The biological processes occurring in the system, namely the aerobic growth of biomass and its loss by decay, are listed in the leftmost column of Table 3.1, while the components are listed by symbol across the top and by name and units across the bottom. The X symbol is given to the insoluble constituents, and the symbol S is given to soluble components; and to identify the different components, the subscripts B for biomass, S for substrate and O for oxygen are used. The index j is allocated to each process ranges from 1 to 2, while the index i is allocated to each component and ranges from 1 to 3. In the rightmost column of Table 3.1, the kinetic expression or rate equations for each process are recorded and are symbolised by ρ_j , and the rate expressions are based on the simple Monod-Herbert (1958) model. As Henze *et al.* (2000) mentioned, the Monod equation, ρ_1 , states that biomass growth is proportional to biomass concentration in a first-order manner and substrate concentration in a mixed order manner. While the Herbert equation, ρ_2 , states that biomass decay is first order regarding biomass concentration. In the lower right corner of Table 3.1, the kinetic parameters used in the rate expression are defined (Grady *et al.*, 1986; Henze *et al.*, 2000).

The elements in the matrix consist of the stoichiometric coefficients, v_{ij} , which set out the mass relationship between the components in the separate processes. For instance, biomass growth (+1) happens at the expense of soluble substrate ($-1/Y$), and oxygen is consumed in the metabolic process [$-(1-Y)/Y$]. The coefficients, v_{ij} , are significantly simplified by working in consistent units. In this case, all organic constituents have been presented as equal quantities of COD; similarly, oxygen is articulated as negative oxygen demand. For consumption, the sign convention used in the matrix is negative and positive for production. All stoichiometric coefficients are displayed in the lower-left corner of Table 3.1 (Grady *et al.*, 1986; Henze *et al.*, 2000).

Table 3.1: Process Kinetics and stoichiometry for heterotrophic bacterial growth in an aerobic environment (Grady *et al.*, 1986; Henze *et al.*, 2000).

Component →		Continuity			Process Rate, ρ_j [$ML^{-3}T^{-1}$]
		i	1	2	
j	Process ↓	X_B	S_S	S_O	
1	Growth	1	$-\frac{1}{Y}$	$-\frac{1-Y}{Y}$	$\frac{\hat{\mu}S_S}{K_S + S_S}X_B$
2	Decay	-1		-1	bX_B
Observed Conversion Rates $ML^{-3}T^{-1}$		$r_i = \sum_j r_{ij} = \sum_j v_{ij}\rho_j$			Kinetic Parameters: Maximum specific growth rate: $\hat{\mu}$ Half-velocity constant: K_S Specific decay rate: b
Stoichiometric Parameters: True growth yield: Y		Biomass [$M(COD)L^{-3}$]	Substrate [$M(COD)L^{-3}$]	Oxygen (negative COD) [$M(-COD)L^{-3}$]	

3.3. Use in material balances

In a system, the concentration of a component may be affected by numerous different processes. The fact that the matrix presentation permits rapid and easy recognition of the outcome of each component can be viewed as a benefit, which helps in the preparation of the material balance equations. This is evident when moving along the column representing a component; hence the arrow marked "Material Balance" is located at the left-hand side of

Table 3.1. The basic equation for a material balance within any defined system boundary is indicated by Equation 3.1 (Grady *et al.*, 1986; Henze *et al.*, 2000):

$$\text{Input} - \text{Output} + \text{Reaction} = \text{Accumulation} \quad (3.1)$$

The input and output terms are transport terms reliant on the physical characteristics of the system being modelled. The system reaction term, r_i , is attained by adding the products of the stoichiometric coefficient v_{ij} and the process rate expression ρ_j for the component i being considered in the material balance as indicated in Equation 3.2 (Grady *et al.*, 1986; Henze *et al.*, 2000):

$$r_i = \sum_j v_{ij} \rho_j \quad (3.2)$$

For example, the rate of reaction, r , for biomass, X_B , at a point in the system is communicated by Equation 3.3 (Grady *et al.*, 1986; Henze *et al.*, 2000):

$$r_{X_B} = \frac{\hat{\mu} S_S}{K_S + S_S} X_B - b X_B \quad (3.3)$$

Equation 3.4 is the material balance for soluble substrate, S_S (Grady *et al.*, 1986; Henze *et al.*, 2000):

$$r_{S_S} = -\frac{1}{Y} \frac{\hat{\mu} S_S}{K_S + S_S} X_B \quad (3.4)$$

Equation 3.5 is the material balance for DO, S_O (Grady *et al.*, 1986; Henze *et al.*, 2000):

$$r_{S_O} = -\left(\frac{1-Y}{Y}\right) \frac{\hat{\mu} S_S}{K_S + S_S} X_B - b X_B \quad (3.5)$$

To generate the material balance for each component in a given system boundary (e.g. a completely mixed reactor), the conversion rate would be joint with the suitable flow terms for the specific system. These terms have not been presented here, for the purpose of the illustration was to prove how the matrix is used to define the fundamental reactions irrespective of the system configuration. Nevertheless, it should be highlighted, that modelling a particular physical system needs the definition of the system boundary with the associated flow terms (Henze *et al.*, 2000).

3.4. Continuity check

Continuity, which may be verified by moving across the matrix, as long as the consistent units used to add the stoichiometric coefficients were equivalent to zero, is another benefit of the matrix. The decaying process can prove this; oxygen is negative COD; therefore, its coefficient must be multiplied by -1 prior to adding. All COD from the biomass due to decay should be balanced oxygen consumption. Likewise, the substrate COD lost from the solution because of growth minus the quantity transformed into new cells must be equivalent to the oxygen used for cell synthesis (Grady *et al.*, 1986; Henze *et al.*, 2000).

3.5. Simulation software packages

As Melcer *et al.* (2003) described, a computer program integrating a biological model with models of other unit operations is a simulator that links units of a specific system to a particular flow scheme. The requirements for setting up a wastewater treatment system simulation that incorporates an AS process are indicated in Figure 3.2, where physical configurations are details such as reactor volume and clarifier dimensions, operating conditions are wastage rate, recycle rates and DO concentrations, and influent loading patten refer to flow rate, COD and TKN. The intended use of the simulator determines the information necessary for the inputs to the process, its configuration and operating conditions. Several software packages assume steady-state for the simulations are run utilizing average day flows and loads, and this is referred to as steady-state modelling. This type of modelling is suitable for checking process unit sizes and performing basic fault findings of treatment processes (Nutt *et al.*, 2004). However, as outlined herein, wastewater treatment processes are dynamic due to the considerable differences in influent wastewater flow rate and composition, and this is revealed by the time-varying reaction parameters thus generating a non-linear and unsteady system (Gazsó *et al.*, 2017). Nutt *et al.* (2004) state that more advanced software packages can perform dynamic modelling using fluctuating conditions over hours, days, weeks or years. This type of modelling provides more information on the performance of the treatment process over

some time. Figure 3.1 illustrates the process input requirements for a well-constructed simulation of a wastewater treatment system.

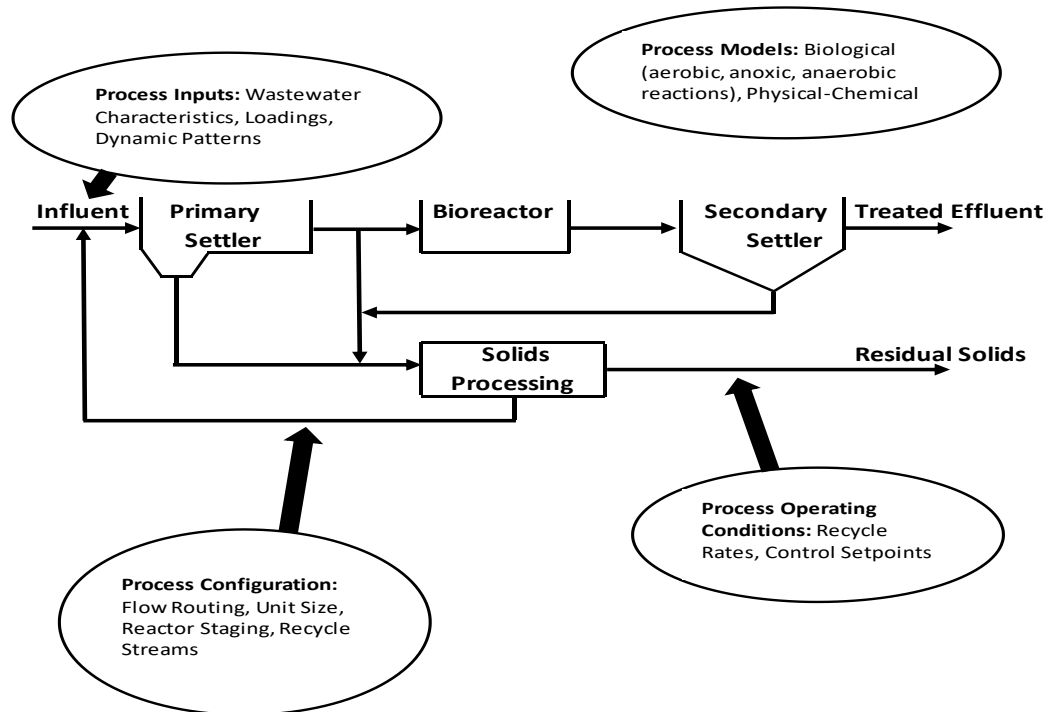


Figure 3.1: Requirements for wastewater treatment system simulation

In a study conducted by Coskun *et al.* (2012) a UASB reactor treating antibiotic fermentation broth wastewater was investigated with regards to COD removal and methane production, using the modified Stover-Kincannon, first-order, substrate mass balance, and Van der Meer and Heertjies kinetic models. With a hydraulic retention time of 13.3 days and an OLR ranging from 0.33 to 7.43kgCOD/m³.day, a 95.7% COD RE was achieved with a maximum yield of 3,700L/day methane gas production, therefore proving that the UASB was capable of effectively treating antibiotic fermentation broth. The Adaptive Neuro-Fuzzy Inference System (ANFIS) based model was used by Yetilmezsoy *et al.* (2015) to predict the effluent COD load from a full-scale EGSB. According to the descriptive statistical performance indicators used for validation, this model accurately predicted the effluent COD. Yang *et al.* (2015) developed a three-dimensional Computational Fluid Dynamics (3D CFD) model that integrated the hydrodynamics and biokinetics of an EGSB reactor treating manufactured municipal wastewater to determine the effect of influent distribution in reactor hydrodynamics, mass transfer and operation details at different points in the reactor. This study concluded that the 3D CFD model successfully predicted the even distribution of the influent. Response Surface

Methodology (RSM) is a mathematical and statistical technique used by Williams (2017) to predict the performance of an EGSB reactor coupled with an anoxic/aerobic hybrid side stream ultrafiltration membrane bioreactor used for treating PSW with regards to tCOD removal. RSM successfully determined the suitable model that would fit the experimental data and the best optimum conditions for a maximum of 93% tCOD removal, and these were found to be an OLR of 2gtCOD/L.day OLR and hydraulic retention time (HRT) of 4.82 days.

3.6. Sumo simulator software

SUMO is a full-featured wastewater treatment process simulator software developed by Dynamita (<http://www.dynamita.com/>) for environmental models, especially for municipal and industrial WWTP modelling. It can simulate biokinetics models dynamically or in steady-state, direct algebraic and mixed equilibrium-kinetic models subject to the simulation mode. Sumo contains internally researched and developed the whole plant and focused models including those for N and P removal (Gazsó et al., 2017), namely, ASM1, ASM2D, ASM2D_TUD, ASM3_BioP, ASM3), Barker_Dold, BUCTPHO plus (Ndebe-Nganongo, 2018). Thus far, no study has been reported with regards to using Sumo to predict the performance of a PSW treatment system consisting of an aerobic pre-treatment tank, EGSB reactor and a submerged membrane tank in South Africa, more specifically Cape Town, with regards to the removal of total suspended solids (TSS), and fats, oil and grease (FOG) in addition to the removal of COD.

According to Gazsó *et al.* (2017), dynamic modelling is based on a differential equation system using the Peterson matrix, which contains processes that influence the change of the state variables. Sumo models are written in an Excel-based open process source code language termed SumoSlang (Sumo Simulation Language) and are responsible for describing of the operational units. Further Gazsó *et al.* (2017) indicated that the SumoSlang based files contain the physical and technological parameters together with mass balance calculations for each unit. The development of the simulation model involved the following:

- 1) building and plant configuration,
- 2) setting up the model parameters and selecting the type of model to be used (Mini_Sumo, Sumo1, Sumo2, Sumo2S or Sumo2C),
- 3) adding calculations to be simulated (i.e. sludge retention time (SRT) or removal efficiencies),
- 4) using measured and parameters of the integrated multi-stage treatment lab-scale plant as well as operating conditions to set up the model,

- 5) selecting either a constant or dynamic input type,
- 6) specifying the variables and the format in which they should be presented and saved during simulation, and
- 7) running the simulation.

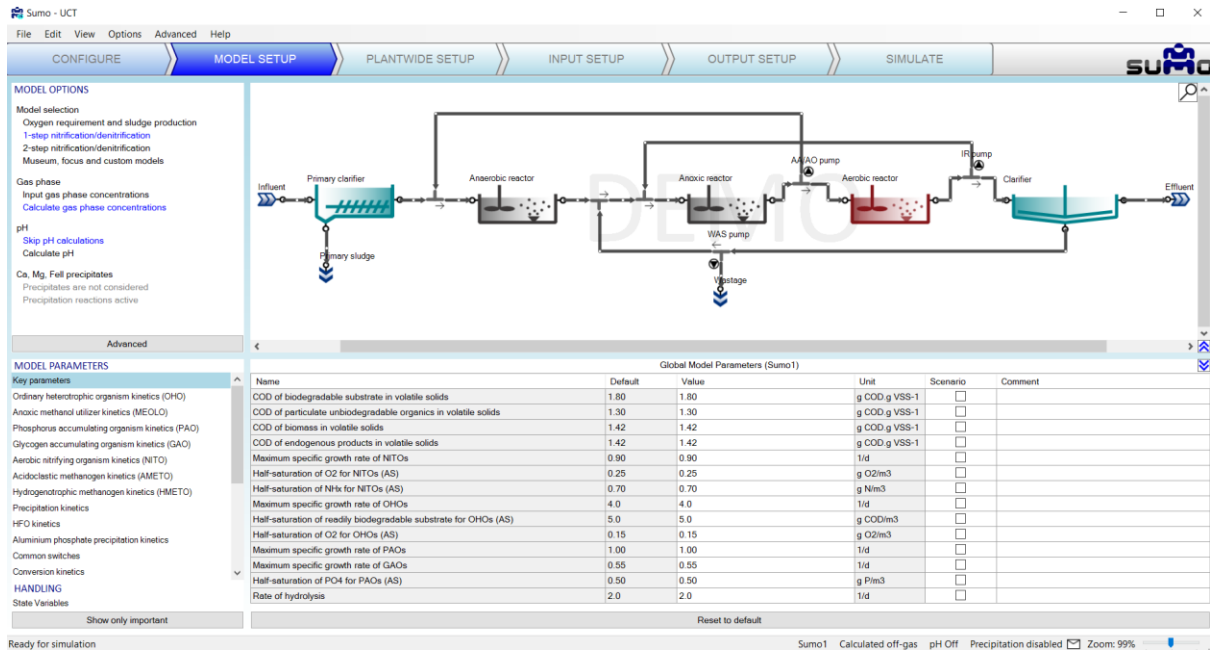


Figure 3.2: University of Cape Town configuration with one-step nitrification/denitrification model

CHAPTER FOUR

Trinal simulator stages for modelling of poultry slaughterhouse wastewater nutrient removal

Parts of this chapter were presented in conferences in 2019 and 2021 as:

Maggie Ntombifuthi Bingo, Seteno Karabo Obed Ntwampe, Lionel Neddy Aymar Ndeba-Nganongo and Moses Basitere, 2019. Trinal Simulator Stages for Modelling of Poultry Slaughterhouse Wastewater Nutrient Removal. 2019 Innovation Conference on Sustainable Wastewater Treatment and Resource Recovery Nov. 24-28, 2019, Shanghai, China. www.nrr2019.com (International conference)

M. N. Bingo, M. Basitere, S. K. O. Ntwampe, and D. N. Dlamini, 2021. Trinal Simulator Stages for Modelling a Pilot Scale Poultry Slaughterhouse Wastewater Treatment Plant Using Sumo. 12th Eastern European Young Water Professionals Conference. 31 March -2 April 2021, Riga, Latvia, <http://iwa-ywp.eu/>. (International conference).

4.1. Introduction

Abattoir waste has been identified as the most challenging food waste type in the Western Cape (WC) Province, South Africa (SA), due to its hazardous nature and potential impact on the environment and human health. For instance, in 2016, poultry slaughterhouse wastewater (PSW) contributed 43% to the total abattoir waste produced in the WC (Western Cape Government, 2016). The poultry abattoir industry is water-intensive, with 90% of the potable water consumed being discharged as wastewater because it is used for cleaning and washing carcasses and meat products, and the sanitation and disinfection of equipment as well as all the processing areas of the slaughterhouse (Molapo, 2009; Department of Environmental Affairs and Development Planning, 2015). High levels of organic compounds typically characterized PSW, thus have a high content of biological oxygen demand (BOD) and chemical oxygen demand (COD), which are in the form of nutrients, suspended solids, and fats, oil and grease (FOG) (Basitere *et al.*, 2017). The presence of organic matter can also result from the slaughtered birds' blood, urine and faeces (Al Smadi *et al.*, 2019). Nitrogen (N) and phosphorus (P) are the primary nutrients present in the PSW (Molapo, 2009). Total nitrogen (TN) is available in the form of ammonia (NH_4^+ , $\text{NH}_3 - \text{N}$), nitrite (NO_2^-) and nitrates (NO_3^-), which are the most stable forms of TN in the PSW and can thus be reduced as a result of treating the PSW. Similarly, orthophosphate (PO_4^{3-}) is a common form of P, and its manifestation in PSW is because of the use of sanitizing detergents for cleaning purposes (Bustillo-Lecompte & Mehrvar, 2017; Yaakob *et al.*, 2018).

