## PRODUCTION OF VALUE-ADDED PRODUCTS FROM WASTE DERIVED VOLATILE FATTY ACIDS: A REVIEW

by

Matthew Donald Hinnecke

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in the program of

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Production of Value-Added Products From Waste Derived Volatile Fatty Acids: A Review Matthew Donald Hinnecke Master of Engineering, 2019 Ryerson University

#### ABSTRACT

Due to the exponential growth of the human population and declining environmental quality in the world, waste derived volatile fatty acids (VFAs) have been identified as a source for the production of value-added products. Throughout this paper, different technologies for the production of value-added products from VFAs, various high content VFA waste streams and value-added products from each process will be discussed. Additionally, an in-depth literature review will be conducted on 5 value added products from VFAs. Highlights of various experiments will be identified as well as common trends in experiments to date. Some considerations will also be given to particular strategies and methods which may enhance the production of a value-added product in the future. Even through the uncertainty it has been proven that waste derived VFAs are a major candidate in contributing to a more environmentally and sustainable society in the immediate future.

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### 1.0 Introduction

As earth's population continues to grow, so does the need for energy. Humans have become more and more reliant on energy as lifestyles are changing. Technology is advancing and quite simply everything that is in use today requires a vast amount of energy. As non-renewable resources are continually being depleted to meet the demands of the current populous, energy sustainability will have to rely on the use of different sources of renewable energy. As the human population has expanded, so has the waste that is being generated every day. For the longest time, the "waste" generated by humans has gone to landfills without the consideration of converting more of this "waste" into a renewable source of energy through newly developing technologies. The generation of electricity, heat and biofuels from renewable energy sources such as human waste has become a high priority in energy strategies at a global scale and will continue to be at the forefront for many years to come (Resch, et al., 2008).

Creating new value-added products from the waste, can not only achieve a decrease in the overall waste, but there is a benefit that has been achieved from this waste; creating a positive effect and further expanding the opportunity for more renewable energy generation. These new value-added products not only provide a benefit to the amount of energy being generated, but they also provide a benefit to the environment. If even the slightest improvement is done to prevent greenhouse gas emissions from incinerators, or soil and water contamination from landfills or dumping in streams there will not only be a better quality of life in the respective area but a better environment globally. Environmental degradation is a major concern amongst many of the worlds most populated countries and anything that can be done to minimize the rate or

even negate the environmental degradation that is currently occurring is something that must be considered.

One way to reduce the environmental degradation rate is through the production of volatile fatty acids (VFA's). The production of VFAs from waste derived sources and their contribution to value added products is a major part of contributing to an environmentally sustainable society

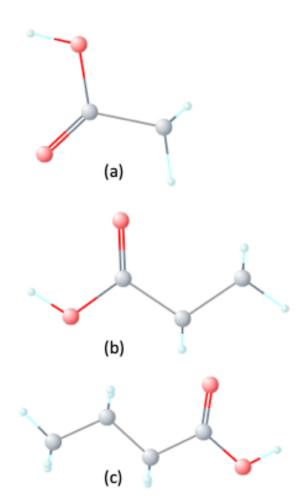


Figure 1: Molecular Structures of (a) Acetic Acid (b) Propionic Acid (c) Butyric Acid

for the forthcoming future. VFA's are short chain fatty acids. In order to be classified as a VFA, the chain of fatty acids must have less than six carbon atoms when distilled (Lee, et al., 2014). The most prominent examples of VFA's would be general acids such as acetic acid, butyric acid and propionic acid as shown in figure 1. The VFA's that are extracted from the wastes are converted through various methods and can provide value added products to a wide range of applications such as plastics, energy, fuels and nutrient removal processes. Until recently, the production of many of the above noted products has been done with fossil fuels.

Plastics, energy and fuels all come from a combination of coal and natural gas or oil. With these resources becoming more and more limited, there is a steep incline in the price to acquire them.

By using VFA's as a source for production of equivalent resources, costs are lowered for the overall process and resources are not depleted at an exponential rate.

Studies indicate that the world is at its highest rate of pollution ever and the World Health Organization estimates that 90% of the world's population breathes polluted air (World Health Organization, 2018). It is also estimated that polluted air alone causes approximately seven million deaths worldwide. With statistics like this, there is a need for a more environmentally sustainable course of action, in order to allow humans to maintain their same status of living while implementing a system that has at least a carbon neutral effect on the environment.

The World Health Organization also notes that there are more countries taking action to reduce the effects of climate change and pollution. One of the ways in which this is being done is through the use of various technologies for the development of VFA's. In an ideal world, max production would be seen on the first try, but it has taken years for different technologies to come to the forefront. Through tedious trials and experiments, technologies for VFA production have been developed, but a global optimum has never been determined due to the wide nature of the types of wastes being used. What can be seen is that if a particular type of waste is used continuously there is a proper process and control implemented in order to manipulate the waste to provide a maximum benefit. That being said, even though a maximum has temporarily been determined, testing still occurs to determine if more can be obtained from a simple waste.

Within this article, various different types of technologies will be discussed. As the production of VFAs is dependent on the waste type that is used, different waste types will be described with the common sources for that waste production. A brief description of the value-added products and their common uses will also be described in this paper. Data has also been

compiled from literature with regards to various types of wastes and operating conditions in order to offer an insight into the optimal waste and operating conditions. Finally, conclusions and remarks will be made on different types of technologies.

### 2.0 Technologies for VFA Production

#### 2.1 Biological Process (Anaerobic Digestion)

VFA production is borne through the use of a few specific technologies. In the industry today, there are two particular technology types that are used for the production of energy and bi-products from VFA's. The most common technology used for the production of VFA's is the anaerobic digestion technology, which is otherwise known as the biological technology for VFA production. The main goals of anaerobic digestion include the destruction of organic material, making it more stable to release into the environment and at the same time producing biogas and fostering pathogen destruction. Within the anaerobic digestion technology, there are three critical steps which are undergone by the waste. Beginning with hydrolysis and fermentation, the waste is converted from carbohydrates and proteins to simple sugars and amino acids. Those sugars and amino acids are then converted into fatty acids and alcohols. The fatty acids and alcohols then undergo acetogenesis and dehydrogenation in which organic acids and alcohols are broken down, resulting in acetic acid and hydrogen. Lastly, is the production step in which the desired bacteria react with the acids and hydrogen to yield the desired product.

The most common types of anaerobic digesters in use today include, but are not limited to, the upflow anaerobic sludge blanket (UASB), packed bed reactor, fluidized bed reactor and the continuously stirred tank reactor (CSTR) (Najafpour, et al., 2016). These types of anaerobic digesters are also further classified into attached and suspended growth technologies, which each have their own pros and cons (Najafpour, et al., 2016). These will be discussed in the upcoming sections.

#### 2.1.1 Continuously Stirred Tank Reactors (CSTR's)

Continuously stirred tank reactors (CSTRs) are the most prominent method for the treatment of wastewater, producing value added products from VFA's. CSTRs are used in a way which directly combines the biomass with the waste, causing a series of reactions within the operational chamber as shown in figure 2. The organic loading rate (OLR) of the reactor must be appropriately designed based on the expected output. Without taking this into consideration,

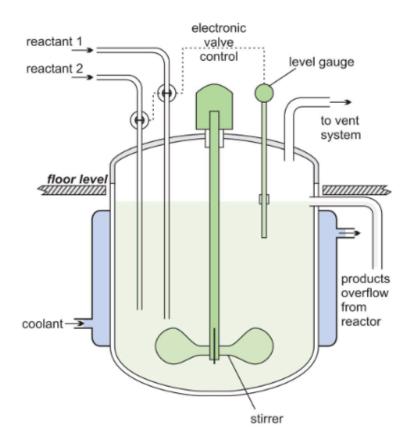


Figure 2: Typical CSTR Reactor Design, (Center for Industry Education and Collaboration, 2013)

there will be a large loss of productivity from the reactor. CSTR's are one of the most basic methods for the production of value-added products from waste and one of the least expensive

as well. They are typically used in the production of methane or ethanol from various wastes. As the waste types undergo the anaerobic digestion process, as noted previously, the resulting value-added product is extracted from the reactor as harnessed for its end result. In a CSTR, the feedstock or waste, is pumped into the reactor where it sits for the determined period and is continually mixed with the biomass. Over time, the anaerobic reactions take place resulting in the value-added product. Once created, these products may be used within the facility or exported to other facilities for their final use. The effluent is then removed from the reactor with a chemical oxygen demand much less than what it entered with. When using a continuous CSTR, there is little down time in comparison to a batch reactor and can offer a greater efficiency in the number of value-added products created. CSTR's are more commonly used in series than any other anaerobic digestion apparatus. By putting the reactors in series, there is an elimination of lag time for the bacteria to begin feasting on the waste. This of course exposes the reactors to a higher risk of contamination (Alexandre, et al., 2008).

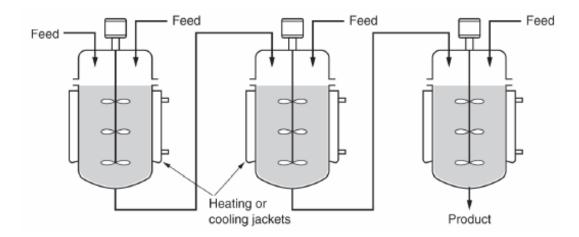
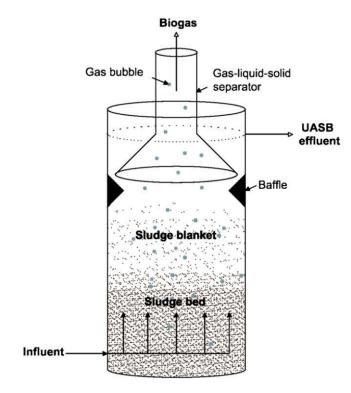


Figure 4: CSTR Reactors in Series (Forger, et al., 2007)

#### 2.1.2 Upflow Anaerobic Sludge Blanket (UASB)

The UASB reactor operates quite simply. It is originally filled with a material such as digested, anaerobic, granular, flocculent or activated sludge (Chong, et al., 2012). From here the reactor has sludge pumped into the bottom. The UASB reactor is a suspended growth technology as there



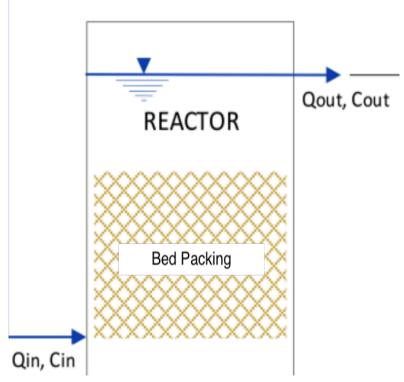
is no material to attach to. In the ideal scenario, the lighter particles will move up the reactor while leaving the heavier particles at the bottom of the reactor. As the lighter particles continue to rise, they travel through the sludge blanket and the soluble organic compounds in the mixture are converted to the desired biogas. The gas then continues upwards carrying the insoluble particles which are removed in the UASB effluent as shown in figure 4. The

gas-liquid-solid separator is essential to the process as it will separate the respective phases and allocate them to the desired area. The reactor itself may be combined with additional technologies in order to ensure that volatile solids are retained ensuring maximum productivity is obtained from the waste. With the ability to ensure that volatile solids are retained, this makes the UASB a suitable reactor for the pretreatment of various wastes, creating maximum efficiency in the next step of the process.

Figure 4: Typical UASB Reactor Design, (Chong, et al., 2012)

#### 2.1.3 Packed Bed Reactor

In a packed bed reactor, the reactor setup is similar to that of the UASB reactor but differs with regards to the material being present within the reactor. The packed bed reactor is an



attached growth system as the biomass grows and attaches to the packing material currently in place. As shown in figure 5, the influent is once again through the bottom of the reactor. The bed may then be packed with a sort of medium. The medium for the packed bed reactor may be a variety of different sources such as

plastics,

rocks

and

ceramics



(Detalina, et al., 2018). In the packed bed reactor, the waste is imputed through the bottom of the reactor. At this point the flow is forced upwards through the packed bed. As the waste or sludge passes through this area, it reacts with the bacteria and biomass which has attached to the packing creating the desired biogas. The biogas is then extracted from the reactor with the remainder of the effluent being removed from the reactor. One disadvantage that is commonly seen with the packed bed reactor is that the system will often clog. This was noticed in the original stages leading to the development of the fluidized bed reactor, which will be discussed in the next section.

#### 2.1.4 Fluidized Bed Reactor

The fluidized bed reactor, similar to the packed bed reactor is an attached growth system. In a fluidized bed reactor, the bacteria attach to the fluidized media within the reactor. This creates a biofilm around the particles and ultimately increases the surface area of the bacterial growth as shown in figure 6. With an increase in the contact between the biofilm and the

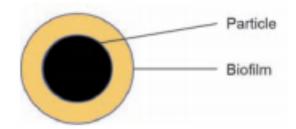
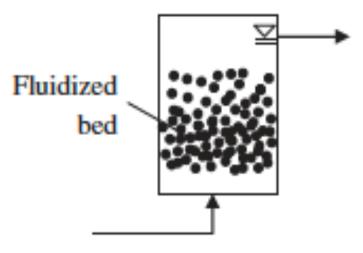


Figure 6: Biofilm Casing Around Fluidized Media Particle, (Nelson, et al., 2017)



wastewater within the reactor, the system is able to break down larger compounds that are typically more difficult to treat in a regular system (Nelson, et al., 2017). Similar to the previously mentioned processes, the fluidized

> bed reactor operates by having the waste material pumped in through the bottom of the reactor. This creates an upward flowing effect for the material as the outlet for the effluent is typically located at the top of the reactor as shown in figure 7. The waste water passes through the fluidized media and

Figure 7: Typical Fluidized Bed Reactor Configuration ( (Lee, et al., 2014)

reacts with the biofilm that has formed on the surface. The media is able to stay suspended due to two possible operational conditions. The first condition is known as conventional fluidization, in which the liquid velocity is sufficient enough to keep the media suspended, but not high enough to cause it to be removed with the effluent (Nelson, et al., 2017). The second condition is known as circulating fluidization in which a high velocity is used to bring the media to the top of the column and then a recycle line will return them to the bottom of the reactor (Nelson, et al., 2017). Once the wastewater has been broken down by the particles, it is removed in the form of effluent.

#### 2.2 Chemical Processes

Second to the biological process, is the chemical processes of oxidation. The oxidation process is primarily used for the production of bioelectricity and the main apparatus that is used is the microbial fuel cell (MFC). The microbial fuel cell works similar to a circuit in which there is an anode and a cathode. The bacteria placed in the anode attach to the electrode as they must transfer the electrons which they inherent from the consumption of waste. This charge is then transferred to the anode, which is exposed to oxygen, creating water in the opposite chamber. A MFC can only be created if the fuel supply for the bacteria is continually renewed as the chambers in the MFC are separated, creating an overall fuel cell, thus completing the oxidation process.

Microbial fuel cells (MFCs) can be used to treat a variety of different waste types. They not only produce a high amount of electricity, but also remove the harmful elements in wastes. Wastewater is one of the most common wastes used in microbial fuel cells as the components are often easily biodegradable. The MFC is a bioelectrochemical system that will allow the production of energy through chemical processes. Energy is produced through the oxidation of organic matter. The organic matter is oxidized by bacteria and microorganisms under relatively ideal conditions, if primary waste is being used as the waste source (Samsudeen, et al., 2016). The oxidization process breaks down the molecules allowing the elements to react with the

bacteria attached to the anode. The anode bacteria sends electricity through the fuel cell to the cathode, where the electrons are discharged creating water. Often times the MFC is used to

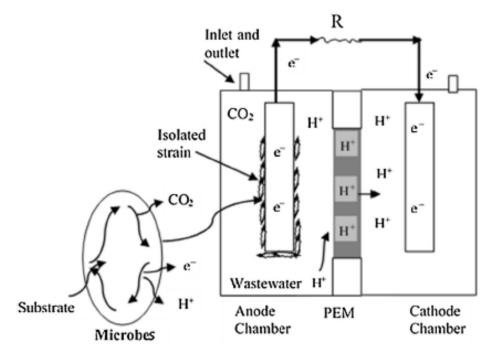


Figure 8: Typical Microbial Fuel Cell Apparatus (Samsudeen, et al., 2016)

compliment an anaerobic digestor in order to maximize the production from a specific waste type. MFC's are able to be used in tandem with anaerobic digestors due to the fact that they can operate under less favorable conditions including a lower temperature and a lower pH (Li, et al., 2013). As most wastes do leave the anaerobic digestor at a lower pH and temperature, MFC's are seen as a viable secondary source of production. When not used in tandem with an anaerobic digester, MFC's are able to generate high voltages and currents through the degradation process which may prove that they are a viable source for electricity production on their own (Li, et al., 2013). MFC's do not offer any value-added products other than electricity. This may make them unfavourable when dealing with a waste such as corn that will produce grains as well as ethanol or methane.

### 3.0 Types of Wastes

The type of waste which is used dictates the amount of VFA's that are produced and ultimately the number of value-added products which can be obtained from it. Common wastes include waste water from municipal wastewater plants, food waste and liquid wastes. Waste types from municipal wastewater plants are the most prominent, as they are so readily available due to the ever-expanding human population. Food waste and liquid wastes are less common than municipal wastewater but are still ever so present in industries. Although it has not been determined which type of waste is the most appropriate to use, to obtain value added products from VFA's, each type of waste has their own particular advantages and disadvantages that will be discussed in the upcoming sections. Additional considerations must be taken into account when dealing with different types of wastes. There is a high variability in the number of organics within a sample, as well as the amount of soluble and insoluble particular matter within the waste, leading to uncertainty in each waste type.

#### 3.1.1 Municipal Waste Water

Municipal waste water (MWW) is prominent all throughout the world and can actually be seen as a resource if the right technologies are in place to process the waste. MWW is commonly used in anaerobic digestion and may exist in a variety of forms. MWW may exist as primary sludge, waste activated sludge of combination of the two known as mixed sludge. Primary sludge is generated during the physical treatment in primary settlers. This may include wastes from kitchen, toilets, vegetables, fruits and more (Ji, et al., 2010). On the other hand, waste activated sludge is generated in the biological treatment of a wastewater treatment plant and may consist of non-hydrolysable particulate materials that have been broken down due to biological metabolism (Pinto, et al., 2016). Mixed sludge is a combination of the primary sludge and waste activated sludge. Often, mixed sludge is a prominent source for VFA production, as it is rich in organic matter from the primary and waste activated sludge. The downside to this is that the mixed sludge contains much more insoluble chemical oxygen demand (COD) than soluble COD, leading to longer retention times as hydrolysis cannot be achieved as quickly as desired (Pinto, et al., 2016). Pretreatment is a possible solution to this but will drive up the cost to run the anaerobic digester.

#### 3.2.2 Food Waste

Although food waste may be a large component of municipal waste water, food waste can be separated at the source from the waste water and considered to be its own waste type. Although this may be a more difficult task to undergo, the production rates for biogas will rapidly increase as the TS and VS content increases. Food waste may have TS and VS contents of 18.1-30.9% and 17.1-26.35% (Zhang, et al., 2014) respectively which will influence the biogas production rate significantly. As food waste typically has a moisture content around 70%, it is seen as a viable source for anaerobic digestion and the production of clean biogas to be used for energy or other renewable measures. Ideally, the purer the food waste is and the less contaminants there are in the waste, the more efficiently the biogas can be produced.

#### 3.2.3 Liquid Waste

Liquid waste may also be a contributor to municipal waste water in some areas, but often times may be already separated from the municipal plants. Examples of liquid waste often include wastes from paper, dairy and agricultural industries. Agricultural wastewaters have been previously experimented with and have been determined to be a viable source for the production of value-added products (Abgenent, et al., 2004). Agriculture wastewater, similar to generic wastewater, is also high in easily degradable organic matter which makes it a prime candidate

for bioprocessing. Dairy wastewater, as a result of substances such as cheese whey, similar to agricultural wastewater, also has a high organic content and has been tested as a viable source for the production of biogas (Escalante, et al., 2018). Pulp and paper mill products have the lowest organic content of readily available liquid wastes. This is due to the fact that there are chemicals used in pulp and paper production process that are expelled in the effluent, as well as a high amount of insoluble organic compounds in the effluent. Although this is a setback, since it is produced in such large quantities, the pre-treatment of pulp and paper mill effluent is often seriously considered in order to develop a sustainable product from waste (Kamali, et al., 2016).