When PSW is discharged into waterways and municipal systems, it can result in harmful algal growth, dissolved oxygen depletion, which would result in the death of fish and other aquatic life, including reducing the efficacy of municipal wastewater works (MWWW), further resulting in water-borne diseases and inadequately treated wastewater (Environmental Integrity Project, 2018; Yaakob *et al.*, 2018). Legislative regulations were implemented, including the "polluter pays," to minimise such behaviour. This was achieved by enacting a national pricing strategy for effluent discharged into the environment or MWWW. This national pricing strategy was established by the Department of Water and Sanitation (DWS) in SA under the National Water Act (Act 36 of 1998), which stipulates the purifying standards of wastewater prior to disposal, and incorporated the Waste Discharge Charge System (WDCS) (Pocock & Joubert, 2017). These industrial discharge standards vary from province to province because they are stipulated by the individual local municipality (Molapo, 2009); therefore, the poultry slaughterhouse located in the Western Cape (WC) are required to comply with the wastewater and industrial effluent discharge standards set out by the City of Cape Town (CCT) municipality (Basitere, 2017). The WDCS was based on the aforementioned "polluter pays"

principle and was meant to create a financial incentive for effluent dischargers who reduce their wastewater and use water resources efficiently. On the other hand, slaughterhouses are fined when their wastewater is not at the required effluent discharge standards as specified by the municipal by-laws, which are based on the Water Services Act (Act 108 of 1997) that indicates the water effluent guidelines and tariffs (Department of Water and Sanitation, 2007; Pocock & Joubert, 2017). The WDCS comprises of two distinct charges; 1) the Waste Mitigation Charge (WMC) that recuperates the expenses incurred for lessening the impacts of discharging waste into water resources, and 2) the Waste Discharge Levy (WDL), which compensates dischargers who reduce their waste (Department of Water Affairs and Forestry, 2007). Therefore, in order to adhere to the effluent discharge standards, PSW has to undergo extensive treatment.

Currently, extensive research has been done experimentally on lab-scale anaerobic bioreactors; however, to upscale these reactors for industrial application, particularly for SMMS's, simulations and process modelling must be conducted to assess the feasibility of the proposed system adequately. Simulation is an accepted tool in engineering practice to assist with plant design, development, operation, and control (Gazsó *et al.*, 2017). The purpose of this study was to simulate a lab-scale PSW biological treatment process for the benefit of SMMS's in the WC, SA. The objectives were to determine different COD fractions essential to generate accurate model input wastewater values, design a model treating PSW using SUMO, and assess the performance of the model designed mainly with regards to the removal efficiency of COD, BOD, NH₃, PO₄³⁻, total suspended solids (TSS) and volatile suspended solids (VSS).

4.2. Treatment methods of poultry slaughterhouse wastewater

The treatment methods applied to PSW are considered dynamic because of the fluctuations of influent flow rate and composition; thus, they combine of physico-chemical, biological, and biochemical processes (Gazsó *et al.*, 2017). Pre-treatment processes are characteristically the first stage for treatment where coarse material (feathers, meat particles, bones, protein and FOG) is removed. This is followed by primary treatment, a physico-chemical process that separates solids from liquids by removing TSS, FOG, COD and BOD. Thereafter, a secondary biological treatment process is employed to reduce the organic compounds and deactivate pathogens through the use of microorganisms (Basitere *et al.*, 2017; Mbulawa, 2017). Finally, a tertiary treatment process is applied to remove the residual suspended or dissolved material not removed by the secondary treatment process (Kang *et al.*, 2007).

Anaerobic treatment is the most suitable biological treatment process because it is advantageous in treating slaughterhouse wastewater (SWW) successfully with minimal complicated equipment requirements, high COD removal and the production of biogas which has the possibility of balancing the energy costs of the slaughterhouses (Al Smadi *et al.*, 2019; Bustillo-Lecompte & Mehrvar, 2017). This treatment process utilises anaerobic bacteria to degrade organic compounds into carbon dioxide (CO₂) and methane (CH₄) in the absence of oxygen (Basitere *et al.*, 2017).

Anaerobic digestion (AD) utilizes microorganisms to break down organic matter in four different phases: hydrolysis, acidogenesis, acetogenesis and methanogenesis, with each phase being dominated by a precise bacterial population. The first phase is hydrolysis in which complex polymers are broken down into monomers such as sugar, amino and fatty acids (Manyi-Loh *et al.*, 2013). Yuan and Zhu (2016) mention that this phase is considered to be the rate-limiting step for the degradation of organic substrates because of the formation of volatile fatty acids (VFA) such as acetate (CH₃COO⁻). This phase is followed by acidogenesis, where the conversion of the hydrolysis products to form CO₂, H₂, NH₃, H₂S, alcohols and VFAs by acidogenic bacteria occurs (Basitere, 2017; Williams, 2017). Acetogens consume alcohols and organic acids to generate more CH₃COO⁻, CO₂ and H₂ in the third phase, known as acetogenesis. In the final phase, methane production takes place by converting acetic acid (CH₃COOH) molecules using acetrophic methanogens or converting CO₂ using hydrogenotrophic methanogens (Manyi-Loh *et al.*, 2013).

4.3. Mathematical modelling

In order to have a better understanding of how wastewater treatment plants (WWTPs) perform under several operating conditions, mathematical modelling is applied (Ndeba-Nganongo *et al.*, 2018). Mathematical modelling of biological treatment processes permits a comprehensive design of the operating parameters, attainable effluent quality and prediction of system reaction for extreme simulations (Gazsó *et al.*, 2017). The activated sludge process is among the prevalent biological wastewater treatment technologies applied, where the utilization of bacterial biomass removes pollutants such as biological N, biological P and organic carbon substance. The advanced knowledge of the different biological processes occurring in activated sludge plants has been interpreted into dynamic models developed to describe the degradation process (Petersen *et al.*, 2003). SUMO simulator is a wastewater process multipurpose simulator software developed by Dynamita Incorporate for environmental models, specifically for municipal and industrial wastewater treatment plant modelling (Ndeba-

Nganongo *et al.*, 2018). Kolovos *et al.* (2016) recommend the application of SUMO because of its user-friendly interface and Excel-based open-source process code language called SumoSlang (Sumo Simulation language), making the calculations behind the simulation model used easily understandable. Sumo contains internally researched and developed the whole plant and focused models such as activated sludge models (ASM1, ASM2d and ASM3) for N and P removal, and is capable of simulating bio-kinetic, mixed equilibrium-kinetic and direct algebraic models in steady-state, subject to the outputs of the intended process (Gazsó *et al.*, 2017).

4.4. Materials and methods

4.4.1. Chemical oxygen demand fractionation

The selection of the treatment process depends on the wastewater's characteristics, compliance with regulations, and the available technology (Bustillo-Lecompte & Mehrvar, 2015). Therefore prior to designing the treatment model, the characteristics of the PSW being treated needed to be determined. This included COD fractionation which involved the identification of inert and biodegradable COD and readily biodegradable and slowly biodegradable fractions (Orhon & Çokör, 1997). By determining the COD fractions, the number of inert pollutants, which reduce the efficiency of the biological treatment, can also be assessed. As a result, the use of total COD content in raw wastewater divided into soluble and particulate fractions is preferred because it permits for subsequent calculations of the content of individual forms of N found in wastewater. The total COD is then calculated using Equation 4.1.

$$\text{Total COD} = S_s + S_I + X_I + S_{col} + X_s \quad (4.1)$$

where S_s is soluble readily biodegradable COD, S_I is inert soluble COD, X_I is inert particulate COD, S_{col} is slowly biodegradable colloidal COD, and X_s is particulate slowly biodegradable COD (Myszograj *et al.*, 2017).

Raw PSW samples were collected during slaughtering and cleaning operations in 5L and 25L polypropylene containers from wastewater disposal facilities of a poultry slaughterhouse located in the WC Province, SA. The total COD, which includes filtered COD and filtered

flocculated COD (ffCOD), was analysed using Merck solutions: A (1.14679.0495) and B (1.14680.0495) for a high range and A (1.14538.0065) and B (1.14681.0495) for a low range, and the readings were recorded on a Merck Spectroquant® UV/VIS Spectrophotometer Pharo 300. In order to determine the filtered COD, a PSW sample was passed through a 1.0µm glass fibre filter, and the residue after filtration is depicted in Figure 4.1.



Figure 4.1: Image showing the residue of PSW on the 1.0µm glass fibre filter after filtration.

The procedure suggested by Mamais *et al.* (1993), which comprises the flocculation of the PSW followed by a filtration process, was implemented with minor changes to remove colloidal COD and determine the ffCOD. In the flocculation step, 1mL of zinc sulphate ($ZnSO_4$) was added to 100mL of PSW, vigorously stirred for one minute. Thereafter, a 6M of sodium hydroxide (NaOH) was added to adjust the pH of the solution to 10.5, and the solution was allowed to settle for a few minutes. The resulting supernatant was filtered using a 0.22µm membrane filter. Figure 4.2 illustrates the test kits tubes used to measure the COD fractions, whereas Figure 4.3 depicts the passage of PSW influent into the different COD components through the 1.0µm glass fibre filter, flocculation, and 0.22µm membrane filter steps.



Figure 4.2: COD fractions (total, filtered and flocculated filtered COD) in analysis test tubes.

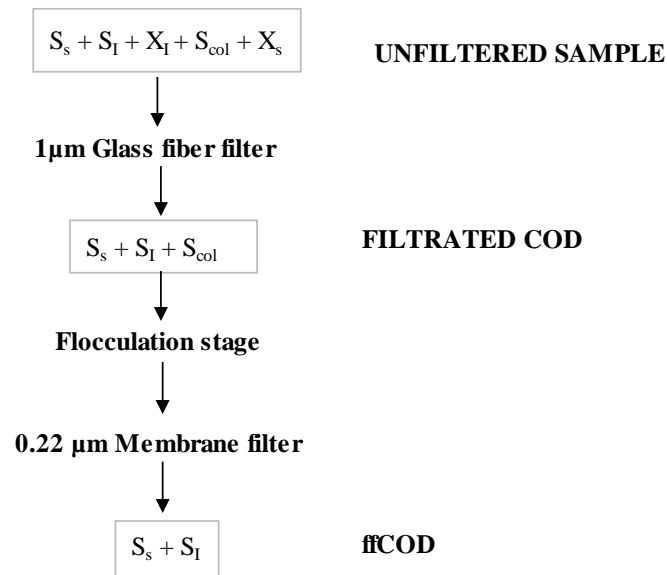


Figure 4.3: Influent PSW passage into different COD components.

4.4.2. SUMO simulator software

Over the past decade, single-stage anaerobic digesters, in which all four phases of AD take place have been widely utilized in wastewater treatment. The digester contents must be thoroughly mixed for a high digestion rate while maintaining a constant mesophilic temperature (Cheremisinoff, 1997). Even though the single-stage system design is simple and economical, biogas production is low, and the feedstock takes longer to digest, causing high hydraulic retention time (HRT) and low throughput rates for treating the PSW. Biogas production is further inhibited by the accumulation of VFA caused by overloading and excessive

macronutrients, leading to souring (Mao *et al.*, 2015). The two-stage digestion splits the anaerobic digester into two operational stages (Blumensaat & Keller, 2005): acid-phase digestion and temperature-phase anaerobic digestion (Zahller *et al.*, 2007). The objective is to encourage the growth of acid-forming bacteria in the first-stage, producing VFA used by methanogenic archaea in the second stage (Pohland & Ghosh, 1971). Even though the two-stage digester has been proven to be advantageous when compared to the single-stage digester, it has a high capital cost and operational instability under certain environmental conditions (Blumensaat & Keller, 2005).

In this study, a three-stage AD model was also evaluated, with the first AD unit focusing on COD reduction, whereas the second AD unit being used for biogas generation and the third AD unit being used for NH₃ removal. The three simulation models designed in SUMO are depicted in Figure 4.4., with Figure 4.4A representing the single-stage AD system, Figure 4.4B representing the two-stage AD system, and finally Figure 4.4C representing the three-stage AD. The models were set to operate at steady-state for 150 days at a temperature of 35°C, an organic loading rate (OLR) of 143.6mgCOD/L.day, and a flow rate of 3 590m³/day. However, the single-stage digester had an SRT of 25 days, while the first digester of the two-stage digestion had an SRT of 15 days and 25 days in the second digester. The remaining influent wastewater characterization parameters used in the models were obtained from literature, and a few of them are listed in Table 4.1. In order to assess the potential biodegradability of the organic matter present in PSW, the COD/BOD ratio was determined and estimated to be 2.15. According to Bustillo-Lecompte and Mehrvar (2015), a ratio below 0.30 may be considered to be non-responsive to treatment. The COD/VSS and BOD/TSS ratios were estimated to be 1.49 and 1.31, respectively.

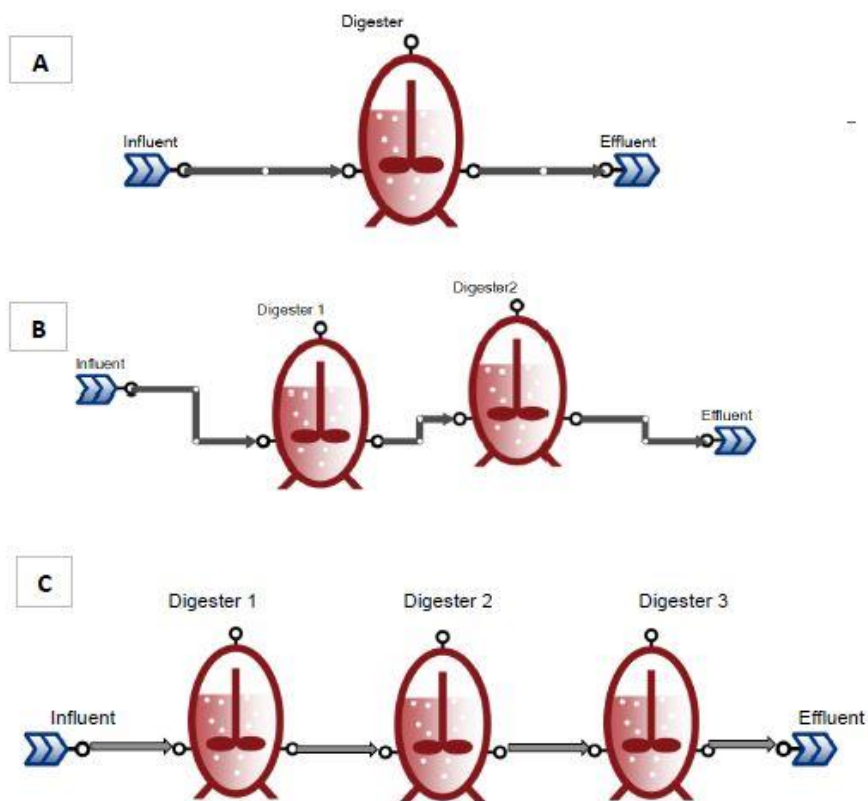


Figure 4.4: The three simulation models designed in SUMO using the ASM2d model.

Table 4.1. Characteristics of wastewater from a poultry slaughterhouse in the Western Cape (Basitere *et al.*, 2017).

Parameter	Unit	Min	Max	Average
pH	-	7	8.0	10.5
Conductivity	$\mu\text{S/cm}$	899	2 450	2 124
Salinity	ppm	529	1 413	1 235.5
Turbidity	NTU	237	997	735.5
TSS	mg/L	313	8 200	4 413
TDS	ppm	372	1 740	1 242
VSS	mg/L	232	8 900	4 682
NH_4^+ -N	mg/L	135	447	358.5
NO_3^+ -N	mg/L	30	235	147.5
PO_4^{3-} -P	mg/L	29	54	56
VFA	mg/L	96	235	213.5
Alkalinity	mg/L	360	926	823
BOD	mg/L	1 100	5 000	3 600
Total COD	mg/L	2 517	12 490	8 762
FOG	mg/L	156	1 710	1 011

4.5. Results and discussion

The main removal efficiencies for the parameters used to evaluate the models' performance were total COD, TSS, VSS, BOD, NH₃ and PO₄³⁻, and Equation 1 was the expression used to determine the removal efficiency of each parameter.

$$\text{Removal efficiency} = (\text{Influent parameter} - \text{effluent parameter}) / \text{Influent parameter} \times 100 \quad (4.2)$$

The single-stage digester was operated as a primary treatment system reduce organic matter and suspended solids; thus, the digester also functioned as a biofilter. The maximum removal efficiencies obtained by the single-stage AD model are indicated in Table 4.2. The maximum removal efficiencies of total COD, TSS, VSS and BOD were obtained by the single-stage AD model that was operating at steady-state for 150 days at a temperature of 35°C, an OLR of 143.6mgCOD/L.day, and a flow rate of 3 590m³/day where 64%, 77%, 84% and 94%, respectively. However, the removal efficiency for NH₃ was -20% and -12.5% for PO₄³⁻, indicating that there was an increase in the amount of nutrients present in the effluent. According to Figure 4.5, the effluent of the single-stage AD model does meet the effluent discharge standards as Stipulated by the CCT regarding total COD, TSS, VSS, BOD, NH₃ and PO₄³⁻; thus, the wastewater can be discharged into the MWWW for further treatment.