## 4.0 Value Added Products From VFA's

#### 4.1 Bio-Methane

Methane is a valuable source for energy production as it is an easily combustible compound and when burned is broken down into carbon dioxide and water. Methane is most typically produced in an anaerobic digestor undergoing a chemical breakdown. As mentioned previously in section 3, the waste is added to the digester where it undergoes the processes of hydrolysis and fermentation, followed by acetogenesis and dehydrogenation. At this point, the waste is broken down into an acid and hydrogen. In order to produce methane from waste, methanogenesis must be undergone. This is done through the use of methanogenic bacteria which will degrade the waste and expel methane. As these are a specialized bacteria they will only produce methane when degrading the waste. Methanogens are obligate methane producers and do not grow using fermentation or alternative electron receptors for respiration (Lyu, et al., 2018). Through anaerobic digestion, the typical composition of biogas will have 50-75% methane (Kamali, et al., 2016). After the methane is refined, it can then be burned creating electricity, steam and heat. The typical yield from 1m<sup>3</sup> of methane is approximately 0.037GJ (Hafez, 2019), but may increase depending on the combination of energy recovery systems used. It is also more beneficial to release carbon dioxide into the environment instead of methane, so burning the methane and obtaining energy lowers the environmental impact substantially.

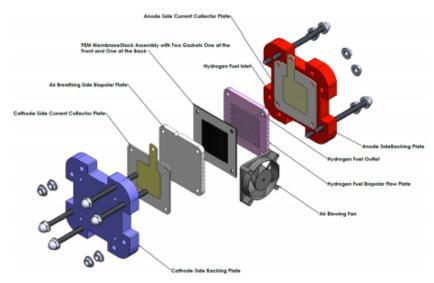
#### 4.2 Bio-Hydrogen

Hydrogen production is very similar to that of the methane process. Biohydrogen can be produced through two process: biophotosis and fermentation. Biophotosis is a process in which hydrogen is produced through photo-synthetically splitting water (Chang, et al., 2008). Fermentation is much less expensive and is the preferred method for biohydrogen production due to its simplistic nature and will be the main focal point for this paper. As the biomethane process extracts the methane from the fermentation process, the biohydrogen process extracts the hydrogen from the fermentative process. Hydrogen is seen as a viable source for renewable energy, as it generates energy at a level three times higher than that of methane (Evvyernie, et al., 2001). In recent years, the option to include a hydrogen fuel cell within vehicles has been implemented, thus lowering the amount of fuel required to operate the vehicle or completely removing it, contributing to capping the global warming increase at 2°C (Eriksson, et al., 2017). Hydrogen fuel cells in vehicles are much more efficient than internal combustion engines as they use 75% of the total fuel energy to run the vehicle in comparison to the 15% from internal combustion engines (Wilberforce, et al., 2017). Hydrogen fuel cells are not solely limited to vehicles, but may also be used for other electrical generation systems and are also looked at as

an option for fuel storage as hydrogen is one of the most abundant elements on earth (Eriksson, et al., 2017).

#### 4.3 Bio-Electricity

Bioelectricity is one of the most self-explanatory value-added products from VFA's. Electricity is generated from the production within the typical microbial fuel cell and can be stored or uploaded right to the energy grid. By producing electricity this way, there are minimal

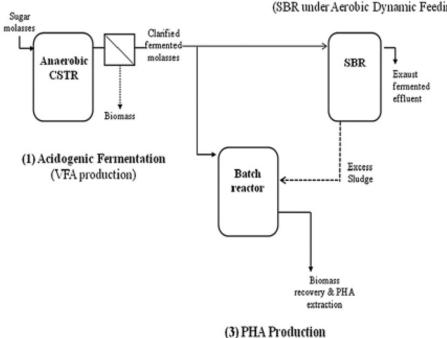


*Figure 9: Typical Polymer Electrolyte Membrane Fuel Cell, (Wilberforce, et al., 2017)* 

bi-products and the waste has been degraded, making this a very attractive technology from both an environmental and economic standpoint. The yield from the microbial fuel cells vary and also depend on the resistance within the fuel cell and the different anode and cathode electrodes are used (Tharali, et al., 2016). Additionally, there is also the potential for the production of bioelectricity to be a downstream process from another type of value added product production.

#### 4.4 Polyhydroalkanoates (PHA's)

Polyhydroalkanoates (PHA's) are produced in a similar fashion to the hydrogen and methane in an anaerobic digestor. A typical 3 stage process is used for the production of PHAs (Strazzera, et al., 2018). For the production of PHA, acidogenic fermentation is undergone producing an effluent (Stage 1). This effluent is clarified and separated between a sequencing batch reactor (SBR). Within the SBR, a portion of the effluent may be converted into an enriched culture, most commonly through aerobic dynamic feeding (Stage 2). Alternatively, an enriched culture can be derived elsewhere and implemented into the reactor but it often a more expensive



(2) Culture selection (SBR under Aerobic Dynamic Feeding)

Figure 10: Typical 3 Stage PHA Production Process Using SBR, (Albuquerque, et al., 2010)

(accumulation under N limitation)

alternative. In the final step, the effluent produced in stage 1 and the enriched culture are combined in another batch reactor. At this point PHA accumulation begins, where the desired product is obtained from the process (Albuquerque, et al., 2010). The concentration and amount of PHAs produced is dependent on the type and amount of VFA's produced during the acidogenic fermentation phase. As this is the major step for the ultimate production of PHAs, it will dictate the net environmental effect and the bi-products from the production process. PHAs are similar to plastics except for the fact that they are biodegradable. The production of thermoplastic polymers from the PHA process is a major step in reducing the amount of petroleum-based plastics currently being created (Strazzera, et al., 2018). Quite simply, the more PHAs that are used in plastics the better the environmental effect, as petroleum is not only bad for the environment at the time of consumption but there is also a significant environmental effect when extracting them from the earth. Plastics produced with petroleum-based products are also not biodegradable, making them a concern for landfills.

#### 4.5 Biological Nutrient Removal (BNR)

Biological nutrient removal (BNR) is an important part in the reduction of nutrients from an effluent stream, that is to be released back into the environment. The BNR process uses naturally occurring microorganisms under variable environmental conditions to facilitate the removal of nitrogen and phosphorus. Nitrogen is removed by a combination of nitrification and denitrification. Nitrification involves the breakdown on ammonia using ammonia-oxidizing bacteria (AOD) to nitrate. Denitrification continues the process and breaks nitrate down into nitrogen gas through nitrate-oxidizing bacteria (NOB) (Alzate Marin, et al., 2016). Phosphorus is removed by phosphate accumulating organisms (PAO's) in a process called enhanced biological phosphorus removal (EBPR) which first breaks down the phosphorus compounds in the anaerobic phase and then consuming the phosphorus in the aerobic phase. In order to complete the denitrification, process a carbon source is required as most of the chemical oxygen demand is consumed upstream of the final reactor (Alzate Marin, et al., 2016). The lack of carbon is often substituted downstream in the form of a carbon bi-product, waste or VFA produced upstream of the process.

As nitrogen and phosphorus are some of the most abundant elements on earth, there are a variety of products that can be retained from the BNR and EBPR process. The most common value-added product is fertilizers. A large amount of nitrogen and phosphorus is used in the creation of fertilizer as they are elements required for the growth of many plants. Another substantial benefit of this process is not a value-added product per say but it does directly improve the quality of the environment as wastewater is often released directly into nearby streams. During this process, wastewater is purified in a way which prevents a harmful effluent from being released into the environment. By doing this, there is a decrease in eutrophication and algal blooms in the surrounding ecosystems, thus fostering a good habitat for plants and animals.

## 5.0 Production & Efficiency of VFA Production Methods

As there is a continual growth for the need of renewable energy sources, there have been multiple experiments and tests done in order to obtain an optimal operating source for the production of value-added products from wastes. The main focus for value added products lies in the following areas: biomethane, biohydrogen, bioelectricity, biological nutrient removal and production of polyalkenoates. For each of the sections below, data has been compiled from literature and summarized in order to identify highlights from previously done experiments and offer insights about the methods based on the conclusive results.

## 5.1 Bio-Methane

Table 1: Summary of Literature Findings for Bio-Methane Production

Waste Type	Reactor Type & Operating Conditions	Methane Yield	Reference
Municipal Wastewater 40% PS 60% WAS	CSTR, 37°C, 15 Days	265 mLCH <sub>4</sub> /gVSS	(Pinto, et al., 2016)
Municipal Wastewater 60% PS 40% WAS	CSTR, 37°C, 15 Days	422 mLCH <sub>4</sub> /gVSS	(Pinto, et al., 2016)
Municipal Wastewater 80% PS 20% WAS	CSTR, 37°C, 15 Days	462 mLCH <sub>4</sub> /gVSS	(Pinto, et al., 2016)
Municipal Wastewater 100% PS	CSTR, 37°C, 15 Days	496 mLCH <sub>4</sub> /gVSS	(Pinto, et al., 2016)
Acidic Effluent From Sugarcane Juice	UASB, 30°C, 6 Days, pH 7	308 mLCH₄/gVSS	(Reungsang, et al., 2016)
Microalgal Biomass	Batch, 38°C, 65 Days	236 mLCH₄/gVSS	(Hernandez et al., 2014)
Perenial Rye Grass	Batch, 38°C, 35 Days, pH 7.2	263 mLCH₄/gVSS	(McEniry, et al., 2013)
Italian Rye Grass	Batch, 38°C, 35 Days, pH 7.2	254 mLCH₄/gVSS	(McEniry, et al., 2013)
Timothy	Batch, 38°C, 35 Days, pH 7.2	255 mLCH₄/gVSS	(McEniry, et al., 2013)
Cocksfoot	Batch, 38°C, 35 Days, pH 7.2	245 mLCH₄/gVSS	(McEniry, et al., 2013)
Tall Fescue	Batch, 38°C, 35 Days, pH 7.2	248 mLCH₄/gVSS	(McEniry, et al., 2013)
Sheep Dung & Corrugated Board Mix (4:1 Mix)	Batch, 37°C, 43 Days, pH 7	152 mLCH <sub>4</sub> /gVSS	(Li, et al., 2018)
Sheep Dung & Office Paper Mix (2:3 Mix)	Batch, 37°C, 43 Days, pH 7	199 mLCH4/gVSS	(Li, et al., 2018)
Pig Manure	Batch, 37°C, pH 7	410 mLCH₄/gVSS	(Li, et al., 2015)
Dairy Manure	Batch, 37°C, pH 7	270 mLCH <sub>4</sub> /gVSS	(Li, et al., 2015)
Raw Sludge, Food Waste Leachate & Algal Biomass	Batch, 35°C, 42 Days, pH 8	176 mLCH₄/gVSS	(Kim, et al., 2015)
Food Waste Leachate & Piggery Wastewater	Batch, 35°C, 30 Days, pH 7.14	323 mL CH <sub>4</sub> /gVSS	(Han, et al., 2012)

Biomethane is a valuable product obtained from the AD process. There are some optimization factors that can be considered. As shown in the study by Pinto, et al. (2016), there is an optimal mixture which may sometimes be obtained between different waste types. WAS and PS are two of the most available waste types. The amount of methane production can be dictated by the total amount of total volatile suspended solids (TVSS). It was noted that the total value of TVSS in the end stages during the primary sludge trial of 100:0 (PS:WAS) had the highest methane yield (Pinto, et al., 2016). As the WAS was not the medium for bacterial production in this test, the 4L inoculum allowed for sufficient production of methane from the PS only (Pinto, et al., 2016).

Additional considerations in Pinto, et al., (2016) were with regards to the carbon nitrogen ratio (C/N) as the C/N ratio for PS to WAS was 2.5:1. This is also a point of emphasis in Han, et al. (2012), as it was hypothesized that a C/N ratio greater than 5.15 may lead to a lower biogas production rate from mixed waste samples. This may be inconclusive depending on the samples that are used as the C/N ratio used for the mixed raw sludge, food waste leachate and algal biomass waste used in the experiment by Kim, et al., (2015), showed that the highest methane production rates occurred at a C/N ratio of approximately 7, in which there was an equal mixture between all three waste types, contributing to a methane production rate of 176mLCH<sub>4</sub>/gVSS.

There are additional mixtures which have been tested as shown in Li, et al., (2015), and Han, et al., (2012), to which agricultural waste was combined with a generic waste. By testing this it was confirmed that the production of methane can be undergone, resulting in a decrease in both waste streams producing a value-added product from anaerobic digestion. As another alternative to reduce the amount of waste, a secondary digestion of a waste source using a dual

recovery system was experimented with in Reungsang, et al., (2016) in which the waste source was first used for the production of hydrogen, then the effluent was used to produce methane creating two value added products from one waste type. Through the experiment conducted by Reungsang, et al., (2016), it was observed that there was a 76% reduction in the initial COD through both processes. As the technologies continue to develop, there will be a larger increase in the amount of wastes which are used for dual recovery. Similar to the dual recovery system for methane and hydrogen, Hernandez, et al., (2014) experimented with microalgal biomass in order to obtain biodiesel and methane. The methane production from this experiment was higher with the effluent from lipid exhausted biomass, than from the production of biodiesel prior to the production of methane.

As a particular area of interest, the time of year in which biomass crops are harvested plays a particular role in the amount of methane which is produced. As experimented in McEniry, et al., (2013), through 5 different types of biomass, there was an overall decrease in the amount of methane that was produced the later into the growing season that it was harvested. In this experiment, the initial harvest done in May yielded 8-15% (McEniry, et al., 2013) more methane than the harvest done in July. The reduction in the methane yield from the two samples may be attributed to the increase in the fibrous components of the plant and the lack of digestibility of the plant at the time of that harvest.

The production of methane is difficult due to the large difference between the optimal growth conditions for microorganisms related to both the acidogenesis step and methanogenesis step of methane production. This may infer that there is a more optimal solution in which a twophase anaerobic digestor is used for the production of biogas in general. In a two-phase digester,

the first digester would involve the acidogens, which typically function better at a lower pH in order to rapidly create the acidogens (Grady, et al., 2011). The second digester would involve the methanogens, which typically operates at a more neutral pH, as concluded by table 1. The twostage system for the production of methane actually allows for the opportunity for the production of methane and hydrogen simultaneously. In the acidogenesis phase for the first reactor, the hydrogen that is released from the waste produce can be collected and used for energy production. Then, as expected, the methane that is produced in the second reactor from the first reactors effluent can be collected and also used for energy production.

Through a two stage anaerobic digestion process, there is much more biogas produced and it is of a higher quality than typical mixed biogas in a one phase anaerobic digestor (Wang, et al., 2011). There is also an estimated 25% higher energy yield than in a typical 1 stage reactor (Wang, et al., 2011). Alternatively, if the hydrogen that is being produced in the first stage of a two stage reactor is not able to be collected and sold at market, it can be used as a heat source for the second reactor, thus making it more environmentally friendly and cost effective to the owner. The implementation of these types of reactors is something that should be very seriously considered amongst producers as although there may be a higher capital cost for the construction of the reactors, the long-term benefits seem to outweigh the upfront costs imposed on the owners.

## 5.2 Bio-Hydrogen

## Table 2: Summary of Literature Findings for Bio-Hydrogen Production

Waste Type	Reactor Type & Operating Conditions	Hydrogen Yield	Reference
Sugarcane Bagasse	Batch, 37°C, pH 5.5	1.73 mol H <sub>2</sub> /mol sugar	(Pattra, et al., 2008)
Glucose	CSTR, 36°C, 14 Days, pH 5.5	2.1 mol H <sub>2</sub> /mol sugar	(Fang, et al., 2002)
Fennel Waste	Batch, 35°C, 10 Days, pH Not Controlled	58 mL H <sub>2</sub> /gVSS	(Ghimire, et al., 2015)
Buffalo Manure	Batch, 35°C, 10 Days, pH Not Controlled	136 mL H <sub>2</sub> /gVSS	(Ghimire, et al., 2015)
Olive Mill Wastewater	Batch, 35°C, 10 Days, pH Not Controlled	46 mL H <sub>2</sub> /gVSS	(Ghimire, et al., 2015)
Potato & Pumpkin Waste	Batch, 35°C, 10 Days, pH Not Controlled	171 mL H <sub>2</sub> /gVSS	(Ghimire, et al., 2015)
Glycerol	CSTR, 37°C, 14 Hours, pH 5.5	163 mL H <sub>2</sub> /gVSS	(Silva-Illanes, et al., 2017)
Cheese Whey	CSTR, 37°C, 65 Days, pH 5.9	2.8 mol H <sub>2</sub> /mol lactose	(Davila-Vazquez, et al., 2009)
Food Waste	Batch, 35°C, pH5.3	1.63 mol H <sub>2</sub> /mol hexose	(Lee, et al., 2014)
Sucrose	CSTR, 35°C, pH 5.7	0.43 mol H <sub>2</sub> /gVSS	(Chen, et al., 2008)
Sucrose	Batch, 35°C, pH 5.5	1.92 mol H <sub>2</sub> /mol sugar	(Mu, et al., 2006)
Starch	Batch, 37°C, pH Not Controlled	5.34 mmol H <sub>2</sub> /g starch	(Lee, et al., 2008)
Glucose	CSTR, 40°C, 43 Days, pH 6.5	1.63 mol H <sub>2</sub> /mol sugar	(Wu, et al., 2008)
Gelidium Amansii & Mixed Microalgal Biomass	Batch, 37°C, 6 Days, pH 7.0	43 mL H <sub>2</sub> /g dry biomass	(Sivagurunathan, et al., 2018)
Textile Wastewater	Batch, 35°C, 25 Days, pH 5.5	0.97 mol H <sub>2</sub> / mol hexose	(Lay, et al., 2014)

The biohydrogen production potential of various wastes is not a definitive set of standards but may merely vary from waste type to waste type. Typically, the performance indicator for hydrogen yields lies in the amount of VFAs produced. Ideally there is a maximum output desired for the amount of input. The maximum outputs are affected by a variety of different factors. pH is a major dictating factor in the area of methane production. As experimented in Pattra, et al., (2008) & Fang, et al., (2002) and across table 2, the requirements for the production from an AD are less than that of methane. The optimal pH appears to be in the range of 5.5 based on the tabulated results and further conclusions from Ghimire, et al., (2015) and Silva-Illanes, et al., (2017). Although not completely known, the pH should be optimized to the design reactor and should not be allowed to skew from the desired pH as the fermentation process as explained in Lee, et al., (2014) may not produce conclusive results. Working in tandem with pH in order to optimize a reactor is the hydraulic retention time (HRT).

In order to appropriately obtain the optimal amount of hydrogen from a reactor, considerations must be made to the HRT as well. From Silva-Illanes, et al., (2017), HRT played an important part in the production of hydrogen as it was directly related to the hydrogen yield and hydrogen production within the reactor. The optimal HRT for the experiment was tested from 8-14 hours, with the optimal being 12 hours, showing that the minimum nor the maximum do not necessarily define the maximum hydrogen yield. In comparison, the HRT from Davila-Vazquez, et al., (2009) was significantly less at a 6 hour HRT in order to obtain the maximum hydrogen yield. It is shown through these two experiments that the HRT is dependent on the waste type that is used. The HRT plays a very significant role especially in CSTR reactors which may remove a lag

phase from the experiment, meaning that the waste source must be refreshed after the HRT period in order to obtain maximum hydrogen production for the owner.

As experimented in Davila-Vazquez, et al., (2009) the OLR was an important factor during the remaining operational periods in order to achieve steady state production of hydrogen. For this experiment, operational periods six and seven had hydrogen molar yields of 2.8 and 2.0 mol H<sub>2</sub>/mol lactose respectively, while their volumetric hydrogen production rates were 46.6 and 45.4 (mmol H<sub>2</sub>/L/h) (Davila-Vazquez, et al., 2009). As can be seen, there is clearly a higher production rate from operational period six in comparison to operational period seven. We see this higher production rate in operational period six which has a lower OLR than operational period seven. This would suggest that the OLR has been maximized during operational period six and the OLR meets the threshold for the bacteria, leaving excess organic matter in the effluent from the reactor (Davila-Vazquez, et al., 2009). A solution to this could be to increase the HRT, but with that other operational parameters may change.