Table 4.2. Characteristics of the single-stage anaerobic digester effluent

Parameter	Unit	Influent	Effluent	CCT By-law limit	% Removal
Total COD	mgCOD/L	3 590	1 273	5 000	64
TSS	mgTSS/L	4 413	999	1 000	77
VSS	mgVSS/L	4 682	744	-	84
BOD	mgO ₂ /L	3 600	212	-	94
NH ₃	mgN/L	147.5	178	-	-20*
PO ₄ ³⁻	mgP/L	56	63	25	-12.5*

*Indicates the accumulation of parameters within the digester system designed.

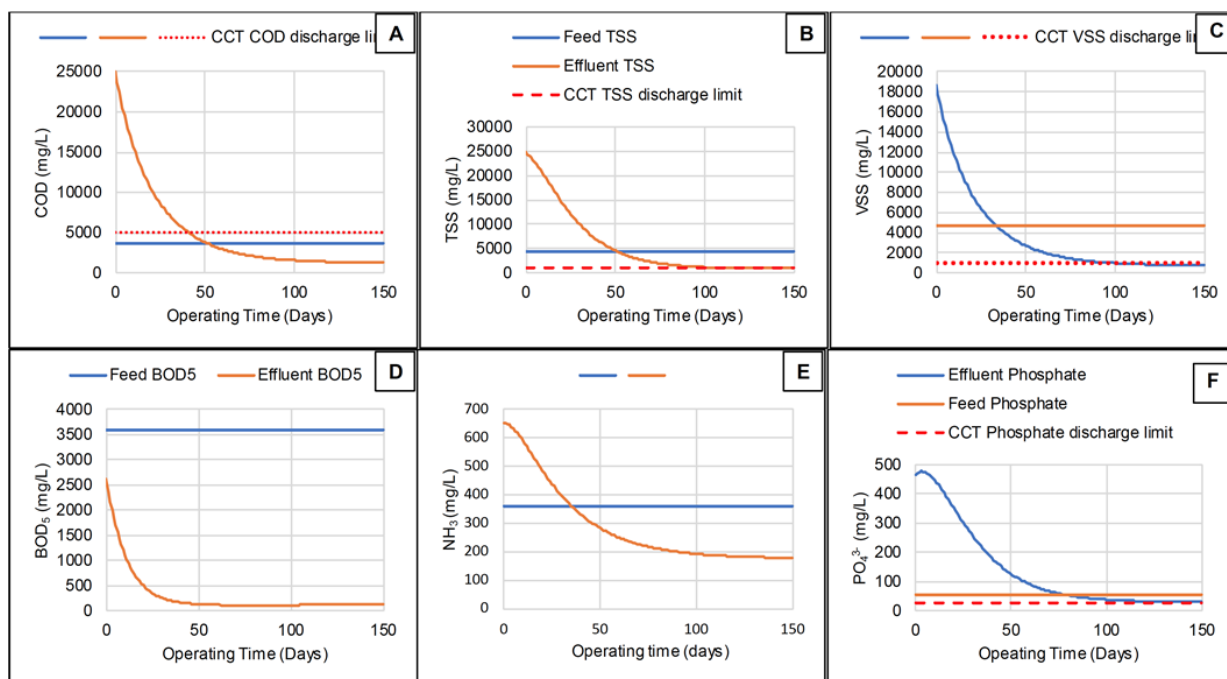


Figure 4.5: Single-stage anaerobic digestion performance – feed and effluent characterisation in relation to the CCT wastewater limits for COD, VSS, TSS, BOD₅, NH₃ and PO₄³⁻.

The two-stage digestion system designed was configured to use acid-phase digestion. Compared to the single-stage digester, the two-stage digestion process performed slightly better at an OLR of 143.6mgCOD/L.day, and the results are displayed in Table 4.3. The maximum removal efficiencies obtained by the two-stage was 69% total COD, 79% TSS, 86% VSS and 97% BOD at the same operating conditions as the single-stage AD. Similar to the single-stage AD, there was an increase in the amount of nutrients present in the effluent. The amount of NH₃ leaving the second AD increased by 23%, and that of PO₄³⁻ increased by 25%, double the amount observed in the single-stage AD. Figure 4.6 also confirms that the quality of the two-stage AD effluent wastewater is well within the standards of the CCT concerning the measured parameters.

Table 4.3: Characteristics of the two-stage anaerobic digester effluent

Parameter	Unit	Influent	Effluent	CCT By-law limit	% Removal
Total COD	mgCOD/L	3 590	1 106	5 000	69
TSS	mgTSS/L	4 413	910	1 000	79
VSS	mgVSS/L	4 682	671	-	86
BOD	mgO ₂ /L	3 600	122	-	97
NH ₃	mgN/L	147.5	182	-	-23*
PO ₄ ³⁻	mgP/L	56	70	25	-25*

*Indicates the accumulation of parameters within the digester system designed.

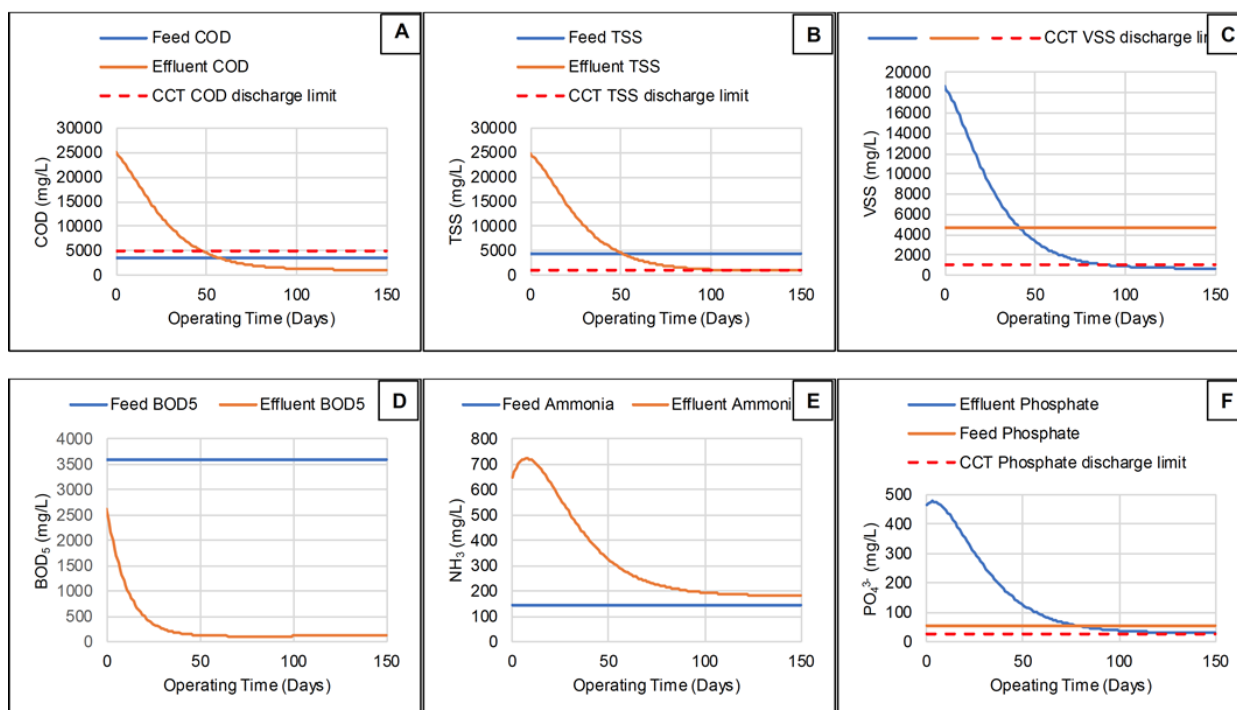


Figure 4.6: Two-stage anaerobic digestion performance – feed and effluent characterisation in relation to the CCT wastewater limits for COD, VSS, TSS, BOD₅, NH₃ and PO₄³⁻.

The maximum removal efficiencies achieved by the three-stage digestion model were 22%, 76%, 83%, 67% and 52% for total COD, TSS, VSS, BOD and PO₄³⁻, respectively. Compared to the single-stage and two-stage AD models, the three-stage AD models were less effective in removing total COD, TSS, VSS and BOD (Table 4.4). The NH₃ content increased further to 38%; therefore, the three-stage effluent failed to meet the CCT industrial effluent discharge standards, and if discharged into MWWWs, it could cause potential damage to the equipment.

Table 4.4: Characteristics of the three-stage anaerobic digester effluent

Parameter	Unit	Influent	Effluent	CCT By-law limit	% Removal
Total COD	mgCOD/L	3 590	2 801	5 000	22
TSS	mgTSS/L	4 413	1 040	1 000	76
VSS	mgVSS/L	4 682	793	-	83
BOD	mgO ₂ /L	3 600	1 192	-	67
NH ₃	mgN/L	147.5	204	-	-38*
PO ₄ ³⁻	mgP/L	56	27	25	52

*Indicates the accumulation of parameters within the digester system designed.

4.6. Summary

The design of single-stage and two-stage PSW treatment systems was successfully employed with the resultant effluents complying with the CCT wastewater and industrial effluent by-law standards. However, the nutrient removal proved to be a challenge, for there was a minute increase in the NH_3 and PO_4^{3-} present in the effluent of both models. In contrast, the three-stage AD model was ineffective in treating the PSW, causing the effluent to not comply with the CCT discharge standards. The results obtained indicate that anaerobic treatment on its own is not sufficient for the complete treatment of PSW. Aerobic treatment processes are commonly applied as post-treatment systems of the AD effluent, particularly with NH_3 removal. They are advantageous for minimising odour production, having a rapid biological growth rate and quickly adjusting to temperature and loading rate alterations (Bustillo-Lecompte & Mehrvar, 2017). With aerobic digestion, microorganisms decompose organic substrates in the presence of oxygen by oxidising carbohydrates into CO_2 and water and converting nitrogenous waste into NO_3^- and sulfates (SO_4^{2-}) (Pocock & Joubert, 2017). As mentioned by Molapo (2009), aerobic effluent can be discharged into water bodies, and according to Roš and Zupančič (2002), this is possible because aerobic effluent is considered to be biologically stable and suitable for disposal. This insinuates that aerobic treatment must be applied as a post-treatment for anaerobic bioreactors if the intention was to discharge the effluent wastewater into local freshwater bodies. In a study conducted by Zupančič and Roš (2008), a two-stage anaerobic-aerobic digestion process treating activated sludge from a wastewater treatment plant in the presence of pure oxygen was evaluated regarding the removal of NH_3 . It was found that after an HRT of 8 days of the aerobic stage, the NH_3 removal efficiency was 85%. Thus, in future mathematical modelling research, the following should be explored:

- Since BOD/COD values are in the range 0.3-0.8, the BOD/COD ratio of 0.11 is low. This means that the BOD values in Table 4.2 might have been overstated.
- The performance of a two-stage anaerobic-aerobic digestion system is evaluated.
- Using data from an existing working digester to assess whether the empirical kinetic and stoichiometric values represent the digester's performance as a means of verifying the model developed.
- Performing a dynamic simulation in order to assess how the model responds to varying SRTs and HRTs

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CHAPTER FIVE

Performance evaluation of an integrated multi-stage poultry slaughterhouse wastewater treatment system

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Waste Management and Disposal (Q2)

5.1. Introduction

Abattoir waste is considered to be the most challenging food waste to process, with poultry slaughterhouse wastewater (PSW) being the largest contributor to the total abattoir waste generated in the Western Cape (WC), South Africa (SA) (Western Cape Government, 2016). The wastewater generated from a poultry slaughterhouse is highly contaminated and thus poses a threat to human health and the environment (Meiramkulova *et al.*, 2020a). PSW is typically categorised by high levels of organic matter (chemical oxygen demand (COD), and biochemical oxygen demand (BOD)) due to the presence of protein and fats, oil including grease (FOG), fibre, pathogens, veterinary pharmaceuticals, total nitrogen (TN), total phosphorus (TP), and total suspended solids (TSS) (Meiramkulova *et al.*, 2020b). Rinqest *et al.* (2019) mentioned, poultry slaughterhouses in SA are permitted to discharge partially treated wastewater directly into municipal sewer systems with limited monitoring, provided they adhere to the municipal by-laws set out by each municipality prescribed by the SA Water Act of 1997. Therefore, PSW must undergo treatment before discharge into the municipal sewer system to avoid municipal levies/charges for discharging wastewater that does not meet the prescribed by-laws (Njoya *et al.*, 2021) or reuse.

The treatment methods applied to PSW must be dynamic because of the fluctuations in influent flow rate and composition; thus, they can combine physico-chemical, biological and biochemical processes (Gazsó *et al.*, 2017). One of the physico-chemical units is a pre-treatment process that is characteristically the first stage for treating PSW, whereby coarse material (feathers, meat debris, bones, protein and FOG) is removed. Pre-treatment options usually practised in poultry slaughterhouses are screening, settling, catch basins and flotation systems (Bingo *et al.*, 2019). Physico-chemical processes do reduce some COD and BOD. Thereafter, a secondary biological treatment process is employed to reduce the organic matter and deactivate pathogens through the use of microorganisms (Basitere *et al.*, 2017; Mbulawa, 2017), some of which have antimicrobial activity. Biological treatment processes are classified under two categories, specifically anaerobic and aerobic treatment systems. The ideal biological treatment for PSW is anaerobic treatment because of its advantage in successfully treating slaughterhouse wastewater with less complex equipment requirements, high COD removal, and biogas production. This has the prospect of offsetting the slaughterhouse treatment system's energy and running costs (Njoya *et al.*, 2021; Bustillo-Lecompte and Mehrvar, 2017; Al Smadi *et al.*, 2019). Even though anaerobic treatment is ideal, anaerobically treated water contains solubilized residual organic matter and nutrients, and these can be successfully treated using aerobic processes after the anaerobic treatment. As a result, aerobic treatment, which operates at rates higher than anaerobic treatment methods, is

applied as a post-treatment for anaerobic effluent (Bustillo-Lecompte and Mehrvar, 2017). Finally, a tertiary treatment process is applied to remove the residual suspended or dissolved matter that the secondary treatment process has not removed and microorganisms therein. These tertiary treatment processes result in the treated water having potable water quality characteristics (Williams, 2017). Overall, the treatment performance of all these processes depends on several factors, such as influent characteristics of the wastewater (substrates composition, macro and micronutrients, toxic compounds), operational conditions (OLR, pH, HRT and sludge retention time (SRT)), temperature variations, biomass concentration and concentration of chemicals (Yetilmezsoy *et al.*, 2015).

Several studies have been conducted on the potential of treating PSW using integrated treatment processes consisting of multi-stage treatment units (Meiramkulova *et al.*, 2020c). The performance of a bench-scale static granular bed reactor (SGBR) coupled with an ultrafiltration (UF) membrane system was evaluated by Basitere *et al.* (2017). The combined effect of the treatment system investigated achieved on average 98%, 99.8% and 92.4% COD, TSS and FOG removal, respectively. The treatment performance of an EGSB, followed by a single-stage nitrification-denitrification (SSND) reactor and a third treatment stage consisting of a UF and microfiltration (MF) membrane bioreactor (MBR) treating PSW, was investigated by Williams (2017). The overall REs of the treatment system were 92% for TSS and 99% for COD. Similarly, Rinquest *et al.* (2019) investigated the performance of an SGBR coupled with an SSND and UF membrane module (ufMM) treating PSW. The combined effect of the treatment processes obtained average REs of 91% COD and 97% TSS. In a study conducted by Meiramkulova *et al.* (2020a), the performance of an electrochemical pre-treatment process connected to an integrated membrane filtration (IMF) system, which consisted of a single module of UF and another module of reverse osmosis (RO), treating PSW was investigated. The overall REs of TSS and COD for the integrated treatment process were approximately 100%, proving the importance of having both a pre- and post-treatment system incorporated.

The use of anaerobic bioreactors has proven to be successful in the treatment of PSW; however, some operational challenges, such as sludge washout due to a reduction in the activity and sloughing of granules, a reduction in the ability to remove particulate matter, and clogging or membrane fouling due to the presence of high FOG, may occur (Williams, 2017; Baker *et al.*, 2020). Thus, this study seeks to mitigate the challenges of clogging and fouling in the bioreactors used by adding a pre-treatment unit using a FOG hydrolysis agent called Eco-flush™ to an integrated multi-stage PSW treatment system consisting of an aerobic pre-

treatment stage, anaerobic EGSB and aerobic submerged UF membrane system. No study has reported on the performance efficacy of the combined use of such a system treating PSW.

The main purpose of this study was to investigate the performance of a lab-scale three-stage PSW treatment system with regards to the REs of COD, FOG and TSS. The results obtained were compared to the CCT discharge by-laws and the Department of Water and Sanitation (DWS) standards of effluent discharge to determine whether the treated PSW would be suitable for discharge into the CCT WWTWs, or for on-site reuse, whereby the water could be recycled and reused in other poultry slaughterhouse operations.

5.2. Materials and methods

5.2.1. Experimental set-up and equipment

The lab-scale multi-stage treatment plant treating PSW that was investigated in this study was conceptualised by researchers in the Bioresource Engineering Research Group (*BioERG*) at the Cape Peninsula University of Technology (CPUT) and manufactured by a South African company Malutsa (Pty) Ltd., located in Wellington, and is depicted in Figure 4.1. The plant consisted of an aerobic pre-treatment tank in which Eco-flush™ was added to hydrolyse FOG and reduce odour, sieves to remove suspended solids and particulate matter, an anaerobic feeding tank, an EGSB to remove organic matter, an EGSB product holding tank, an anoxic tank followed by a submerged membrane tank, and finally the membrane unit product tank. The temperature for the EGSB was maintained using a water bath, while Tedlar bags were used to collect the biogas from the unit.