As the technology for hydrogen production continues to expand, new types of wastes may be utilized in order to obtain a value added product such as hydrogen from a digestion process. From Chen, et al., (2008), hydrogen production was undergone with a waste that was high in sulfate concentration around 3g/L. From this, hydrogen yields were seen to be comparable to previous studies from pure waste or wastewater, showing that there is a promising future for the reuse of wastewater with high chemical concentrations. Additional advancements have also been made with regards to the predictability of specific wastewaters and reactors as shown in Wu, et al., (2008), in which a response surface methodology (RSM) was used to accurately predict the amount hydrogen produced within an experiment. From the model

that was produced, the calculated values were within the calculated error regions of the measured experimental data (Wu, et al., 2008). This too shows advancements in technology which have contributed to the optimization of the biohydrogen production process.

The last factor that has yet to be discussed is the temperature. As temperature plays a vital role in any reactor, significant results were seen in Lee, et al., (2014) where there was a comparison done between mesophilic and thermophilic reactors. In this experiment, the reactors were operated at 37°C and 55°C respectively, keeping all of the other operating conditions the same. From this, a yield of 5.34 mmol H<sub>2</sub>/g starch was obtained from the 37°C test which was significantly higher than the yield from the 55°C test of 1.44 mmol H<sub>2</sub>/g starch (Lee, et al., 2014). The mesophilic test also showed a much higher amount of hydrogen produced as well as a higher hydrogen production potential as shown in figure 11 below, which is contrary to the typical hydrogen yield which increases with an increased temperature. Through this test, some considerations can be made that some hydrogen producing bacteria may produce better under mesophilic conditions.

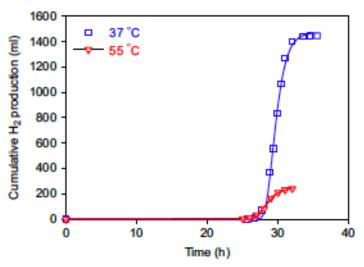


Figure 11: Mesophilic Vs. Thermophilic Hydrogen Production (Lee, et al., 2014)

## 5.3 Bio-Electricity

## Table 3: Summary of Literature Findings for Bio-Electricity Production

Waste Type	Reactor Type & Operating Conditions	Bio-Electrical Power Yield	Reference
Food Waste Leachate	Fed Batch, 28°C, pH 5.39	316.12 mW/m <sup>3</sup>	(Li, et al., 2013)
Food Waste Leachate	Fed Batch, 28°C, pH 5.15	445.61 mW/m <sup>3</sup>	(Li, et al., 2013)
Food Waste Leachate	Fed Batch, 28°C, pH 4.8	453.9 mW/m <sup>3</sup>	(Li, et al., 2013)
Powdered Orange Peel Waste	Batch, 30°C, 10 Days, pH 6.45	227.5 mW/m <sup>2</sup>	(Miran, et al., 2016)
Filtered Orange Peel Waste	Batch, 30°C, 10 Days, pH 6.45	358.8 mW/m <sup>2</sup>	(Miran, et al., 2016)
Food & Dairy Wastewater	Batch, 22°C, 19 Days, pH 5.3	150 mW/m <sup>2</sup>	(Nimje, et al., 2012)
Activated Sludge	Fed Batch, 24°C, 150 Days, pH 4.4	280.0 mW/m <sup>2</sup> or 12.9 W/m <sup>3</sup>	(Kim, et al., 2014)
Food Waste	Batch, 25°C, 6 Days, pH 4	19.15 W/m <sup>3</sup>	(Hou, et al., 2016)
Food Waste Leachate from LBR Reactor	Batch, 27°C 17 Days, p H Variable	14.42 mW/m <sup>2</sup>	(Moharir, et al., 2018)
Food Waste Leachate from LBR Reactor	Batch, 27ºC 17 Days, pH Variable	29.25 mW/m <sup>2</sup> or 974.89 mW/m <sup>3</sup>	(Moharir, et al., 2018)
Distillery Wastewater	Batch, 31°C, 8 Days, pH 8	80 mW/m <sup>2</sup>	(Samsudeen, et al., 2016)
Organic Fraction of Municipal Solid Waste	Fed Batch, 20-35°C, 7 Days, pH 7.2	47.6 mW/m <sup>2</sup> or 0.72mW/m <sup>3</sup>	(Karluvali, et al., 2015)
Cattle Manure	Sequential Batch, 22-24°C, 65 Days, pH 7	165.03 mW/m <sup>2</sup> or 6.6W/m <sup>3</sup>	(ElMekawy, et al., 2014)
Acetate	Fed Batch, 28°C, 7 Days, pH 7	64.3 mW/m <sup>2</sup>	(Chae, et al., 2009)
Butyrate	Fed Batch, 28°C, 7 Days, pH 7	51.4 mW/m <sup>2</sup>	(Chae, et al., 2009)
Propionate	Fed Batch, 28°C, 7 Days, pH 7	58.0 mW/m <sup>2</sup>	(Chae, et al., 2009)
Glucose	Fed Batch, 28°C, 7 Days, pH 7	156.0 mW/m <sup>2</sup>	(Chae, et al., 2009)

Bio-electrical production may be one of the most effective ways to obtain a renewable source of electricity. As the electricity that is produced can be directly utilized in the electrical grid without the need to burn or convert the energy to another source, thus reducing the amount of carbon dioxide emissions and improving the ecological footprint for the process. In conducting the conversion of various wastes to bio-electricity, the optimization of a MFC is essential. As shown in table 3, the pH of the waste and type of inoculum has a very influential output capacity from the MFC. In the study conducted by Li, et al., (2013) the production of bioelectricity from food waste (bread, rice, cabbage and pork mixture) was tested under different scenarios.

The waste source was consistent throughout the experiment, but the inoculum source changed from wastewater to activated sludge to anaerobic sludge, thus changing the pH of the MFC from 5.39 to 4.18 as shown in table 3 (Li, et al., 2013). With the change in pH and inoculum type, there was an increase in the amount of power obtained from the MFC from 316.12 mW/m<sup>3</sup> to 453.9 mW/m<sup>3</sup>. As suggested in Li, et al., (2013), the highest value obtained in the anaerobic sludge may be a result of the enrichment of the fermentative and electrogenic species within the fuel cell, but evidence can be drawn that the low pH has a significant effect on the power generation as also seen in Samsudeen, et al., (2016) and Kim, et al., (2014). Although there was a noticeable difference between the amount of power obtained between the 3 different MFCs, the experiment bolstered an 87% COD and VFA removal rate which suggests it is an effective method for the purification of effluent prior to its release (Li, et al., 2013).

In contrast to changing the inoculum type, a change in the substrate will also have an adverse effect on the bioelectrical production from an MFC. As experimented in Chae, et al., (2009), 4 different substrate types were compared, in which the highest power density was

obtained from glucose. This is not to say that other substrates were not ideal for electricity generation, as all of the substrates demonstrated properties that would make each of them ideal for the production of bioelectricity (Chae, et al., 2009). Acetate had the highest columbic efficiency as it did not have electron loss due to competing bacteria in comparison to glucose which did (Chae, et al., 2009). This could have been minimized by pre-treating the inoculum in order to remove the methanogenic bacteria. As the glucose yielded the highest power density, it was not sustainable due to the presence of the competing bacteria, thus making it less ideal in this particular experiment for the production of bioelectricity (Chae, et al., 2009). The effect of particle size was also experimented in Miran, et al., (2016) where smaller granulated particle sizes in the substrate prior to treatment yielded the highest power yield from this experiment. From this article the conclusion can be drawn that the smaller the particle size and the more treatment prior to implementation in the MFC, the higher the energy yield will be.

Equally important is the effect of temperature to a MFC. As experimented in Karluvali, et al., (2015), the organic fraction of municipal solid waste (MSW) underwent a series of experiments in which the temperature was altered from 20-35°C in 5°C increments. As the temperature increased the columbic efficiency, COD removal rate and bio-electrical production rate increased (Karluvali, et al., 2015). This should be expected as particulate and bacterial matter becomes more active with an increase to the temperature. From Karluvali, et al., (2015), it was suggested that MFCs would be an alternative to typical digesters in colder environments as methanogens are very sensitive to thermal changes, whereas MFCs are not.

Experimented in Moharir, et al., (2018) and ElMekawy, et al., (2014), are the effects of recirculation and OLR within a batch MFC. As shown in table 3, the effects of recirculation bolstered the production of electricity as the yields were much higher than that of the typical batch method. The COD removal efficiency of the recirculating reactor was also higher than the typical batch reactor for all of the different OLRs (Moharir, et al., 2018). As recirculation promotes a higher mixing of components, as well as a more active waste source mirroring the effects of an increased temperature, it was concluded that the recirculation of waste is effective in increasing the output of the MFC. If a recirculating reactor is used, there is even more benefits that can be seen from this experiment setup as there would be a reduction in the amount of lag time, allowing the MFC to produce electricity at an optimal rate.

Similar to previous studies done for the production of biomethane and biohydrogen, the OLR for a MFC plays an important part in the production of bioelectricity. ElMekawy, et al., (2014) experimented with the production of bioelectricity from different 3 OLRs. As the experiment progressed, the OLR rate was increased to triple the original concentration. As there is a limitation to the amount of waste the bacteria can convert, the lowest of the 3 organic loading rates provided the highest power generation potential as well as the highest COD removal rate and columbic efficiency (ElMekawy, et al., 2014). As expected, there is a defined number of organisms within the MFC and the reactor can only handle a certain OLR in order to produce electricity at an optimum rate.

## 5.4 Polyhydroalkanoates (PHA's)

Table 4: Summary of Literature Findings for PHA Production

Waste Type	Reactor Type & Operating Conditions	PHA Composition	Reference
Municipal Wastewater	Batch, 20°C, 225 Days, pH 4.5	49%	(Bengtsson, et al., 2017)
Glucose	Batch, 30°C, 2 Days, pH Uncontrolled	32%	(Munir, et al., 2018)
Glucose & Propionic Acid	Batch, 30°C, 2 Days, pH Uncontrolled	35%	(Munir, et al., 2018)
Fresh Cheese Whey	CSTR, 25°C, 4 Days, pH 8.8	659 gPHA/kgVSS	(Colombo, et al., 2016)
Sterilized Cheese Whey	CSTR, 25°C, 4 Days, pH 8.8	814 gPHA/kgVSS	(Colombo, et al., 2016)
Glucose	Batch, 37°C, 8 Days, pH Uncontrolled	68%	(Ray, et al., 2017)
Crude Glycerol	Batch, 37°C, 8 Days, pH Uncontrolled	71%	(Ray, et al., 2017)
Fermented Molasses	Fed Batch, 30°C, 300 Days, pH 6	75%	(Albuquerque, et al., 2010)
Fermented Molasses	Continuous, 30°C, 10 Days, pH 8.4	77%	(Albuquerque, et al., 2011)
Cheese Whey Powder	Fed Batch, 23-25°C, 46 Days, pH 8	65%	(Duque, et al., 2014)
Sugar Cane Molasses	Fed Batch, 23-25°C, 29 Days, pH 8	56%	(Duque, et al., 2014)
Candy Factory Wastewater	Fed Batch, 30°C, 58 Days, pH 7.5	760 gPHA/kg VSS	(Tamis, et al., 2014)
Food and Plant Sludge from MWWTP	Fed Batch, 25°C, 60 Days, pH Uncontrolled	48%	(Zhang, et al., 2014)

The synthesis of PHAs from VFAs, may yield a variety of different PHA concentrations based on the type of waste used as shown in table 4. Similar to many of the other production processes previously discussed, the production of PHAs also exhibited a relationship in the feed regime which was not linear. From Albuquerque, et al., (2010), a PHA yield increase was seen from feeding concentrations of 30 to 45 Cmmol VFA/L followed by a decrease when the feeding concentration was further increased to 60 Cmmol VFA/L. This can once again conclude that if the organisms are not readily available to digest the waste and convert it to the desired product than the increase in the rate is not effective. In this particular experiment, the optimal production was seen at a concentration of 45 Cmmol VFA/L as there was no limitations on the concentration of the feast ratio of the bacteria thus creating an optimal result and yielding a 75% PHA content (Albuquerque, et al., 2010).

Also similar to many of the previous studies, it was seen that the lag time was eliminated through the use of a continuous reactor in comparison to batch reactors. As shown in Albuquerque, et al., (2011), there was a reduction in the amount of lag time once new waste was implemented into the system. Additionally, there was an increase in the hydroxyvalerate (HV) content by about 8% (Albuquerque, et al., 2011), showing that the continuous feeding strategy may allow for the manipulation of polymers and increase the desired structures required for optimal PHA production. The manipulation of polymers was also experimented with in Duque, et al., (2014), where it was found that polymer composition can be manipulated by altering the pH and HRT in the acidogenic phase to yield the desired polymers. This has been a particular challenge when using a real feedstock for the production of PHAs, as it is difficult to obtain a polymer composition that is consistent in order to have a continuous production of PHAs (Duque,

et al., 2014). It was also seen that in the production of particular VFAs from manipulated polymers, there was a narrow scope of VFAs produced (Duque, et al., 2014). From this it may be concluded that by using a mixed culture there is the potential to widen the scope of polymers produced and by extrapolation, the type of VFAs produced, ultimately yielding a higher PHA content.

The amount of solids within the reactor at the time of PHA production may have a significant impact on the production of said PHAs. There is a strong correlation between the amount of suspended solids in the effluent after the fermentation stage and the amount of PHAs produced (Tamis, et al., 2014). The amount of suspended solids after the fermentation stage should be kept at a minimum in order to reduce the amount of ethanol and methane produced during the PHA production process (Tamis, et al., 2014). With the production of either of these other value added products, there is valuable VFAs or other PHA producing compounds used, thus lowering the overall amount of PHAs produced and rendering the process less effective than it should be.

If there are any suspended solids within the reactor at the time of PHA production, a lower sludge retention time (SRT) should be desired (Bengtsson, et al., 2017). With a decrease in the SRT, an increase in the PHA yield and overall process productivity and economic feasibility is seen. The PHA production process is easily compatible with processes which work to remove nitrogen, as well as lower the total chemical oxygen demand within a waste stream (Bengtsson, et al., 2017).

## 5.5 Biological Nutrient Removal (BNR)

Table 5: Summary of Literature Findings for Biological Nutrient Removal

Carbon Source	Reactor Type & Operating Conditions	BNR Rate	Reference
Wasterwater from Quafa WWTP	Continuous, 24°C, 60 Days, pH 7.2	90% Phosphorus Removal	(Rashed, et al., 2015)
Sludge From WWTP	Batch, 20 Days, pH 7.5	75.7% Nitrogen Removal	(Tong, et al., 2009)
Sludge From WWTP	Batch, 20 Days, pH 7.5	83% Phosphorus Removal	(Tong, et al., 2009)
Waste Activated Sludge from Shanghai WWTP	Batch, 21°C, 90 Days, pH 7.5	82% Nitrogen Removal	(Zheng, et al., 2010)
Waste Activated Sludge from Shanghai WWTP	Batch, 21°C, 90 Days, pH 7.5	95% Nitrogen Removal	(Zheng, et al., 2010)
Waste Activated Sludge from Qingdao WWTP	Batch, 24°C, 60 Days, pH 7.5	96.4% Nitrogen Removal	(Shao, et al., 2019)
Waste Activated Sludge from Langfang WWTP	Batch, 30°C, 5 Days, pH 6.9	86.4% Nitrogen Removal	(Pang, et al., 2015)
Waste Activated Sludge from Chunliu WWTP	Batch, 30°C, 45 Days, pH 8.2	95% Nitrogen Removal	(Wang, et al., 2016)
Food Waste Fermentation Liquid	Batch, 25°C, 2.5 Days, pH 7.5	98% Nitrogen Removal	(Zhang, et al., 2016)
Glucose	Batch, 25°C, 2.5 Days, pH 7.5	98% Nitrogen Removal	(Zhang, et al., 2016)
Sodium Acetate	Batch, 25°C, 2.5 Days, pH 7.5	39.6% Nitrogen Removal	(Zhang, et al., 2016)
<b>Oil-Removed Food Waste</b>	Batch, 25°C. 7 Days, pH 7.8	88.1% Nitrogen Removal	(Yan, et al., 2018)
Oil-Added Food Waste	Batch, 25°C. 7 Days, pH 7.8	92.8% Nitrogen Removal	(Yan, et al., 2018)
Sodium Acetate	Batch, 25°C. 7 Days, pH 7.8	90% Nitrogen Removal	(Yan, et al., 2018)
Waste Activated Sludge from Qingdao WWTP	Batch, 25°C, 40 Days, pH 7.5	96.3% Nitrogen Removal	(Guo, et al., 2017)

Biological nutrient removal (BNR), is proven to be an effective method for the reduction of specific nutrients from waste streams prior to their release back into the environment. As shown in table 5, the most prominent nutrient removed from waste streams is nitrogen, with phosphorus being the second most prominent. The BNR process is not only effective, but also efficient. Using specific wastes as a carbon source for the reduction of nutrients has yielded high reduction rates (Table 5) if the appropriate waste type is selected for the digester. With a full nutrient removal from the waste, the bi-products, most prominently nitrogen, can be used for other processes in the nitrogen cycle as it is one of the most abundant elements on earth. Within each of these processes there are specific considerations for the removal of nutrients.

The use of waste activated sludge fermentation liquid has been a focal point for the removal of nutrients from various wastewater streams. In using alkaline fermentative liquid, there is seen to be an increase in the overall nutrient removal for nitrogen and phosphorus (Tong, et al., 2009). In comparison to acetic acid as a carbon source for nutrient removal, the waste activated sludge fermentative liquid yielded an 8% higher removal rate for both nitrogen and phosphorus (Zheng, et al., 2010). There was also better PHA utilization performance for both nitrogen and phosphorus uptake, which is a key indicator for the overall nutrient removal process (Zheng, et al., 2010). In the trade off for removing more of the nutrient from the waste stream, it has been seen that there is a slight effluent COD increase for alkaline fermentative liquid reactor in comparison to the acetic acid, which may be attributed to the humic acid which is produced during the fermentation period (Tong, et al., 2009). This may not be seen as a particular issue due to the fact that the removal rates for BOD and COD can be very high (93%) as seen in (Rashed, et al., 2015).

Essential to the nutrient removal process is the C/N ratio as it can often dictate the amount of remaining nitrogen in the effluent. In Yan, et al., (2018), is was discovered that with a C/N ratio of 7 and a temperature of 25°C, the general discharge standards could be met for nitrogen and COD content in effluents that were to be discharged into Chinese wastewater streams of watercourses in the region. This is quite similar to what was determined in Zhang, et al., (2016) where it was determined that the optimal C/N ratio and temperature was 6 and 25°C respectively. Although higher temperatures will achieve a high hydrolysis rate, 25°C was determined as an optimum due to the fact that acidogenesis yields were higher with a lower temperature (Zhang, et al., 2016).

Using glucose and acetic acid as the basis for nutrient removal, there is seen to be a higher percentage of nutrient removal with the incorporation of additional wastes as carbon sources downstream. As experiment in Zhang, et al., (2016), the fermentative liquid from food waste allowed the optimal nitrogen removal to occur two hours sooner than the glucose under the same conditions. Compared to sodium acetate, the removal efficiency of the fermented liquid (study dependant) was always higher than that of sodium acetate as shown in table 5. This may be attributed to the fact that using sodium acetate as a carbon source only provides that waste with a singular compound and there are various types of bacteria within the reactor. Using different waste streams as a source for the nutrient removal provides the bacteria with alternatives to acetate that may be more favoured, thus engaging all bacteria and making them active instead of one particular type (Yan, et al., 2018). This would imply that using various fermentative liquids may have a beneficial reaction and actually work to increase the efficiency and productivity of a general reactor (Guo, et al., 2017).

## 6.0 Conclusion

The production of value-added products from waste streams is becoming a hot topic in the area of environmental engineering due to the large amount of benefits that can be obtained from the production process. In an attempt to produce a circular economy, the goal is to keep resources in use for as long as possible, which will even include waste products for various streams (Neligen, 2018). By obtaining value added products from wastes, the life of a product or its bi-products is extended further to essentially double down on the production process. Through various studies, there is further research being done on the optimal operating conditions in order to obtain the maximum output from a waste stream. As shown in tables 1-5 it can be determined that VFA based wastes as a suitable substrate for the production of methane, hydrogen and electricity as well as bioplastics from PHAs and the removal of nutrients in the biological nutrient removal process.