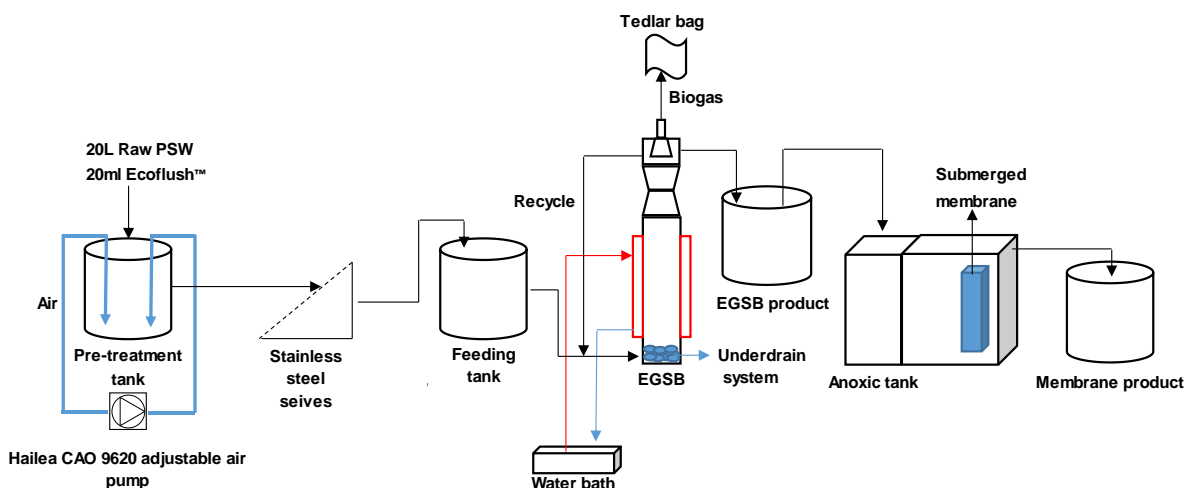


Figure 5.1: Process flow diagram of the experimental set-up.

5.2.2. Poultry slaughterhouse wastewater

The raw wastewater was collected in 25L polypropylene containers from a PSW outlet stream to a collection discharge point of a poultry slaughterhouse located in the Western Cape Province, (SA) – Figure S1. The wastewater stream was a combination of water used for cleaning and washing carcasses and meat products, and the sanitation of equipment and processing areas of the slaughterhouse. The collected raw PSW was stored in a refrigerator at less than 4°C until it was fed into the integrated lab-scale biological treatment system.

5.2.3. Pre-treatment process

The pre-treatment tank was a 25L container in which 20L of raw PSW was added together with 20mL of Ecoflush™, which is a hydrolysis remediation product supplied by Mavu Bio-Technologies (Pty) Ltd., a South African company located in Cape Town, SA – see Figure S2. Ecoflush™ consists of a cluster of bacteria that occur naturally and are extracted from soil, bottled in an inactive state, and activated when they come into contact with wastewater rich in nutrients. The constituents in Ecoflush™ include glucids and essential amino acids that encourage the natural proliferation of certain bacteria to generate enzymes capable of breaking hydrocarbon chains in FOG and oxidising ammonia (NH₃) into nitrite (NO₂⁻) and nitrate (NO₃⁻), including the elimination of pathogenic bacteria and those generating odour. It further reduces the components contributing to COD and BOD in the wastewater (Ergofito, 2019). The composition of Ecoflush™ is given in Table 5.1.

Table 5.1: Composition of the Ecoflush™ Bioremediation Agent (Ergofito, 2019)

Component	%
Enzyme and microbial blend	40
Organic nitrogen (N)	3
Potassium (K ₂ O)	4
Organic Carbon (C)	10
Organic matter	31
Water	12

The raw PSW with the Eco-flush™ mixture was aerated for 24h using air spargers (n = 2) attached to the Hailea ACO-9620 adjustable six-outlet air pump (Hailea Group Co. Ltd., Guangdong, China) because the bacteria in the hydrolysis agent is aerobic. During this time, agglomerated FOG, including FOG entrapped solids, were removed via scraping. Thereafter, the mixture was allowed to settle for 24h to reduce dissolved oxygen (DO) concentration in the mixture because the next treatment process was anaerobic. The pre-treatment process was operated at room temperature ranging from 20 to 23°C in a fed-batch mode to allow for the hydrolysis and agglomeration of the FOG. This resulted in a product on the third day, which was screened using two different stainless-steel sieves of pore sizes 1.18mm and 53µm respectively, before being fed into a 25L feeding tank that was continuously stirred using a magnetic stirrer under anaerobic conditions.

5.2.4. EGSB set-up and inoculation

Since the EGSB is a modification of the up-flow anaerobic sludge blanket (UASB) reactor, it relies on microorganism self-immobilization and the generation of well-settling granular biomass. It consists of an up-flow configuration. The wastewater was introduced at the bottom of the reactor, then passed through the anaerobic sludge-bed, which facilitates the conversion of organic matter into biogas, including other by-products collected at the top through a three-phase separator. The reason for the three-phase separator is to ensure the separation of biogas (gas phase) and treated wastewater (liquid phase) at the top of the reactor while the anaerobic biomass (solid phase) remains within the reactor (Mbulawa, 2017; Mchugh et al., 2003; Njoya et al., 2019). The difference between the EGSB and UASB is that the EGSB utilizes a higher up-flow velocity due to the high settleability of the granular sludge employed by the EGSB as its operation ensues and to maintain high methanogenic activity. The introduction of an effluent recirculation stream resulted in a slight expansion of the fluidized sludge-bed meant to improve the hydraulic mixing, and reduce gas entrapment while increasing the organic matter loading rate (Mbulawa, 2017; Basitere, 2017). Consequently, the EGSB is appropriate for treating high-strength wastewater with OLRs up to

30KgCOD/m³.d. When treating wastewater with a high FOG content, most of the FOG gets entrapped within the granular sludge, and over time, the FOG gradually disintegrates and is recovered through the recirculation stream. Thus this bioreactor's bed displays filtration capabilities (Njoya, 2017), and the efficiency of the EGSB was determined to be dependent on the retention of the granular sludge (Mbulawa, 2017). As a means of further increasing the contact time between the wastewater and the sludge granules, which in turn increases the production of biogas, the EGSB was designed to have a height/width ratio of 4-5 and a height/diameter ratio greater than 20 (Williams, 2017; Basitere, 2017).

The EGSB used in this study was made of clear polyvinylchloride (PVC) with an inner diameter (ID) of 0.0814m, a height of 0.612m and a working volume of 2L. Although a gas-liquid-solid separator was installed at the top of the EGSB, the utilization of the Eco-flush™ in the pre-treatment stage prevented the generation of biogas; hence, biogas was not collected during the operation of the plant. The liquid effluent was divided into two streams: 1) the recirculation stream, which was mixed with the effluent of the pre-treatment feeding tank, and 2) the EGSB product stream. About 50 glass marbles were used as the underdrain system for the EGSB to retain the granular sludge – see Figure S3. For the inoculation of the reactor, 0.4L of anaerobic granular sludge wastewater mixture with a TSS of 1 182mg/L and VSS of 24.5% was added, together with 1.6L of untreated PSW. This wastewater mixture was collected from a full-scale UASB reactor treating brewery wastewater at the local South African Brewery (SAB) plant located in Newlands, Cape Town, and stored in 25L polypropylene drums at 25°C. Mchugh *et al.* (2003) described anaerobic granular sludge as “particulate biofilms formed spontaneously by auto immobilization of anaerobic bacteria in the absence of a support material.” As such, it is constituted by a combination of interdependent anaerobic microorganisms essential for the methanogenic degradation of organic matter, and according to Mchugh *et al.* (2003), this sludge is a major contributor to modern, high-rate anaerobic digesters successfully treating high-strength wastewater. The inoculation stage, a milk solution was also added to provide the sludge microorganisms with the nutrients they required to grow (Rinquest *et al.*, 2019). Six scoops of Lactogen Starter Infant Formula 1 powder milk were added to 400mL of water, of which 200mL of the milk solution was added into the reactor. The recirculation stream, which was operated independently of the product and feed streams, was then switched on and left to run for 3 days before the reactor was put into operation to increase the tolerance of the microorganisms to the raw PSW.

5.2.5. EGSB start-up and operation

During the start-up period, the feed to the bioreactor was fed from the bottom using an Antech Aspendose A 5.1 L/0.5B peristaltic dosing pump (Enelsa Endüstriyel Elektronik Ltd., Antalya, Turkey) at a flow rate of 0.619L/h, and the recirculation stream was continuously operated at 1.29L/h. On the 8th day after start-up, there was minor sludge wash-out in the recirculation stream (Figure S5) and the biogas gas port, and this was due to the product stream not being pumped at the same rate as the feed, which led to an overflow in the reactor. As a result, the feed rate to the reactor was reduced to 0.350L/h for the entire duration of the study. The EGSB was operated at an upflow velocity (V_{up}) of 0.107m/h, an HRT of 5.71h, and the OLR fluctuated between 200 and 700mgCOD/L.h. A water bath was used to maintain the reactor's temperature between 33°C and 40°C, as noted by Sheldon and Erdogan (2016) due to mesophilic temperatures being ideal for the anaerobic digestion (AD) to take place.

5.2.6. Membrane tank set-up and inoculation

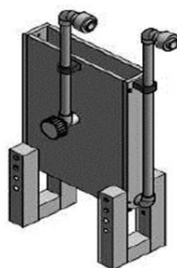
Membrane filtration is a pressure-driven process that uses semi-permeable membranes to separate small particles such as pathogens from wastewater. It is considered ideal for disinfection and water polishing to a point whereby the treated water meets the standards of potable water. Membrane systems are exceptional at removing residual nitrogen (N) and phosphorus (P). Membrane filtration does not require temperature monitoring; however, the presence of FOG may result in the formation of thick biofouling layers on the surface of the membrane, thus decreasing the permeation rate of the wastewater (Williams, 2017; Baker *et al.*, 2020; Rinqest, 2017). The two most frequently applied membrane filtrations for wastewater treatment are MF and UF due to the affordability of the membranes, including their life cycle (Rinqest, 2017). For the past 20 years, UF has been extensively used in the food processing industry for it is capable of providing a barrier for the removal of pathogens which could be critical for recycling the treated PSW (Sardari *et al.*, 2018).

The membrane tank made of high-density polyethylene (HDPE) was purchased from Maizey Plastics (Pty) in Cape Town, SA. The membrane tank comprised a 37.3L anoxic zone, and a 75.7L aerobic zone that contained the submerged UF membrane Nadir®-UP150 (Microdyn-Nadir GmbH, Wiesbaden, Germany) – see Figure S5. The membrane was made of hydrophilic polyethersulfone (PES) with a pore size of 0.04µm and a nominal M.W.C.O. of 150kDa, resulting in a high flux (permeability $\geq 285(\text{L}/\text{m}^2\cdot\text{h})/\text{bar}\cdot\text{g}$), low fouling and good cleanability. The membrane sheet had a thickness of 2mm, making it possible for a packing density to be twice that of plate membrane systems. The low specific weight allows for high crossflow rates,

and the active membrane surface was 0.34m². Product specifications of the membrane are listed in Table 5.2. The membrane tank was inoculated with 10L of the raw PSW, 90L of tap water and 90mL of the Eco-flush™, with an acclimatization period of 3 days.

Table 5.2: Membrane product specifications (dimensions).

MEMBRANE CHARACTERISTICS	Membrane	NADIR® UP150
	Membrane Polymer	Polyethersulfone (PES)
	Nominal Pore Size	0.04 µm
	Preservative	Glycerine 20% / Sodium benzoate 3%
MODULE SPECIFICATIONS	Housing Material Options	Polyvinyl chloride (PVC)
	Drainage Layer	Polyester (PET)
	Nominal Membrane Area	0.37 m ²
PHYSICAL DIMENSIONS	Dry weight	6.73 kg
	Length	315 mm
	Width	182 mm
	Height	696 mm



5.2.7. Membrane tank start-up and operation

The product of the EGSB was fed into the membrane tank (see Table 5.2 for specifications) from the top of the tank using an Antech Aspendose A 5.1L/0.5B peristaltic dosing pump (Enelsa Endüstriyel Elektronik, Antalya, Turkey), with the membrane unit operating at a pressure of -0.4bar, at a flow rate of 0.717L/h, and the permeate was discharged at the top of the tank at a flow rate of 0.813L/h. The membrane tank was operated at ambient temperature (20–24°C), and the OLR varied between 50-450mgCOD/L.h. After 120 days of the MBR unit, implementing a soak cycle was deemed not necessary, as the unit still maintained the initial flux.

5.2.8. Sampling and analytical methods

Two samples of the raw PSW and effluents of the pre-treatment, EGSB and membrane tank units were collected in 1L sample bottles three times a week. One of the two samples was

analysed for temperature, pH, electrical conductivity (EC) and DO using a Lovibond SensoDirect 150 Water Testing Multimeter (Tintometer GmbH, Dortmund, Germany). The second samples collected during the week were combined to form a weekly representative sample that was sent to an independent SANAS accredited laboratory (Scientific Services, Cape Town, SA) for the analyses of alkalinity, COD, FOG and TSS EPA method 310.1, EPA method 410.4, EPA method 10056 and EPA method 160.2.

5.3. Results and discussion

5.3.1. Poultry Slaughterhouse Wastewater characteristics

The chemical analysis results for the raw PSW used in this study, pre-treatment and EGSB product streams, and submerged membrane permeate were compared to the CCT discharge by-laws and DWS standards effluent discharge (see Table 5.3). The average COD, FOG and TSS of the raw PSW were 5 280mg/L, 510mg/L and 1207mg/L, respectively and were above the CCT discharge standards as expected. However, the pH of 6.61 and the electrical conductivity (EC) of 215.4mS/m were within the CCT discharge standards.

Table 5.3: Raw PSW characteristics compared to the CCT discharge by-laws and the DWS standards of effluent discharge (Council of the City of Cape Town, 2014; DWS, 2017).

Parameter	Raw PSW			Pre-treatment Product			EGSB Product			MBR Permeate			CCT Discharge By-Laws	Standards for Effluent Discharge
	Min	Max	Ave	Min	Max	Ave	Min	Max	Ave	Min	Max	Ave	Not to exceed	General Limits
pH	6.57	6.71	6.61	6.82	6.97	6.88	6.81	6.91	6.86	6.5	8.12	7.685	12	5.5 - 9.5
Alkalinity (mg/L)	1005	1019	1012	492	993	709	264	1196	735	105	294	246	-	-
Electrical conductivity, EC at 25 °C (mS/m)	171.2	331.3	215.4	61.4	102.4	78.6	56.8	78.8	67.5	-	-	-	500	150
Dissolved oxygen, DO (%)	3.7	7.2	5.45	0.85	2.3	1.76	2.6	3.40	3	8	10.7	9.3	-	-
OLR (mgCOD/L.day)	-	-	-	-	-	-	154	717	417	69	456	190	-	-
COD (mg/L)	3210	13250	5280	881	4092	2382	394	2602	1085	68	134	100	5000	75
FOG (mg/L)	35	2870	510	8	1478	182	4	45	25	2	15	8	400	2.5
TSS (mg/L)	198	3100	1207	100	2650	576	13	308	152	5	13	7	1000	25

5.3.2. Evaluation of the pre-treatment performance

It was observed that the pungent odour of the raw PSW was reduced drastically within an hour after adding the Eco-flush™ to the pre-treatment unit. Therefore, it was deduced that the microbial consortium present in the Eco-flush™ was successful in generating enzymes capable of oxidising H₂S, resulting in the reduction of odour. On the third day of pre-treatment, there was a light-brown, very fine, fluffy scum floating on the surface of the water while denser particles sank to the bottom of the tank, as indicated in Figure 5.2 (a). The Eco-flush™ therefore functioned as a flocculant, for it caused the aggregation of fine suspended solids (SS), including some FOG. After screening, the agglomerated FOG-SS flocs that were

removed consisted of feathers, small meat particles and other small solids, as depicted in Figure 5.2 (b).



(a) Pre-treatment tank on the third day.

(b) Screening residue.

Figure 5.2: Illustration of the recovered solids from the pre-treatment process.

Figure 5.3 depicts the pre-treatment performance with regards to the removal of COD, FOG and TSS. The graphs in the first row indicate the variation of the concentration for each parameter in the feed, while those in the second row depict the effluent stream concentrations. The third row illustrates the REs for the same parameters. As indicated by Table 5.3, the average FOG content in the pre-treatment product stream was reduced to 182mg/L, which met the discharge standard of the CCT. For this stage, the average RE achieved was 66%. After the pre-treatment process, the pH and EC fluctuated from 6.82 to 6.92 and 61.4 to 102.4mS/m, respectively, and these were within the standards for effluent discharge, as indicated by Table 5.3. Baker *et al.* (2020) mentioned that sieves could be used for the removal of TSS, and according to Bustillo-Lecompte and Mehrvar (2015), screening is capable of removing up to 60% solids, and in this study, the average TSS RE attained by the pre-treatment system was only 49%, which is within the range. The effluent COD ranged between 881mg/L and 4 092mg/L,33 which was below the CCT discharge standards, and the average RE of COD achieved was 52% in the pre-treatment unit. The DO present in the pre-treatment product stream was of interest, as the EGSB is an anaerobic process. An average DO percentage of 1.76% maintained in the pre-treatment unit was low enough for anaerobic digestion to take place in the EGSB using the effluent from the pre-treatment stage.