In order to maximize the amount of VFAs within the waste, pre-treatment may be necessary in order to isolate the VFAs for the production of the desired products. In doing this, an economic benefit must be seen to justify the additional step in the production process. Further studies must be done on each waste in particular in order to optimize the number of value-added products that are obtained from the waste stream itself. Through this research, owners will have the opportunity to truly know what the composition of their waste is and to utilize it for production of a particular value-added product. Similarly, to analyzing the waste prior to beginning the production process it is important to understand the bacterial community which is producing the value-added product. Seen in almost all the experiments in the tables for this

paper, each of the wastes are analyzed prior to their use in the reactor along with the type of microbial community, leading to successful results being obtained.

Optimal conditions for the digestion of waste are quite variable as no one waste is the same. The uncertainty of some wastes already produced, in addition to the wastes which are currently being experimented with is the main reason why the use of some wastes is still in the experimental stage instead of them being implemented at a field level. Although optimal conditions can be speculated, a large variety of operating conditions are seen in tables 1-5 for the production of the desired product from a particular waste stream. There has been some conclusive evidence for the production of value-added products such as temperature and pH, particularly in the PHA production process and BNR process, but there is a large variability in retention time and the type of reactor being used for almost all of the processes.

In order to successfully create a circular economy, further understanding must be made in the area of production of value-added products from waste streams. Through this paper it can be seen that waste streams comprised of VFA based materials can be a viable source for the production of value-added products. In cases such as (Wang, et al., 2011), a particular waste stream can be used for the production of more than one value added products. In the coproduction of value-added products, the core values of a circular economy are further exemplified, driving the overall environmental impact even lower. Though there has been a large amount of advancements seen in this field, there is further research that must be done, in regards to optimal operating conditions, reactor configurations and wastes types in order to achieve the maximum potential from these wastes.

## References

- Abgenent, L. T., Karim, K., Al-Dahhan, M. H., Wrenn, B. A., & Domiguez-Espinosa, R. (2004). Production of bioenergy and biochemical from industrial agricultural wastewater. *TRENDS in Biotechnology*, 477-485.
- Albuquerque, M., Martino, V., Pollet, E., Averous, L., & Reis, M. (2011). Mixed culture polyhydroxyalkanoate (PHA) production from volatile fatty acid (VFA)-rich streams: Effect of substrate composition and feeding regime on PHA productivity, composition and properties. *Journal of Biotechnology*, 151, 66-76.
- Albuquerque, M., Torres, C., & Reis, M. (2010). Polyhydroxyalkanoate (PHA) production by mixed microbial culture using sugar molasses: effect of the infleunt substrate concentration on culture selection. *Water Research*, 3419-3433.
- Alexandre, G., Amorim, H., Lopes, M., & Oliviera, M. (2008). Continuous and batch fermetation processes: advantages and disadvantages of these process in the Brazilian ethanol production. *International Sugar Journal*, 175-181. Retrieved from Research Gate: https://www.researchgate.net/publication/289874380\_Continuous\_and\_batch\_fermen tation\_processes\_Advantages\_and\_disadvantages\_of\_these\_processes\_in\_the\_Brazilia n\_ethanol\_production
- Alzate Marin, J. C., Caravelli, A. H., & Zaritzky, N. E. (2016). Nitrification and aerobic denitridication in anoxic-aerobic sequencing batch reactor. *Bioresrouce Technology*, 380-387.
- Bengtsson, S., Karlsson, A., Alexandersson, T., Quadri, L., Hjort, M., Johansson, P., . . . Alan Werker. (2017). A process for polyhydroxyalkanoate (PHA) production from municipal wastewater treatment with biological carbon and nitrogen removal demonstrated at pilot-scale. *New Biotechnology*, 35, 42-53.
- Center for Industry Education and Collaboration. (2013, March 18). Chemical Reactors. Retrieved from University of York:

http://www.essentialchemicalindustry.org/processes/chemical-reactors.html

- Chae, K.-J., Choi, M.-J., Lee, J.-W., Kim, K.-Y., & Kim, I. S. (2009). Effect of different substrates on the performance, bacterial diversity, and bacterial viability in microbial fuel cells. *Bioresource Technology, 100*, 3518-3525.
- Chang, J.-J., Wu, J.-H., Wen, F.-S., Hung, K.-Y., Chen, Y.-T., Hsaio, C.-L., . . . Huang, C.-C. (2008). Molecular monitoring of microbes in a continuous hydrogen producing system with differnet hydraulic retention times. *International Journal of Hydrogen Energy*, 1579-1585.
- Chen, C.-C., Chen, H.-P., Wu, J.-H., & Lin, C.-Y. (2008). Fermentative hydrogen production at high sulfate concentration. *Internation Journal of Hydrogen Energy*, *33*, 1573-1578.
- Chong, S., Tushar, K. S., Kayaalp, A., & Ang, H. M. (2012). The performance enhancements of upflow anaerobic sludge blanket (UASB) reactors for domestic sludge treatment a state of the art review. *Water Research*, 3434-3470.
- Colombo, B., Sciarria, T. P., Reis, M., Scaglia, B., & Adani, F. (2016). Polyhydroxyalkanoates (PHAs) production from fermeneted chesse whey by using a mixed microbial culture. *Bioresource Technology, 218*, 692-699.

- Davila-Vazquez, G., Cota-Navarro, C. B., Rosales-Colunga, L. M., de Leon-Rodriguez, A., & Razo-Flores, E. (2009). Continuous biohydrogen production using cheese whey: Improving the hydrogen production rate. *International Journal of Hydrogen Energy*, *34*, 4296-4304.
- Detalina, M., Pradanawati, S. A., Widyarani, Mamat, Nilawati, D., & Sintawardani, N. (2018). The influence of bamboo packed configuration to mixing charecterisitcs in a fixed bed reactor. *Journal of Physics*.
- Duque, A. F., Oliveira, C. S., Carmo, I. T., Gouveia, A. R., Pardelha, F., Ramos, A. M., & Reis, M. A. (2014). Response of a three-stage process for PHA production by mixed microbial cultures to feedstock shift: impact on polymer composition. *New Biotechnology*, *31*, 276-288.
- ElMekawy, A., Srikanth, S., Vanbroekhoven, K., De Wever, H., & Pant, D. (2014). Bioelectrocatalytic valorization of dark fermenetation effleuents by acetate oxidizing bacteria in bioelectrochemical system (BES). *Journal of Power Sciences, 262*, 183-191.
- Eriksson, E., & Gray, E. A. (2017). Optimization and integration of hybrid renewable energy hydrogen fuel cell sysyems A critical review. *Applied Energy*, 348-364.
- Escalante, H., Castro, L., Amaya, M., Jaimes, L., & Jaimes-Estevez, J. (2018). Anaerobic digestion of cheese whey: Energetic and nutritional potential for the dairy sector in developing countries. *Waste Management*, 711-718.
- Evvyernie, D., Morimoto, K., Karita, S., Kimura, T., Sakka, K., & Ohmiya, K. (2001). Conversion of chitinous wastes to hydrogen gas by clostridium paraputrificum M-21. *Journal of Bioscience and Bioengineering*, 339-343.
- Fang, H. H., & Liu, H. (2002). Effect of pH on hydrogen prodcution from glucsose by a mixed culture. *Bioresource Technology, 82*, 87-93.
- Forger, & Gurman. (2007). *Industrial Reactors*. Retrieved from University of Michigan: http://umich.edu/~essen/html/01chap/html/industrial.htm
- Ghimire, A., Frunzo, L., Pontoni, L., d'Antonio, G., Lens, P. N., Esposito, G., & Pirozzi, F. (2015). Dark fermentation of complex waste biomass for biohydrogen production by pretreated thermophilic anerobic digestate. *Journal of Environmental Management, 152*, 43-48.
- Grady, J., Leslie, C., Daigger, G. T., Love, N. G., & Filipe, C. D. (2011). *Biological Wastewater Treatment.* Boca Raton: CRC Press.
- Guo, Y., Guo, L., Sun, M., Zhao, Y., Gao, M., & She, Z. (2017). Effect of hydraulic retention time (HRT) on denitrification using waste activated sludge thermal hydrolysis liquid and acidogenic liquid as carbon sources. *Bioresource Technology*, 224, 147-156.
- Hafez, H. (2019, February). Lecture 3: Fundamentals of Anaerobic Digestion. Toronto, Ontario, Canada.
- Han, M. H., Behera, S. K., & Park, H.-S. (2012). Anaerobic co-digestion of food waste leachate and piggery wastewater for methane production: statistical optimization of key process parameters. *Journal of Chemical Technology & Biotechnology*, 1541-1550.
- Hernandez, D., Solana, M., Riano, B., Garcia-Gonzalez, M. C., & Bertucco, A. (2014). Biofuels from microalgae: Lipid Extraction and methane production from the residual biomass in a biorefinery apporoach. *Bioresource Technology*, 370-378.
- Hou, Q., Pei, H., Hu, W., Jiang, L., & Yu, Z. (2016). Mutual facilitations of food waste treatment microbial fuel cell bioelectricty generation and Chlorella vulgaris lipid production. *Bioresource Technology*, 203, 50-55.