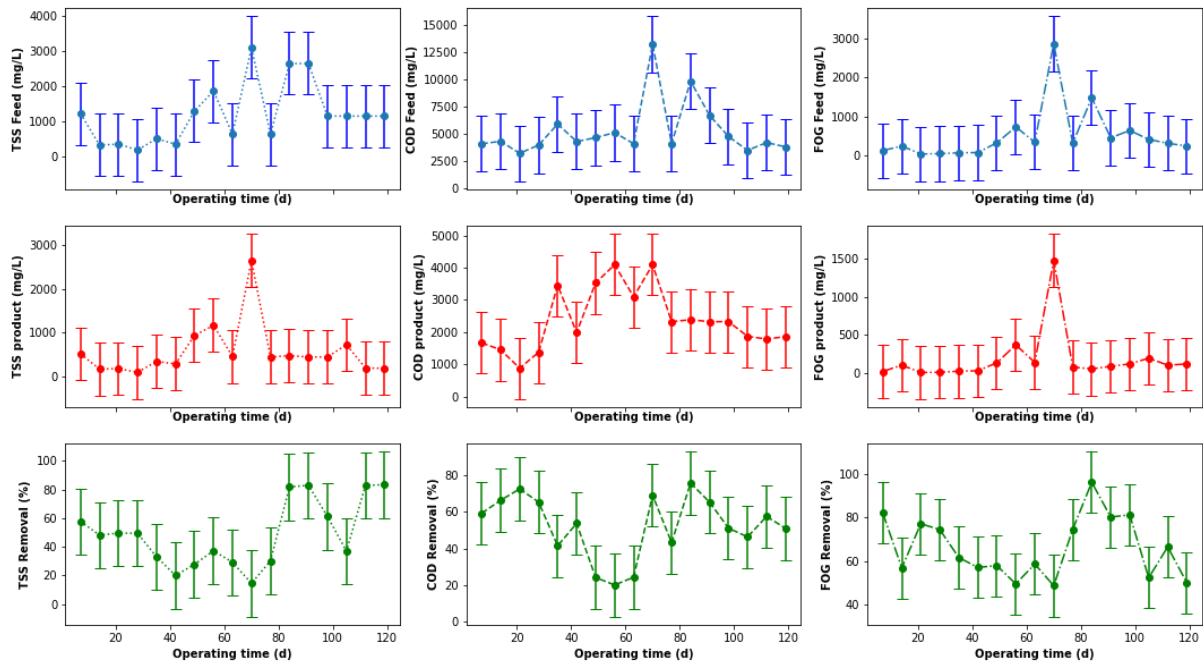
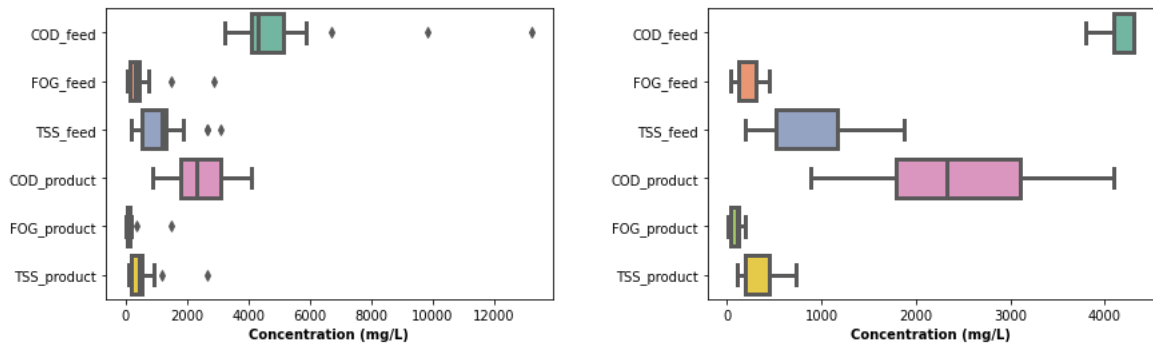


Figure 5.3: Performance of the pre-treatment stage before data processing.

To evaluate the veracity of the fluctuations in concentrations and REs achieved as illustrated in Figure 5.3, visual outlier detection was implemented using boxplots (See Figure 5.4 (a)) to detect outliers from each distribution evaluated. Outliers were noticed in the COD, FOG, and TSS concentrations in the feed stream to the pre-treatment process. Similarly, outliers were also identified in the concentration of the FOG and TSS in the effluent of the pre-treatment stage, as depicted in Figure 5.4 (a). These outliers could be numerically detected by using the interquartile range rule (where the box symbolises the middle 50% of the ranked data and is drawn from the lower quartile value, which is the 25th until the 75th percentile), and replaced or eliminated using the mean or median value (the middle data observation in the ranked data of any dataset and as a measure for central tendency of the data, i.e. similar as the 50th percentile value of a data set) of each distribution (Nadiatul Adilah *et al.*, 2020). However, in this study, the outliers were replaced by the median of respective distributions to keep the parameters within respective interquartile ranges. The result of this data processing is depicted in Figure 5.4 (b), from which an absence of outliers is evident.



(a) Boxplots before the replacement of outliers (b) Boxplots after the replacement of outliers

Figure 5.4: Boxplots of the distribution of performance features before and after the replacement of outliers.

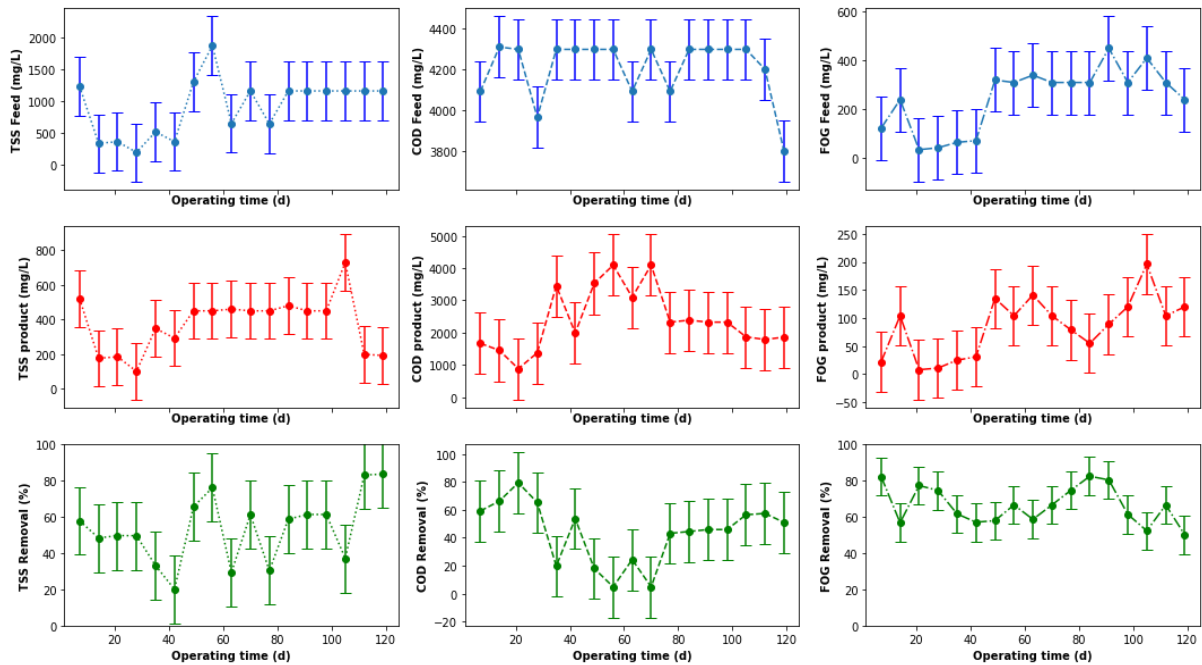


Figure 5.5: Performance of the pre-treatment stage after data processing.

This outlier replacement led to a second evaluation of the performance of the pre-treatment stage, as illustrated in Figure 5.5. From the third row, it can be observed that the COD, TSS, and FOG REs varied less than the initial data processing done and therefore displayed a more stable, even distribution.

Figure 5.6 provides a correlation matrix between the three REs investigated in this study after the data processing, with univariate distributions across the diagonal of the correlation matrix,

heatmaps of bivariate distributions above the diagonal and bivariate density contours under the diagonal. This matrix correlation does not display a strong correlation between the three REs as illustrated by the bivariate density contours, which means that the pre-treatment stage performed differently to remove the COD, FOG, and TSS.

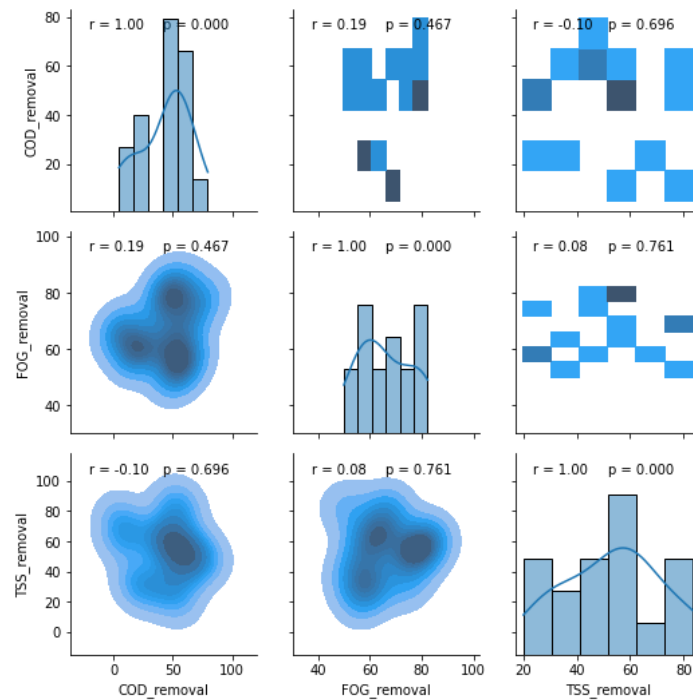


Figure 5.6: Correlation matrix between the COD, FOG and TSS.

This was further noticed in Figure 5.7, which provides the distribution, skewness, and kurtosis of the distribution of the COD, FOG, and TSS REs in the pre-treatment stage of the PSW. It was observed that the REs of the FOG are distributed within a smaller range than the distribution of the COD and TSS REs in the same treatment stage, with REs varying between 50 and 82% and a mean value of 66%. This was also confirmed by the REs observed, with the FOG standard deviation being the lowest. The REs for TSS ranged from 20% to 82%, with a mean value of 53%. Of the three REs investigated, the removal values of the COD were scattered across a broader range than the others, with values varying between 5 and 80% and a mean RE of about 44%. These observations imply that the pre-treatment stage was more effective in removing FOG than other contaminants present in the PSW, which can be attributed to the Eco-flush™ facilitating FOG removal in the raw PSW.

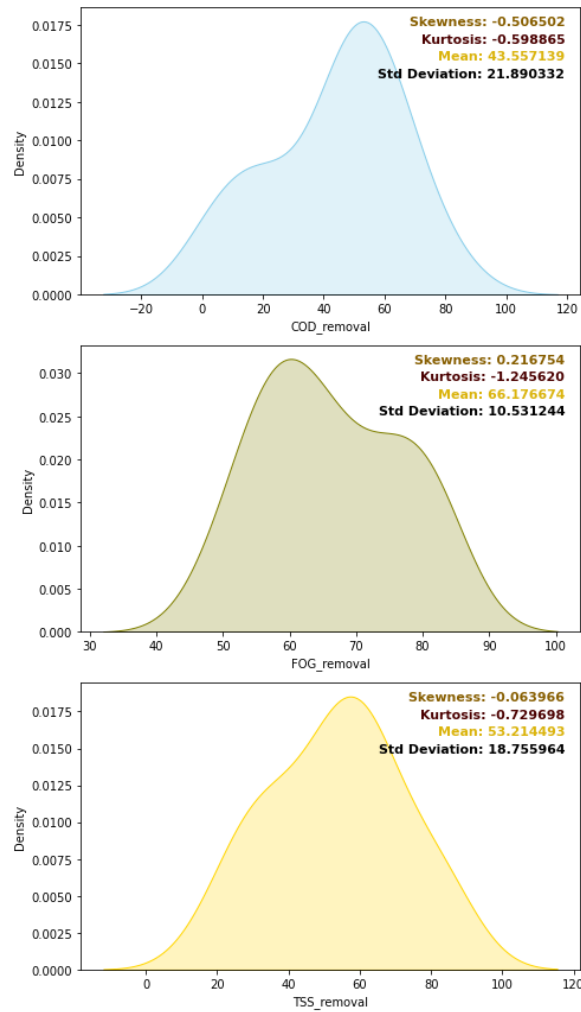


Figure 5.7: Distribution of the COD, FOG and TSS REs.

Figure 5.8 depicts the colour change of the raw PSW from brick brown to clear mustard after the screening process. The bottle on the left contains raw PSW, the bottle on the right contains pre-treated PSW before the screening, and the bottle in the middle contains pre-treated PSW after screening. Upon close examination of the bottles, it was noted that the bottle on the right contained small particles, resulting in the sample looking cloudy, while the bottle in the middle had a clearer appearance resulting from the screening process.



Figure 5.8: Bottles containing raw PSW (left), pre-treated PSW after screening (middle) and pre-treated PSW before screening (right).

5.3.3. Evaluation of the EGSB performance

Hydrolysis being the first stage of AD, increased production of non-desirable VFA must be minimal. Irshad *et al.* (2016) stated that the increased production of VFA causes the hydrolysis process to stop by reducing the pH of the wastewater being treated. Therefore, the pH needs to be maintained between 7.0 and 7.2 to ensure that the first stage of AD takes place. According to Basitere *et al.* (2016), the optimal pH conditions for methanogens activity to occur is 6.5-8. Throughout this study, the pH of the wastewater being treated ranged between 6.5 and 8.12, therefore, it can be deduced that there was no increased production of VFA, and the organic matter degradation process was not compromised.

The presence of FOG can hinder the efficiency of any biological treatment by reducing the biodegradation process due to blockage and clogging of piping systems and the encapsulation of the granules with grease or oil; therefore, the success of the anaerobic process is dependent on the physico-chemical pre-treatment used prior to feeding the wastewater to the anaerobic digester (Mbulawa, 2017; Rinquest, 2017). The pre-treatment process used in this study was highly efficient at removing the FOG content in the raw PSW as the effluent FOG from this stage was consistently lower than the CCT discharge by-law limit of 400mg/L. Therefore, there was no clogging in the EGSB, and its degradation process was not compromised by the FOG content in the feed stream. The EGSB further reduced the FOG content of the wastewater, with 67% being removed. This was due to the high microbial activity, which ensured the entrapment and further hydrolysis of the FOG present in the pre-treated PSW feed (Yetilmezsoy *et al.*, 2015).

The average REs achieved by the EGSB were 54% COD and 68% TSS, which was less than the 69% COD and 98% TSS achieved by the lab-scale EGSB that was used in the study conducted by Williams (2017). From the average overall REs of the pre-treatment coupled with the EGSB, it can be resolved that the treatment was successful; however, the characteristics of the EGSB effluent indicated that the treated PSW still required further treatment to meet the DWS effluent discharge standards of 75mg/L COD, 25mg/L FOG and 25mg/L TSS. The same data processing and data analysis methodologies were applied to evaluate the performance of the EGSB. Figure 5.9 provides the variation of the concentrations and REs of the COD, FOG, and TSS throughout the study.

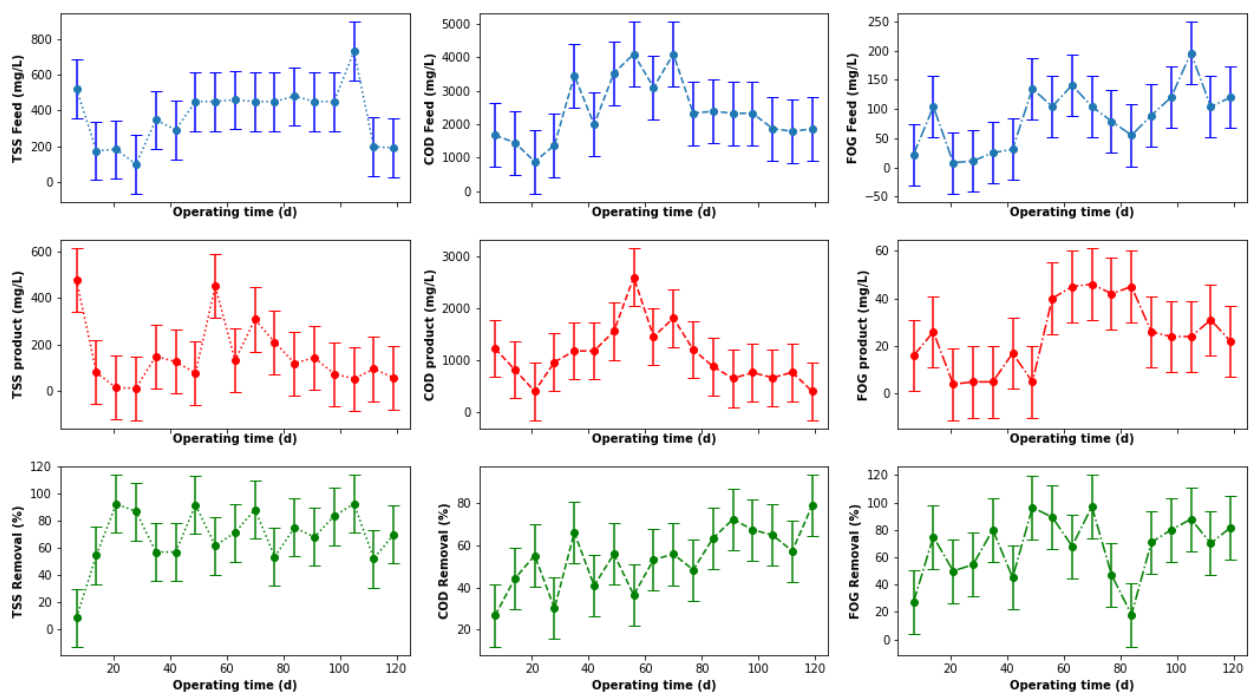
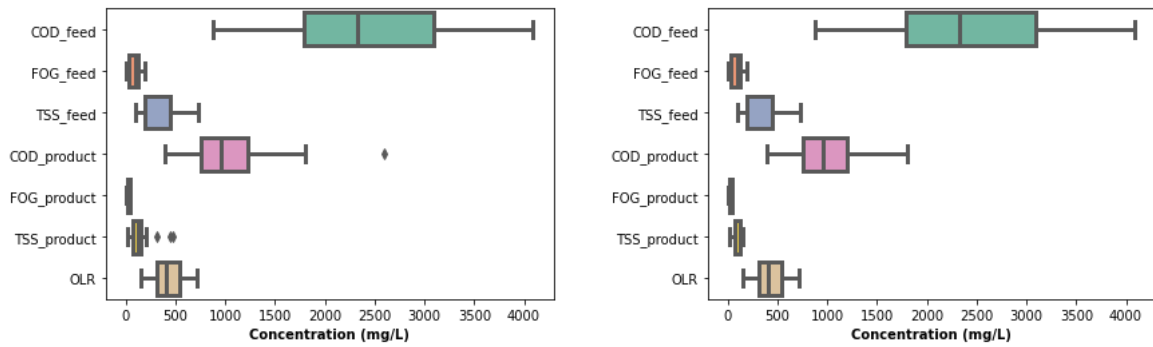


Figure 5.9: Performance of the EGSB before data processing.