- Ji, Z., Chen, G., & Chen, Y. (2010). Effects of waste activated sludge and surgactant addition on primary sludge hydrolysis and short-chain fatty acids accumulation. *Bioresource Technology*, 3457-3462.
- Kamali, M., Gameiro, T., Costa, M. E., & Capela, I. (2016). Anaerobic digestion of pulp and paper mill wastes - an overview of the developments and improvement oppurtunities. *Chemical Engineering Journal*, 162-182.
- Karluvali, A., Koroglu, E. O., Manav, N., Cetinkaya, A. Y., & Ozkaya, B. (2015). Electricity generation from organic fraction of municipal solid wastes in tubular microbial fuel cell. *Seperation and Purification Technology*, *156*, 502-511.
- Kim, H., Kim, B., Kim, J., Lee, T., & Yu, J. (2014). Electricity generation and microbial community in microbial fuel cell using low-pH distillery wastewater at different extremal resistances. *Journal of Biotechnology*, 186, 175-180.
- Kim, J., & Kang, C.-M. (2015). Increased anaerobic production of methane by co-digestion of slidge with microalgal biomass and food waste leachate. *Bioresource Technology*, 409-412.
- Lay, C.-H., Sen, B., Kuo, S.-Y., Chen, C.-C., & Lin, C.-Y. (2014). Biohydrogen production from mixed textile wastewater by mixed microflora in an intermittent-flow, stirred tank reactor: effect of feeding frequency. *Journal of the Chienese Chemical Society*, 61, 791-796.
- Lee, C., Lee, S., Han, S.-K., & Hwang, S. (2014). Effect of operational pH on bioghydrogen production from food waste using anaerobic batch reactors. *Water Science and Technology*, 1886-1893.
- Lee, K.-S., Hsu, H.-F., Lo, Y.-C., Lin, P.-J., Lin, C.-Y., & Chang , J.-S. (2008). Exploring optimal environmental factors for fermentative hydrogen production from starch using mixed anaerobic microflora. *International Journal of Hydrogen Energy*, *33*, 1565-1572.
- Lee, W. S., Chua, A. S., Yeoh, H. K., & Ngoh, C. G. (2014). A Review of the production and applications of waste-derived volate fatty acids. *Chemical Engineering Journal*, 83-99.
- Li, K., Liu, R., & Sun, C. (2015). Comparison of anaerobic digestion characteristics and kinterics of four livestock manures with different substrate concentrations. *Bioresource Technology*, 133-140.
- Li, W., Siddhu, M. A., Amin, F. R., He, Y., Zhang, R., Liu, G., & Chen, C. (2018). Methane production through anaerobic co-digestion of sheep dung and waste paper. *Energy Conversion and Management*, 279-287.
- Li, X. M., Cheng, K. Y., Selvam, A., & Wong, J. W. (2013). Bioelectricity production from acidic food waste leachate using microbial fuel cells: Effect of microbial inocula. *Process Biochemistry*, 283-288.
- Lyu, Z., Shao, N., Akinyemi, T., & Whitman, W. B. (2018). Methanogensis. *Current Biology*, 727-732.
- McEniry, J., & O'Kiely, P. (2013). Anaerobic methane production from five common grassland species at sequential stages of maturity. *Bioresource Technology*, 143-150.
- Miran, W., Nawaz, M., Jang, J., & Lee, D. S. (2016). Conversion of orange peel waste biomass to bioelectricity using a mediator-less microbial fuel cell. *Science of the Total Environment*, 547, 197-205.

- Moharir, P. V., & Tembhurkar, A. R. (2018). Effect of recirculation of bioelectricity generation using microbial fuell cell with food waste leachate as substrate. *International Journal of Hydrogen Energy*, *43*, 10061-10069.
- Mu, Y., Wang, G., & Yu, H.-Q. (2006). Response surface methodologicla analysis on biohydrogen production by enriched anaerobic cultures. *Enzyme and Microbial Technology, 38*, 905-913.
- Munir, S., & Jamil, N. (2018). Polyhydroxyalkanoates (PHA) production in bacterial co-culture using glucose and volatile fatty acids as carbon source. *Journal of Basic Microbiology, 58*, 247-254.
- Najafpour, G., & Ebrahimi, A. (2016). Biological Treatment Processes: Suspended Growth vs. Attached Growth. *Iranica Journal of Energy and Environment*, 114-123.
- Neligen, A. S. (2018). *EU Circular Economy Package: A Challenging Yet Important Impulse.* The Conference Board.
- Nelson, M. J., Naklha, G., & Zhu, J. (2017). Fluidized-Bed Bioreactor Application for Biological Wastewater Treatment: A Review of Research and Developments. *Engineering*, 330-342.
- Nimje, V. R., Chen, C.-Y., Chen, H.-R., Chen, C.-C., Huang, Y. M., Tseng, M.-J., ... Chang, Y.-F. (2012). Comparative bioelectricity productino from various wastewaters in microbial fuel cells using mixed cultures and a pute strain of Shewanella oneidensis. *Bioresource Technology*, 104, 315-323.
- Pang, L., Ni, J., Tang, X., & Chen, Q. (2015). Short-cut waste activated sludge fermentation and application of fermentation liquid to improve heterotrophic aerobic nitrogen removal by Agrobacterium sp. LAD9. *Chemical Engineering Journal, 259*, 911-917.
- Pattra, S., Sangyoka, S., Boonmee, M., & Reungsang, A. (2008). Bio-hydrogen production from the fermentation of sugarcane bagasse hydrolysate by clostridium butyricum. *Internation Journal of Hydrogen Energy*, 5256-5265.
- Pinto, N., Carvalho, A., Pacheco, J., & Duarte, E. (2016). Study of different ratios of primary and waste activated sludges to enhance the methane yield. *Water and Environment Journal*, 203-210.
- Rashed, E. M., & Massoud, M. (2015). The effect of effective microorganisms on EBPR in modified contact stabilization system. *HBRC Journal*, *11*, 384-392.
- Ray, S., & Kalia, V. C. (2017). Polyhydroxyalkanoate production and degreadation patterns in Bacillus species. *Indian Journal of Microbiology*, *57*, 387-392.
- Resch, G., Held, A., Faber, T., Panzer, C., Toro, F., & Haas, R. (2008). Potentials and Prospects for Renewable Energies at Global Scale. *Energy Policy 36*, 4048-4056.
- Reungsang, A., Sittijunda, S., & Sreela-or, C. (2016). Methane production from acidic effluent discharged after the hydrogen fermentation of sugarcane juice using batch fermentation and UASB reactor. *Renewable Energy*, 86: 1224-1231.
- Samsudeen, N., Radhakrishnan, T., & Matheswaran, M. (2016). Effect of isolated bacterial strains from distillery wastewater on power generation in a microbial fuel cell. *Process Biochemistry*, 1876-1884.
- Shao, M., Guo, L., She, Z., Gao, M., Zhao, Y., Sun, M., & Gup, Y. (2019). Enhancing denitrification efficiency for nitrogen removal using waste sludge alkaline fermentation liquid as external carbon source. *Environmental Science and Pollution Research, 26*, 4633-4644.

- Silva-Illanes, F., Tapia-Venegas, E., Sciappacasse, C., Trably, E., & Ruiz-Filippi, G. (2017). Impact of hydraulic rentention time (HRT) and pH on dark fermentative hydrogen production from glycerol. *Energy*, *141*, 358-367.
- Sivagurunathan, P., Kumar, G., Kobayashi, T., Xu, K., Kim, S.-H., Nguyen, D. D., & Chang, S. W. (2018). Co-digestion of untreated macro and microalgal biomass for biohydrogen production: impact of incoculum augmentation and microbial insights. *Internation Journal of Biohydrogen Energy*, 43, 11484-11492.
- Strazzera, G., Battista, F., Garcia, N. H., Frison, N., & Bolzonella, D. (2018). Volatile fatty acids production from food wastes for biorefinery platforms: A review. *Journal of Environemental Management*, 278-288.
- Tamis, J., Luzkov, K., Jiang, Y., van Loosdrecht, M. C., & Kleerebezem, R. (2014). Enrichment of Plasticicumulans acidivorans at pilot-scale for PHA production on industrial wastewater. *Journal of Biotechnology, 192*, 161-169.
- Tharali, A. D., Sain, N., & Osborne, J. (2016). Microbial fuel cells in bioelectricity production. *Frontiers in Life Science*, 252-266.
- Tong, J., & Chen, Y. (2009). Recovery of nitrogen and phosphorus from alkaline fermenetation liquid of waste activated sludge and application of the fermentation liquid to promote biological municpal wastewater treatment. *Water Research*, *43*, 2969-2976.
- Wang, W., Xie, L., Chen, J., Luo, G., & Zhou, G. (2011). Biohydrogen and methane production by co-digestion of cassava stillage and excess sludge under thermophilic conditions. *Bioresource Technology*, *102*, 3833-3839.
- Wang, X., Zhang, Y., Zhang, T., Zhou, J., & Chen, M. (2016). Waste activated sludge fermenation liquid as carbon source for biological treatment of sulfide and nitrate in microaerobic conditions. *Chemical Engineering Journal, 283*, 167-174.
- Wilberforce, T., El-Hassan, Z., Khatib, F., Al Makky, A., Baroutaji, A., Carton, J. G., & Olabi, A. G.
  (2017). Develops of electric cars and fuel cell hydrogen electric cars. *International Journal of Hydrogen Energy*, 25695-25734.
- World Health Organization. (2018, May 2). 9 out of 10 people worldwide breathe polluted air, but more countries are taking action. Retrieved from World Health Organization: https://www.who.int/news-room/detail/02-05-2018-9-out-of-10-people-worldwidebreathe-polluted-air-but-more-countries-are-taking-action
- Wu, S.-Y., Hung, C.-H., Lin, C.-Y., Lin, P.-J., Lee, K.-S., Lin, C.-N., . . . Chang, J.-S. (2008). HRTdependent hydrogen production and bacterial community structure of mixed anaerobic microflora in suspended, graular and immobilized sludge systems using glucose as the carbon source. *International Journal of Hydrogen Energy*, 33, 1542-1549.
- Yan, F., Jiang, J., Zhang, H., Liu, N., & Zou, Q. (2018). Biological denitrification from mature landfill leachate using a food-waste-derived carbon source. *Journal of Environenmental Management*, 214, 184-191.
- Zhang, C., Su, H., Baeyens, J., & Tan, T. (2014). Reviewing the anaerobic digestion of food wastes for biogas production. *Renewable and Sustainable Energy Reviews*, 383-392.
- Zhang, M., Wu, H., & Chen, H. (2014). Coupling of polyhydroxylalkanoate production with volatile fatty acid from food wastes and excess sludge. *Process Safety and Environmental Protection, 92*, 171-178.

- Zhang, Y., Wang, X. C., Cheng, Z., Li, Y., & Tang, J. (2016). Effect of fermentation liquid from food waste as a carbon source for enhancing denitrification in wastewater treatment. *Chemosphere, 144*, 689-696.
- Zheng, X., Chen, Y., & Liu, C. (2010). Waste Activated Sludge Alkaline Fermentation Liquid as Cabon Source for Biological Nutrient Removal in Anaerobic Followed by Alternating Aerobic-Anoxic Sequencing Batch Reactors. *Chinese Journal of Chemical Engineering, 18,* 478-485.