From Figure 5.10 (a), it can be observed that outliers were present in the distribution of the concentration values for the COD and TSS in the effluent of the EGSB. The median of relevant distributions replaced these outliers, and the new distribution is illustrated in Figure 5.11. Figure 5.10 (b) depicts the absence of outliers in the new distribution values, which allowed further data analyses.



(a) Boxplot before the replacement of outliers (b) boxplot after the replacement of outliers

Figure 5.10: Boxplots of the performances before and after the replacements of outliers.

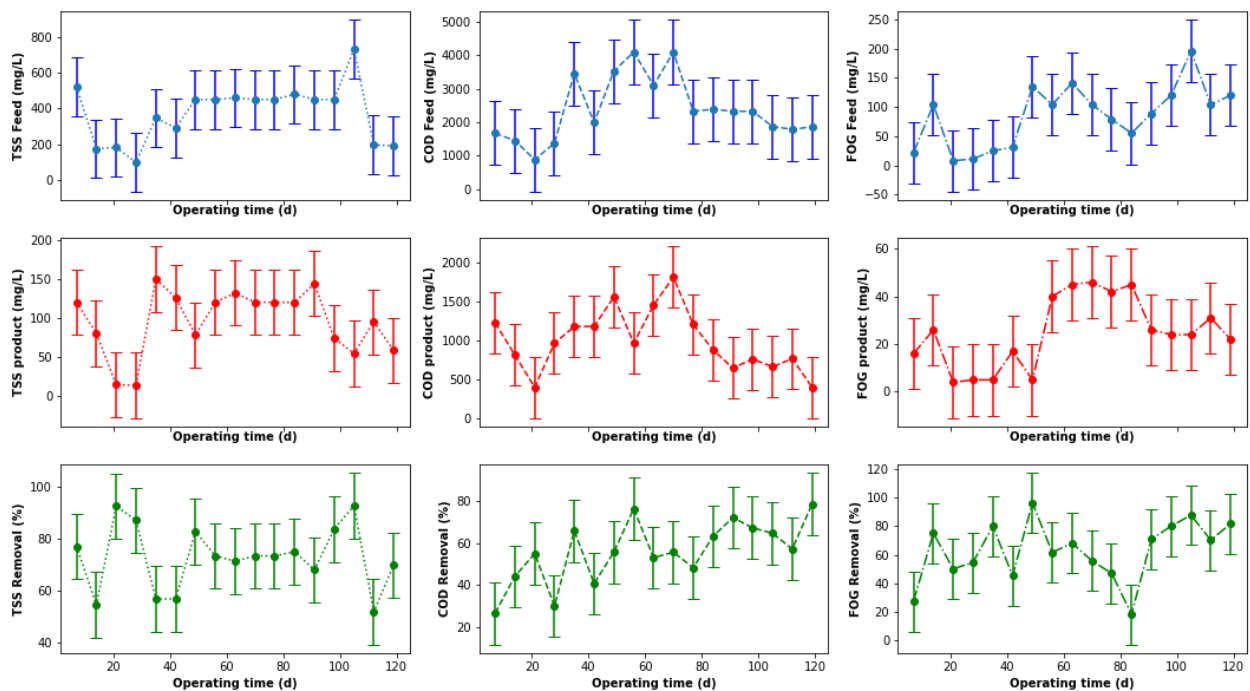


Figure 5.11: Performance of the EGSB after the replacement of outliers.

Therefore, there was a fluctuation in the OLR as expected, with a minimum of 200mgCOD/L.h and a maximum of 700mgCOD/L.h. In a study conducted by Lim and Fox (2011), where swine wastewater treatment was conducted at room temperature using an SGBR, it was observed that the COD RE was proportional to the OLR, as earlier studies by other researchers have indicated. In a study conducted by Rinquest *et al.* (2019), whereby the treatment efficiency of an SGBR-SSND-ufMM system treating PSW was investigated, the REs of COD, TSS and BOD increased with increasing OLR, while the FOG RE was not a function of the OLR. Thus, it was expected that a similar trend would be observed in this study; however, this was not the case, the REs were not a function of the OLR, and no trend was established. The variation of

the RE for FOG, COD, and TSS with the change in OLR throughout the study can be observed in Figures 5.12 (a), (b), and (c), respectively. These figures show that the EGSB performed adequately for the removal of FOG and TSS from PSW, and to a lesser extent, the removal of COD. There was no observable trend of increased REs with increased OLR.

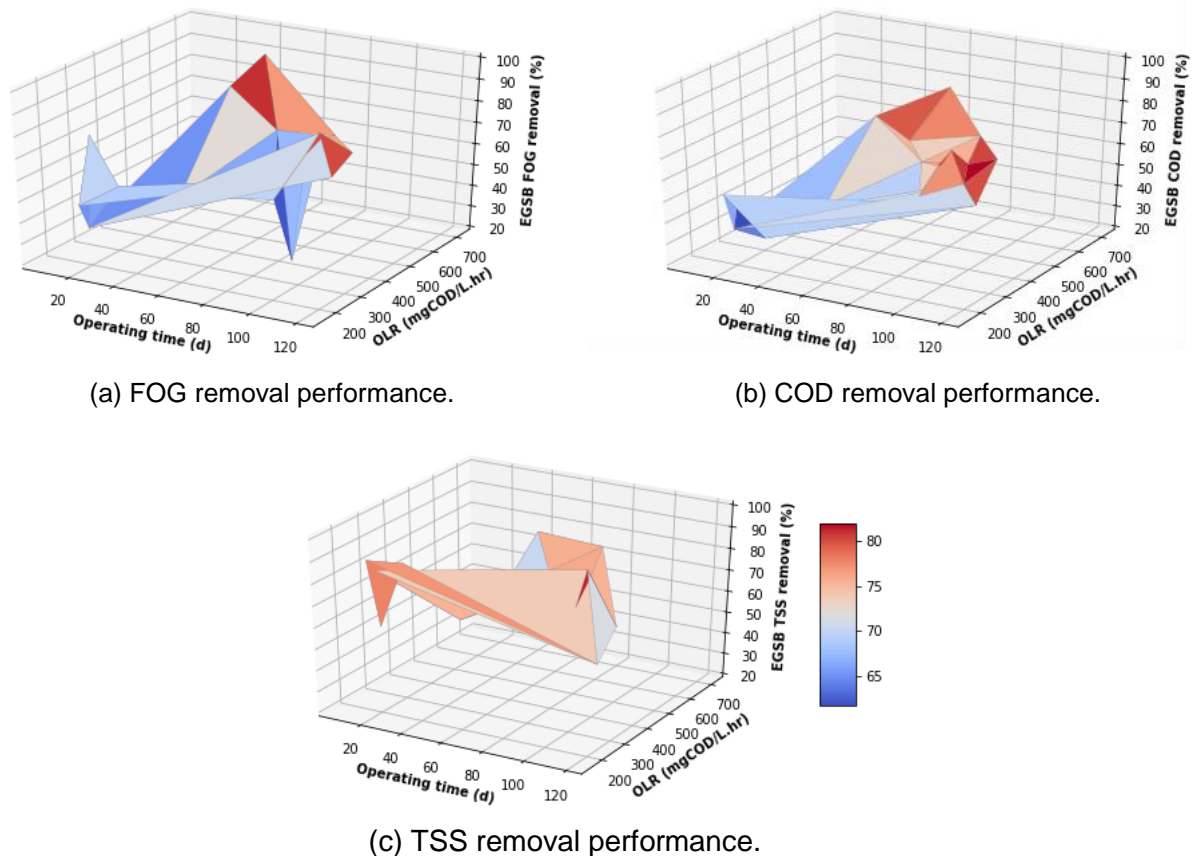


Figure 5.12: REs for the three parameters concerning operational time and varying OLR.

The investigation of a significant correlation between the RE of each parameter is shown in Figure 5.13, which depicts a lack of a significant correlation between the COD, FOG, and TSS REs. However, it can be noticed that the average percentage removal for each parameter was above 50%, with maximum REs reaching 99% for the FOG, 92% for the TSS, and 78% for the COD. Figure 5.14 further confirms these observations and provides more details on the skewness, kurtosis, and distribution range of each RE. It can be observed that the distribution of the TSS removal in the EGSB is highly skewed and heavy-tailed. Figure 5.11 reveals that this skewness and the high kurtosis values of the TSS removal can be attributed to low values concentration of TSS in the inlet stream to the EGSB. As it pertains to the skewness of the COD and FOG REs, these can be attributed to the poor performance of the EGSB at the

beginning of the treatment process. This was partly due to the period required for the acclimatization of the anaerobic bacteria inoculated inside the EGSB at the beginning of the EGSB operation. Figure 5.14 further provides the distribution range of each RE, and the mean of each distribution, with the mean of TSS being the highest of the three REs with a value slightly above 72%, indicating that the EGSB was more efficient at removing TSS from the wastewater.

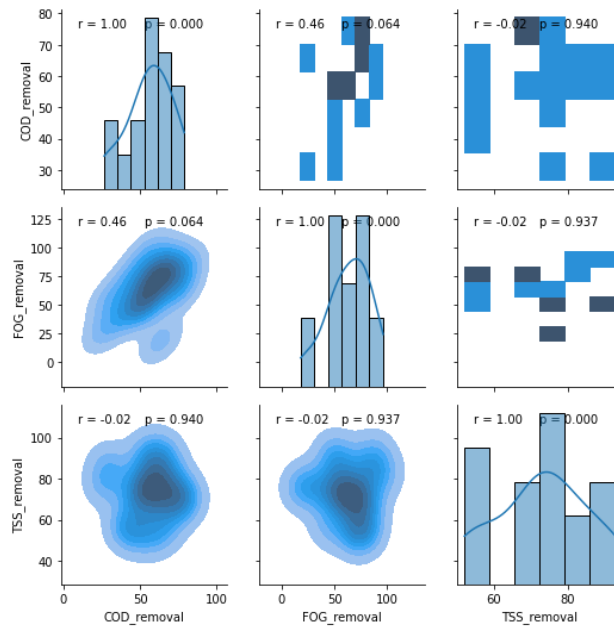


Figure 5.13: Correlation matrix of the COD, FOG and TSS REs in the EGSB.

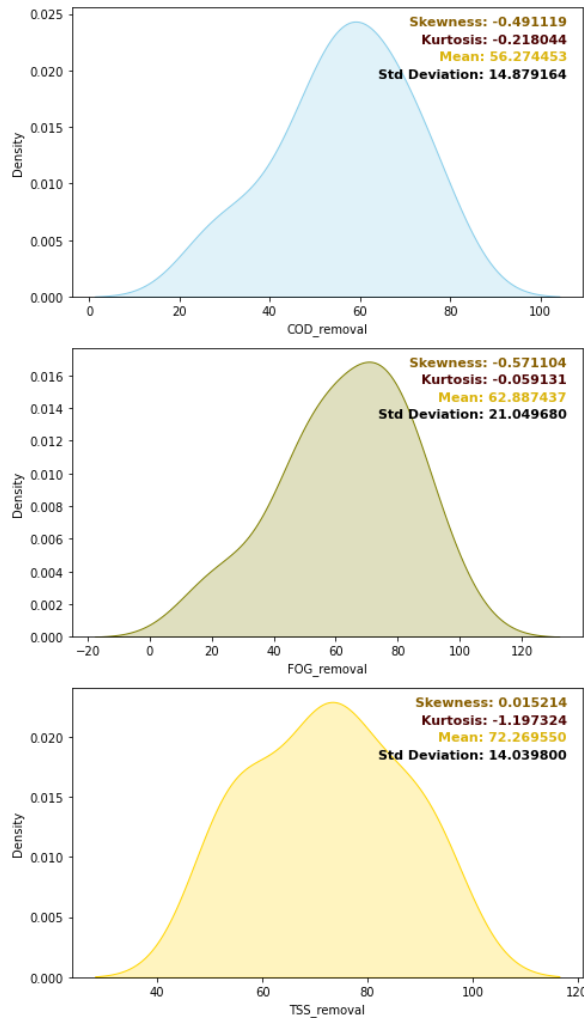


Figure 5.14: Distribution of the COD, FOG and TSS REs in the EGSB.

5.3.4. Evaluation of the membrane tank performance

The membrane system permeate's average COD, FOG and TSS content was 101mg/L, 8mg/L and 7mg/L, respectively. This indicated that further treatment might be required for the treated water to meet potable water standards. The average REs attained were 88% COD, 64% FOG and 90% TSS, which was higher than the 64% for COD, 48% for FOG and 88% for TSS REs accomplished by Basitere *et al.* (2017). This suggests that a comparative analysis using other modified membrane materials must be conducted to assess whether further improvements can be attained. Figure 4.15 depicts the variation of the concentration and REs of the COD, FOG, and TSS of the PSW for the membrane system. As illustrated in Figure 5.16, there were no outliers in each distribution; therefore, there was no need to implement a second analysis of the performance of the submerged membrane tank. Overall, as illustrated in the third row of Figure 5.15, the membrane performed well to remove COD, FOG, and TSS, with maximum REs close to 99% for the TSS, 96% for the COD and 81% for the FOG. This indicates that the performance of the membrane was not hindered, and the fact that the pre-treatment stage

was highly effective at removing the FOG present in the PSW contributed to the membrane not experiencing any fouling during the treatment process. Therefore, the addition of the Eco-flush™ successfully mitigated the challenges of clogging and fouling in the treatment units used in this study.

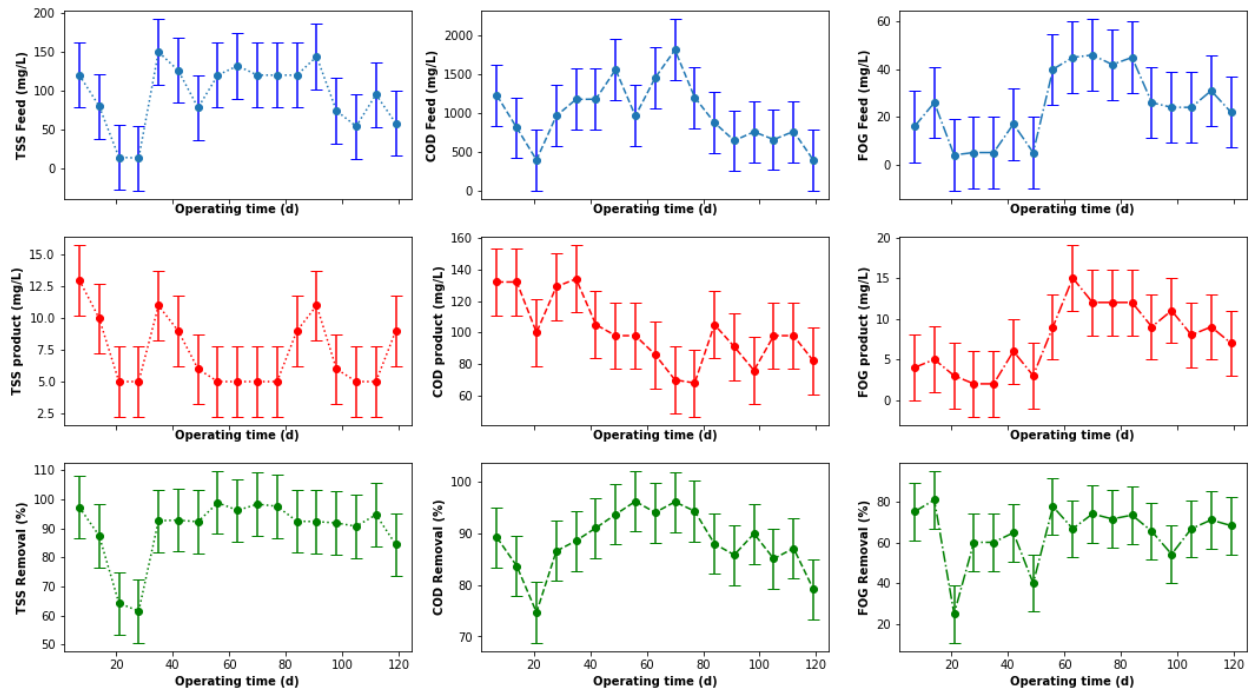


Figure 5.15: Performance of the submerged membrane unit.

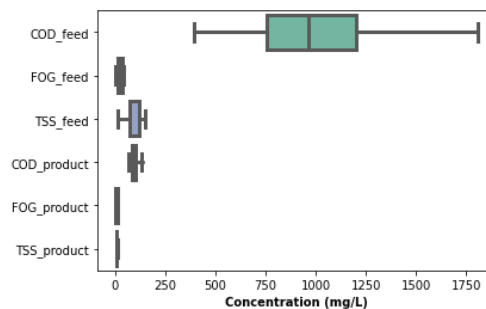
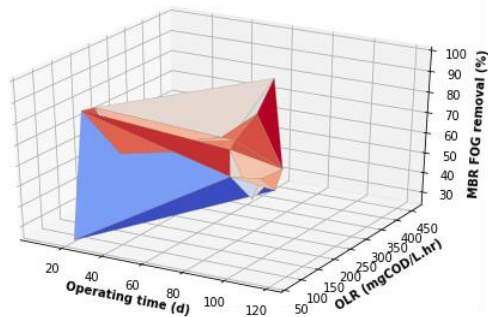


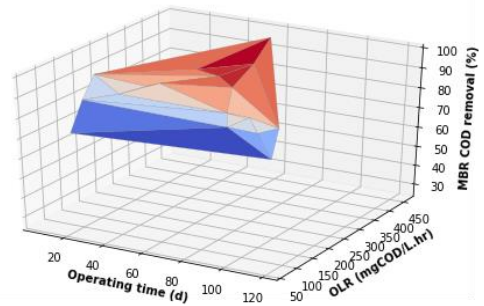
Figure 5.16: Boxplot of the COD, FOG and TSS at the feed and effluent of the submerged membrane.

When analysing the results of the membrane unit, it was noted that the post-treatment had the most stable performance for the removal of COD with REs ranging from 75 to 96%, followed by the TSS REs maintained within the range of 62 to 99%; and lastly, the FOG REs which had a wider range of 25 to 81% removal, albeit at OLRs varying between 69 and 456mgCOD/L.h.

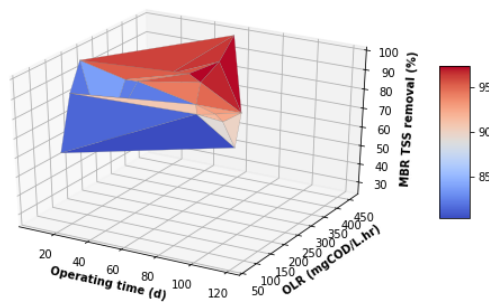
Figures 5.17 (a), (b), and (c) illustrate the performance of the membrane unit with the variation of the OLR throughout the study concerning FOG, COD, and TSS removal percentages, respectively. A comparison between Figures 5.12 (a), (b), and (c) and Figures 5.17 (a), (b), and (c) also shows that the membrane unit was dealing with lower OLRs than the EGSB.



(a) FOG removal performance.



(b) COD removal performance.



(c) TSS removal performance.

Figure 5.17: REs for the three parameters concerning operational time and varying OLR.

The correlation matrix between the REs of the COD, FOG, and TSS is illustrated in Figure 5.18, from which an insignificant correlation was observed between the parameters investigated. Further analyses of the distribution of the REs is provided in Figure 5.19, from which the density, skewness, kurtosis, and mean of each distribution are provided. From the same figure, it can be observed that the membrane performed well for the removal of COD and TSS, with mean removal values of 88% and 90%, respectively; however, the distribution of the TSS REs was more skewed.

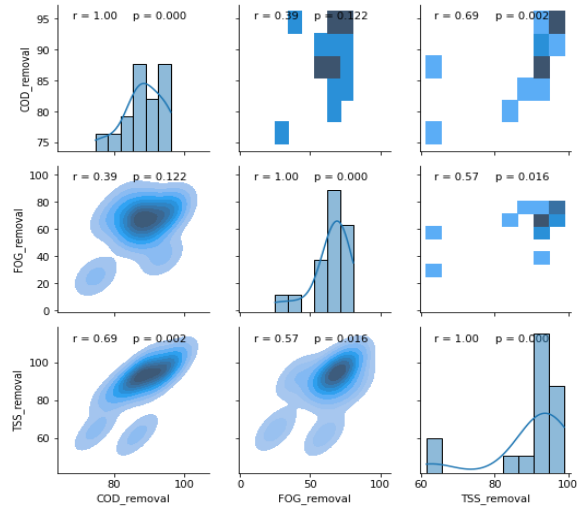


Figure 5.18: Correlation matrix of the COD, FOG and TSS REs in the submerged membrane.

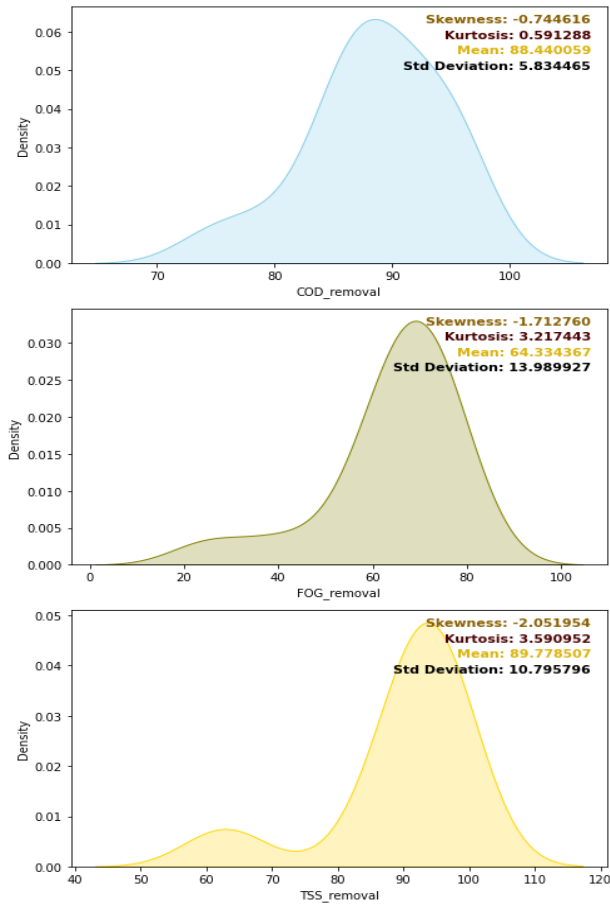


Figure 5.19: Distribution of the COD, FOG and TSS REs in the submerged membrane.

5.3.5. Overall treatment performance

The overall REs of the lab-scale PSW treatment plant was consistently above 90%, averaging at 98% COD, 97% FOG and 99% TSS. Overall, as depicted in Figure 5.20, the combination of the pre-treatment stage, the EGSB, and the submerged membrane provided worthy COD, FOG and TSS REs, especially after day 40, when the removal percentages remained high and fluctuated less for the three parameters evaluated. As verified in Figure 5.21, there were no outliers in the distribution of the parameters, which consolidates the observations made.

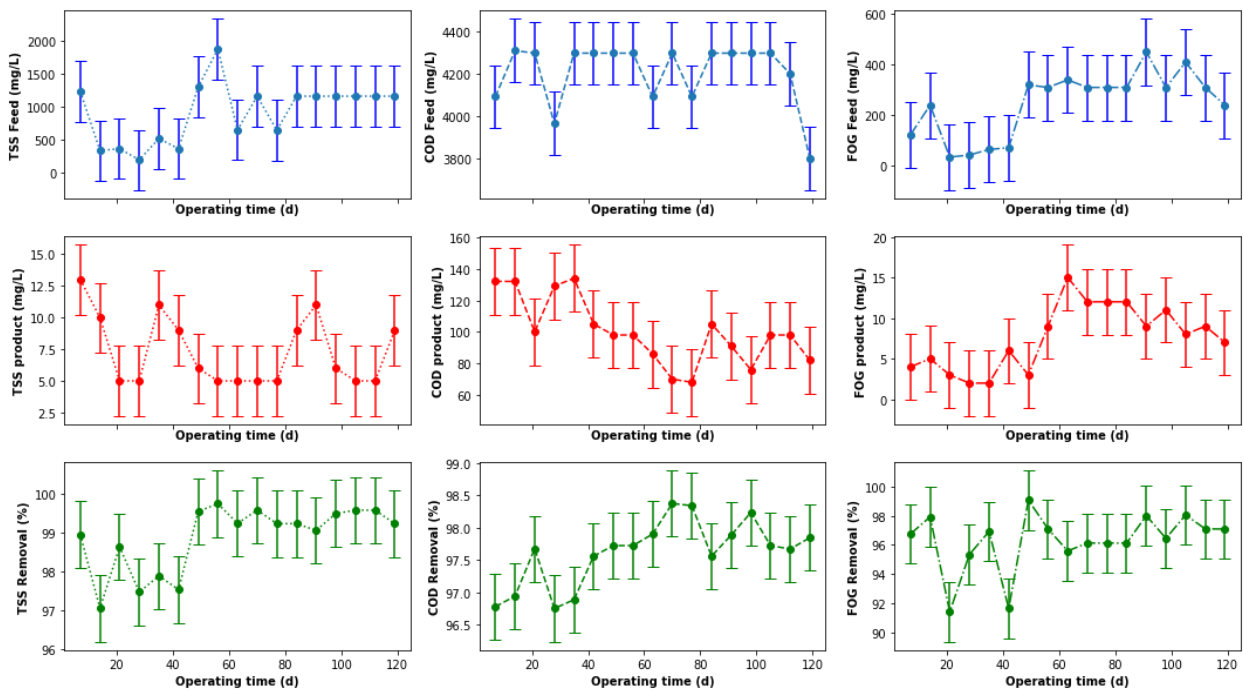


Figure 5.20: Performance of the overall treatment system.

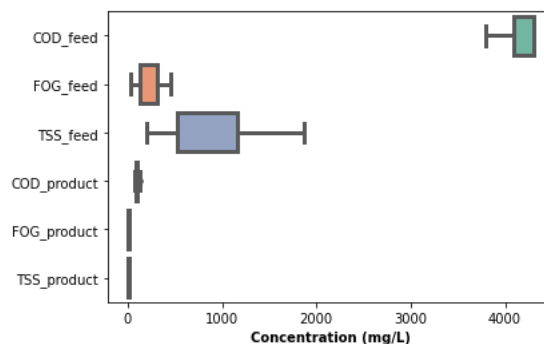


Figure 5.21: Boxplots of the COD, FOG and TSS at the feed and effluent of the entire system.

The observation of the correlation matrix provided in Figure 5.22 reveals a correlation between the TSS and COD REs, but there was a high skewness of the distribution of TSS REs, as depicted in Figure 5.23. However, Figure 5.23 also shows that the overall performance of the entire system had a mean above 95% for the COD, FOG, and TSS REs, and the standard deviation values confirm this. This suggests that this treatment system is suitable for the treatment of PSW or similar high-strength wastewater.

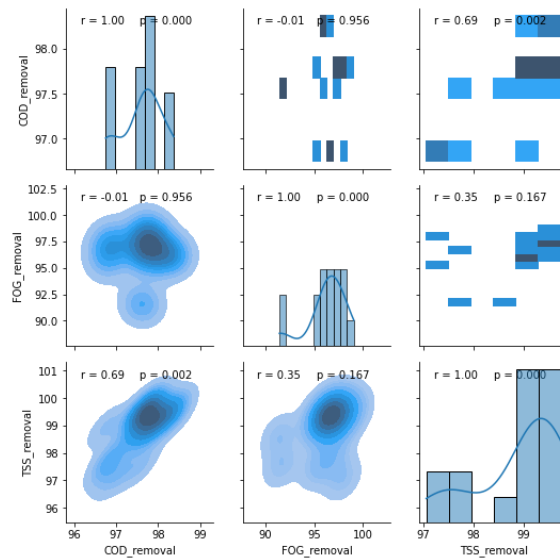


Figure 5.22: Correlation matrix of the COD, FOG and TSS overall REs.

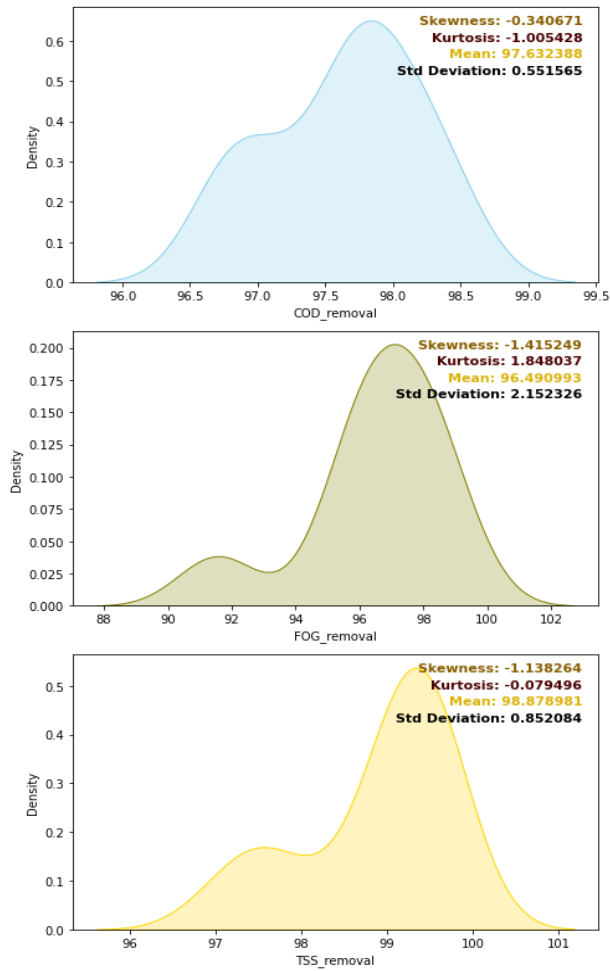


Figure 5.23: Distribution of the COD, FOG and TSS REs in the entire treatment process.

Table 5.4 displays a comparison of the current study to other integrated multi-stage treatment processes.

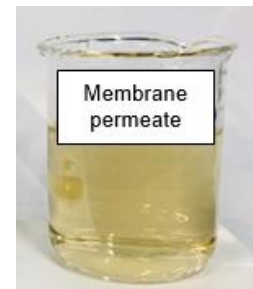
Table 5.4: Comparison of integrated multi-stage treatment systems treating PSW.

Technology Used	Wastewater treated	Results	References
Aerobic pre-treatment connected to an EGSB and UF submerged membrane	PSW	97.6% COD, 96.5% FOG, 98.9% TSS	Current study
Electrochemical pre-treatment connected to a UF module and RO module	PSW	99.7% COD, 99.7% TSS	(Meiramkulova <i>et al.</i> , 2020a)
SGBR connected to an SSND and ufMM	PSW	91% COD, 97% TSS	(Rinquest <i>et al.</i> , 2019)
SGBR connected to a UF membrane	PSW	98% COD, 92.4% FOG, 99.8% TSS	(Basitere <i>et al.</i> , 2017)
EGSB connected to an SSND and MBR	PSW	99% COD, 92%TSS	(Williams, 2017)
UASB connected to packed-bed with polyethylene rings	Swine wastewater	99% COD	(Gonzalez-Tineo <i>et al.</i> , 2020)

Upon examination of the appearance of the wastewater at the different treatment stages, it is evident that the membrane permeate is cleaner, as depicted in Figure 5.24. The image of the treated PSW in the beaker is a sample of the submerged membrane permeate.



(a) Treated PSW at different treatment stages



(b) Sample of membrane permeate

Figure 5.24: Samples of the treated PSW.

5.3.6. Summary

The aerobic pre-treatment process proved to be highly efficient in removing organic constituents present in the raw PSW because, on average, the effluent content was within the CCT discharge by-laws. The average REs achieved by this process for COD, FOG and TSS were 50%, 62% and 56%, respectively. The EGSB successfully removed on average 56% COD, 63% FOG and 73% TSS. The submerged membrane had the most stable RE performance, achieving average REs of 88% for COD, 64% for FOG and 90% for TSS. The overall treatment of the pilot plant achieved average REs of 98% COD, 97% FOG and 99%

TSS successfully reducing the content of the treated wastewater to 215.3mg/L COD, 24.3mg/L FOG and 15.3mg/L TSS and rendering it safe for discharge into the CCT WWTWs. However, the effluent did not meet the DWS standards for effluent discharge. Therefore, it is recommended that a further treatment stage, such as UV radiation, be implemented to deactivate pathogens adding additional chemicals to the system that might result in the formation of harmful by-products. It is also recommended that 1) a comparative analysis be conducted to assess other modified membrane materials in order to determine whether further RE improvements can be attained and 2) oxidation mechanisms of pollutants in particular for the treatment stage be studied.

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Declaration of competing interest

The authors declare no conflict of interest.

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CHAPTER SIX

Conclusions and Recommendations

6.1. Conclusions

In this study, three basic models, namely single-stage, two-stage and three-stage, were developed using Sumo19 simulator software to simulate the performance of a lab-scale anaerobic digester treating PSW. The models were assessed to predict the removal of organic matter, TSS and VSS in PSW. The resultant effluents of the single-stage and two-stage AD were compliant with the CCT wastewater and industrial effluent by-laws standards. However, there was a miniature increase in the quantity of NH_3 and PO_4^{3-} present in the effluent of both models. With the three-stage model, the effluent did not meet the CCT discharge standards; thus, it can be concluded that it was ineffective at treating PSW and that anaerobic treatment on its own is not sufficient for the complete treatment of PSW.

Secondly, the performance of a lab-scale treatment plant consisting of an aerobic pre-treatment tank to which a bioremediation agent called Eco-flush™ was added, screening sieves, an EGSB and submerged membrane, treating PSW collected from a poultry slaughterhouse located in the WC, was evaluated with regards to the REs of COD, FOG, and TSS. The results obtained were compared to the CCT discharge by-laws and the DWS standards of effluent discharge to determine whether the treated PSW would be suitable for discharge into the CCT WWTWs or on-site re-use since generally abattoirs struggle to meet these by-laws and there is a limited number of WWTRs that are in working condition. The data from the treatment pilot plant was used to develop a model that simulated the treatment plant's performance using Sumo, a wastewater process simulator developed for municipal and industrial wastewater modelling.

Due to the fact that the on average the contents of the pre-treatment effluent were within the CCT discharge by-laws, it is evident that the aerobic pre-treatment process proved to be highly efficient in removing organic constituents present in the raw PSW. The average REs achieved were 52% COD, 66% FOG and 49% TSS; therefore, the pre-treatment process was more effective at removing FOG, and this was because of the addition of Ecoflush™. The EGSB performed slightly better than the pre-treatment stage and successfully removed on average 54% COD, 67% FOG and 68% TSS with the OLR of the feed fluctuating between 200 and 700mgCOD/L.hr. Visual outliers were detected in the pre-treatment and EGSB processes through the use of boxplots, and these outliers were replaced with the median of the respective distributions, which allowed for a second analysis of the performance of the two processes. After the data processing, the REs varied less resulting in the REs of the pre-treatment being 44% for COD, 66% for FOG and 53% for TSS, while those for the EGSB changed to 56%

COD, 63% FOG and 73% TSS. Even though the combined treatment of the pre-treatment and the EGSB could reduce the organic content of the treated water considerably, the effluent still required further treatment to meet the DWS effluent discharge standards. The post-treatment, submerged membrane treatment process, showed the most performance, and no outliers were detected. In the membrane tank, the OLR ranged between 69 and 450mgCOD/L.hr and the REs attained by the post-treatment for COD, FOG and TSS were 88%, 64% and 90%, respectively. The correlation matrices performed on the REs for the three treatment stages indicated that there were no relationships between them. The overall treatment of the pilot plant achieved average REs of 98% COD, 96% FOG, and 99% TSS. Despite the MBR reducing the content of the treated water to 215.3mg/L COD, 24.3mg/L FOG and 15.3mg/L TSS, rendering it safe for discharge into the CCT WWTWs, the permeate did not meet the DWS standards for effluent discharge. Therefore, on-site re-use of the treated water is not possible.

6.2. Recommendations

- It would be beneficial for the lab-scale treatment plant to be operated at different EGSB and submerged membrane flowrates and HRTs to assess the plant's performance and determine the optimum operating conditions.
- It is suggested that the analysis of the sample include the REs of the nutrients such as nutrients such as ammonia (NH_3) and phosphate (PO_4^{3-}), especially in the anoxic and submerged membrane tanks.
- Since the effluent did not meet the DWS standards for effluent discharge, it is recommended that a further treatment stage and UV radiation be implemented to deactivate pathogens without adding additional chemicals to the system that might result in the formation of harmful by-products.
- It is also recommended that a comparative analysis be conducted to assess other modified membrane materials in order to determine whether further RE improvements can be attained.
- It would be beneficial for the oxidation mechanisms of pollutants, particularly for the treatment stage be studied.
- Performing a cost analysis of the multi-stage treatment system proposed in this study is highly recommended.
- The performance of a two-stage anaerobic-aerobic digestion system is evaluated.
- Using data from an existing working digester to assess whether the empirical kinetic and stoichiometric values actually represent the digester's performance as a means of verifying the model developed.

- Finally, performing a dynamic simulation to assess how the model responds to varying SRTs and HRTs.

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Appendices

APPENDIX A: Analytical methods

A.1: Method to determine the temperature, pH, EC and DO using the Lovibond SensoDirect 150 Water Testing Multimeter.

pH measurement (with manual temperature setting)

1. Attach the pH Electrode by installing the “Probe Plug” into the “pH Socket/BNC Socket”.
2. Power on the meter by pressing the “Power” button once.
3. Keep pressing the “Mode” button until the display of the bottom right shows “pH” and “Manual Temp.” indicator.
4. Adjust the Manual Temp. value exactly to the same temperature as the solution.
5. Remove cap and hold the pH Electrode body and completely immerse the “Sensing Head” in the solution to be measured and gently swirl the probe.
6. The upper display will show the pH value, the bottom left display will show the Manual Temp. setting.

pH calibration procedure

1. Attach the pH Electrode by installing the “Probe Plug” into the “pH Socket/BNC Socket”.
2. Power on the meter, set the mode to pH measurement, and the bottom right display will show “pH”.
3. Adjust the “Temperature Compensation Value” to make it the same temperature as the pH buffer solution.
4. Hold the “pH Electrode body” and completely immerse the “Sensing Head” in the buffer solution and gently swirl the probe. The display will show the pH value.
5. Press the “REC” button and “HOLD” button at the same time.
6.
 - If the buffer solution is pH 7.0 (± 1 pH), the upper display will show 7.00 automatically.
 - If the buffer solution is pH 4.0 (± 1 pH), the upper display will show 4.00 automatically.
 - If the buffer solution is pH 10.0 (± 1 pH), the upper display will show 10.00 automatically.
 - If the buffer solution value is beyond pH 7.00, pH 4.00, pH 10.00 (for example 7.01, 4.02, 10.03) then use “▲” button, “▼” button to adjust the display value to exactly match the pH buffer solution value.
7. Press the “Enter” button twice to save the calibration data and finish the calibration procedure.
8. The described procedure can be performed for the following calibration points:

pH7 calibration

pH4 calibration

pH10 calibration

- Calibration should always start with pH7, followed by pH 4 and / or pH10 calibration.
- Rinse the electrode with distilled water before each calibration point.
- Repeat the above calibration procedure at least twice to ensure accuracy.

Electrical Conductivity (EC) measurement

1. Attach the Conductivity Probe by installing the “Probe Plug” into the “CD Socket”.
2. Power on the meter by pressing “Power” button once.
3. Keep pressing the “Mode” button until the bottom right display shows a value (e.g. “200 mS”) and “Auto Range”.
4. Remove probe cap and hold the probe body and completely immerse the “Sensing Head” in the solution to be measured. Swirl the probe to let any air bubble escape from the sensing head.
5. The display will show the conductivity values in either “mS/cm” or “ μ S/cm”. At the same time the bottom left display will show the Temp. value of the measured solution.

EC calibration procedure

1. Obtain the standard conductivity solution:
For example:
 - 2 mS range calibration solution:
1.413 mS Conductivity Standard Solution
 - 200 μ S range calibration solution:
80 μ S Conductivity Standard Solution
 - 20 mS range calibration solution:
12.88 mS Conductivity Standard Solution
Or other Conductivity Standard Solution.
2. Install the “Probe Plug” into the “CD Socket”.
3. Power on the meter, and set the mode to conductivity measurement (μ S, mS).
4. Hold the probe body and completely immerse “Sensing Head” in the standard solution. Swirl the probe to let any air bubble escape from the sensing head. The display will show the conductivity (mS) value.
5. Press the “REC” button and “HOLD” button at the same time. The display will show the following screen, as an example. Now release.

6. Use “▲” button, “▼” button to adjust the upper display value to match the standard conductivity value.
7. Press the “Enter” button twice to save the calibration data, and finish the calibration procedure.
 - If only one calibration point is needed, just set the 2 mS range (1.413 mS Cal.).
 - A multi-point calibration procedure should always start with 2 mS range (1.413 mS Cal.), then proceed to other ranges (20 μ S range, 20 mS range or 200 mS range) if necessary.

ATTENTION: Make sure the Oxygen probe is filled with Electrolyte!

Dissolved Oxygen (DO) measurement

1. Attach the Oxygen Probe by installing the ‘Probe Plug’ into the “DO Socket”.
2. Power on the meter by pressing the “Power” button once.
3. Keep pressing the “Mode” button until the bottom right display shows “%O₂”.

CAUTION! Ensure calibration on air before measurement. Wait approx. 2 minute until the reading value stabilises. If the reading value on air is not within 20.7 to 21.1 (20.9 \pm 0.2), then proceed with calibration procedures first.

After completing the calibration procedures, the display should show a value between 20.8 and 21.0 (20.9 \pm 0.1).

4. Press the “Function” button once, and the bottom right display will show “mg/L”. Now the meter is ready for the Dissolved Oxygen measurement.
5.
 - Remove the protective cover from the probe head and immerse the probe to a depth of at least 10 cm in the measured liquid in order for the automatic temperature compensation to take effect.
 - Thermal equilibrium must occur between the probe & the measurement sample, which usually takes a few minutes if the Temp. difference between the two is only a few degrees Celsius.
 - To measure the dissolved oxygen content in any given liquid, it is sufficient to immerse the tip of the probe in the solution, making sure that the velocity of the liquid coming into contact with the probe is at least 0.2 – 0.3 m/s. This is achieved by swirling the probe in the solution.

- During laboratory measurements, the use of a magnetic stirrer/ agitator is recommended. In this way, errors due to air diffusion in the solution are reduced to a minimum.
6. The display will show the Dissolved Oxygen values (mg/L). At the same time, the bottom left display will show the Temp. value of the measured solution.
 7. Rinse the probe carefully with normal tap water after each series of measurements.

DO calibration procedure

1. Install the “Probe Plug” into the “DO Socket”.
2. Power on the meter by pressing the “Power” button once.
3. Keep pressing the “Mode” button until the bottom right display shows “%O₂”.
Wait for at least 5 minutes until the display reading values stabilise with no fluctuation.
4. Press the “Enter” button twice. This will save the calibration data and finish the calibration procedure. Finally, the lower display will show “O₂ CAL. OK”. Return to the normal screen. The complete calibration procedure will take approximately 30 seconds.

Calibration – additional information:

As oxygen in the air is typically 20.9%, use ambient air O₂ for quick & precise calibration.

Table A.1: Summary of the analysis methods done on the representative samples that were sent to the independent accredited laboratory (EPA, 1999; Integral Laboratories, 2019; ZDHC Wastewater Guidelines, 2019).

Parameters	Units	Method	ISO	European Standards	United States
COD	mg/L	M2	ISO 6060**	ISO 6060**	US EPA 410.4, APHA 5220 D**
FOG	mg/L	5520B	ISO 9377*2	EN ISO 9377-2	US EPA 10056
TSS	mg/L	M8	ISO 11923	ISO 11923	US EPA 160.2

APPENDIX B: Auxiliary operation parameters used

B.1: Determination of the hydraulic retention time

The hydraulic retention time, which is the average amount of time that liquid and soluble compounds stay in a reactor or tank, is given by Equation B.1:

$$HRT = \frac{V}{Q} \quad (B.1)$$

where HRT – hydraulic retention time, hr

V – working volume of the bioreactor, m³

Q – influent feed flowrate, m³/hr

B.2: Determination of the organic loading rate

The organic loading rate is given by Equation B.2:

$$OLR = \frac{COD}{HRT} \quad (B.2)$$

where OLR – organic loading rate, mgCOD/L.hr

COD – influent COD, mg/L

HRT – hydraulic retention time, hr

B.3: Determination of the recycle stream up-flow velocity

The recycle stream up-flow velocity is determined by Equation B.3:

$$V_{up} = \frac{H}{HRT} \quad (B.3)$$

where V_{up} – up-flow velocity, m/hr

H – bioreactor height, m

HRT – hydraulic retention time, hr

B.4: Determination of the removal efficiency of organic matter

In this study, the treatment performance was evaluated through the deduction of the percentage removal of COD, FOG and TSS. The formula used to determine the removal

efficiency is given by Equation B.4:

$$RE = \frac{\textit{Influent}-\textit{Effluent}}{\textit{Influent}} \times 100 \quad (\text{B.4})$$

where RE – removal efficiency, %

APPENDIX C: Tables used to plot graphs

Table C.1: Pre-treatment performance

Days	Influent (mg/L)			Effluent (mg/L)			Removal efficiency (%)		
	COD	FOG	TSS	COD	FOG	TSS	COD RE	FOG RE	TSS RE
7	4092	123	1230	1667	22	520	59	82	58
14	4310	240	340	1450	104	176	66	57	48
21	3210	35	365	881	8	184	73	77	50
28	3965	43	198	1374	11	100	65	74	49
35	5900	65	520	3452	25	348	41	62	33
42	4298	72	365	1988	31	292	54	57	20
49	4650	320	1302	3525	135	940	24	58	28
56	5120	730	1870	4092	370	1173	20	49	37
63	4092	340	650	3096	141	460	24	59	29
70	13250	2870	3100	4092	1478	2650	69	49	15
77	4092	310	645	2324	79	450	43	75	30
84	9850	1478	2650	2385	55	480	76	96	82
91	6700	450	2650	2324	89	450	65	80	83
98	4750	640	1160	2324	120	450	51	81	61
105	3484	410	1160	1868	196	730	46	52	37
112	4200	310	1160	1785	104	198	58	66	83
119	3800	240	1160	1860	120	192	51	50	83
Average	5280	510	1207	2382	182	576	52	66	49

Table C.2: EGSB performance

Days	Influent (mg/L)				Effluent (mg/L)			Removal efficiency (%)			OLR (mgCOD/L.h)
	COD	FOG	TSS	OLR	COD	FOG	TSS	COD RE	FOG RE	TSS RE	
7	1667	22	520	291.74	1223	16	476	26.6	27.3	8.5	291.7
14	1450	104	176	253.76	809	26	80	44.2	75.0	54.5	253.8
21	881	8	184	154.18	395	4	14	55.2	50.0	92.4	154.2
28	1374	11	100	240.46	961	5	13	30.1	54.5	87.0	240.5
35	3452	25	348	604.13	1174	5	150	66.0	80.0	56.9	604.1
42	1988	31	292	347.92	1176	17	126	40.8	45.2	56.8	347.9
49	3525	135	940	616.91	1556	5	78	55.9	96.3	91.7	616.9
56	4092	370	1173	716.14	2602	40	450	36.4	89.2	61.6	716.1
63	3096	141	460	541.83	1450	45	132	53.2	68.1	71.3	541.8
70	4092	1478	2650	716.14	1809	46	308	55.8	96.9	88.4	716.1
77	2324	79	450	406.72	1200	42	210	48.4	46.8	53.3	406.7
84	2385	55	480	417.4	874	45	120	63.4	18.2	75.0	417.4
91	2324	89	450	406.72	643	26	144	72.3	70.8	68.0	406.7
98	2324	120	450	406.72	757	24	74	67.4	80.0	83.6	406.7
105	1868	196	730	326.92	657	24	54	64.8	87.8	92.6	326.9
112	1785	104	198	312.39	762	31	95	57.3	70.2	52.0	312.4
119	1860	120	192	325.52	394	22	58	78.8	81.7	70	325.5
Average	2382	182	576	417	1085	25	152	53.9	66.9	68.4	416.8

Table C.3: Submerged membrane performance

Days	Influent (mg/L)			Effluent (mg/L)			Removal efficiency (%)			OLR (mgCOD/L.h)
	COD	FOG	TSS	COD	FOG	TSS	COD RE	FOG RE	TSS RE	
7	1223	16	476	132	4	13	89	75	97	8.8
14	809	26	80	132	5	10	84	81	88	5.8
21	395	4	14	100	3	5	75	25	64	2.8
28	961	5	13	129	2	5	87	60	62	6.9
35	1174	5	150	134	2	11	89	60	93	8.4
42	1176	17	126	105	6	9	91	65	93	8.4
49	1556	5	78	98	3	6	94	40	92	11.2
56	2602	40	450	98	9	5	96	78	99	18.7
63	1450	45	132	86	15	5	94	67	96	10.4
70	1809	46	308	70	12	5	96	74	98	13.0
77	1200	42	210	68	12	5	94	71	98	8.6
84	874	45	120	105	12	9	88	73	93	6.3
91	643	26	144	91	9	11	86	65	92	4.6
98	757	24	74	76	11	6	90	54	92	5.4
105	657	24	54	98	8	5	85	67	91	4.7
112	762	31	95	98	9	5	87	71	95	5.5
119	394	22	58	82	7	9	79	68	84	2.8
Average	1085	25	152	100	8	7	88	64	90	8

Table C.4: Overall performance of the integrated multi-stage treatment

Days	Influent (mg/L)			Effluent (mg/L)			Removal efficiency (%)		
	COD	FOG	TSS	COD	FOG	TSS	COD RE	FOG RE	TSS RE
7	4092	123	1230	132	4	13	97	97	99
14	4310	240	340	132	5	10	97	98	97
21	3210	35	365	100	3	5	97	91	99
28	3965	43	198	129	2	5	97	95	97
35	5900	65	520	134	2	11	98	97	98
42	4298	72	365	105	6	9	98	92	98
49	4650	320	1302	98	3	6	98	99	100
56	5120	730	1870	98	9	5	98	99	100
63	4092	340	650	86	15	5	98	96	99
70	13250	2870	3100	70	12	5	99	100	100
77	4092	310	645	68	12	5	98	96	99
84	9850	1478	2650	105	12	9	99	99	100
91	6700	450	2650	91	9	11	99	98	100
98	4750	640	1160	76	11	6	98	98	99
105	3484	410	1160	98	8	5	97	98	100
112	4200	310	1160	98	9	5	98	97	100
119	3800	240	1160	82	7	9	98	97	99
Average	5280	510	1207	100	7.6	7.3	97.8	96.9	99

APPENDIX D: Supplementary figures



Figure S1: The raw PSW was collected in 25L polypropylene drums from a poultry slaughterhouse in the WC Province, SA.



Figure S2: The pre-treatment process in which 20ml Ecoflush™ was added to 20L PSW, aerated for 24h, then allowed to settle for a further 24h before screening and placing it into the 25L feeding tank.



Figure S3: Photographic illustration of the glass marbles underdrain system and the EGSB reactor.



Figure S4: Sludge washout in the EGSB recirculation stream (left), and biogas gas line (right).



Figure S5: Photographic illustration of the submerged ultrafiltration membrane (left), and membrane tank (right).