VALORIZATION OF FOOD WASTE FOR BIOETHANOL PRODUCTION AND OTHER VALUABLE PRODUCTS

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A PROJECT SUBMITTED TO THE DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF BACHELOR OF SCIENCE (B.SC) HONOUR DEGREE IN SCIENCE LABORATORY TECHNOLOGY (CHEMICAL/PETROLEUM TECHNIQUES)

NOVEMBER, 2025.

CERTIFICATION

This is to certify that, this project work titled **“VALORIZATION OF FOOD WASTES FOR BIOETHANOL PRODUCTION AND OTHER VALUABLE PRODUCTS ”** was carried out by **Courage Okwukwe IKEHI** with Matriculation Number **LSC2007305**, under the supervision of **Prof. J. O. Osarumwense** of the Department of Science Laboratory Technology (Chemical/Petroleum Techniques), University of Benin, Benin City, Edo state, Nigeria

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DEDICATION

This project is dedicated to Almighty God for His love and guidance all through the period of my study and for making this project successful.

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I wish to express my profound gratitude to Almighty God who gave me wisdom, knowledge, strength and provided the needed resources for the completion of my study in the University.

My sincere appreciation goes to my supervisor, Prof. J. O. Osarumwense for his useful assistance, supervision, constructive criticism and encouragement, which contributed immensely to the successful completion of this work. May the Almighty God bless him.

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ABSTRACT

This study focused on the valorization of food wastes, particularly banana and pineapple peels, for the production of bioethanol and other valuable products using *Aspergillus Niger*. The fruit wastes were collected, cleaned, oven-dried, pulverized, and subjected to fermentation for 21 days a. Proximate and physicochemical analysis revealed high carbohydrate content (17.50–21.29%) and moderate protein and ash levels, indicating their suitability as substrates for microbial fermentation. During fermentation, there was a gradual decline in pH and reducing sugar concentration, confirming active microbial metabolism and sugar utilization. The mixed banana–pineapple substrate yielded the highest ethanol concentration (7.60 mL/100 mL), followed by banana (7.20 mL/100 mL) and pineapple (6.80 mL/100 mL). In addition to ethanol, *A. Niger* produced significant quantities of citric acid (4.30 mg/mL), gluconic acid (2.60 mg/mL), and carbon dioxide (4.50 g/L), reflecting multiproduct valorization. These findings demonstrate that fruit wastes can serve as low-cost, renewable feedstocks for sustainable bioethanol and organic acid production. The research underscores the potential of waste-to-wealth conversion as an effective strategy for environmental protection, renewable energy generation, and the promotion of a circular bio economy through sustainable food waste management.

CHAPTER ONE

INTRODUCTION

1.1 Background of Study

Every year, about 1.3 billion tons of food, nearly one-third of all food produced are wasted worldwide (FAO, 2013). In addition to making food insecurity worse, this also increases greenhouse gas emissions and degrades the environment. Research on biofuels, especially bioethanol, as fossil fuel substitutes has increased concurrently with the rising demand for renewable energy (Nigam and Singh, 2011). Sustainable solutions to the energy and environmental problems can be achieved by valuing food waste and turning it into bioethanol and other products with added value. Food wastes are good substrates for the fermentation of ethanol because they are high in carbohydrates. Fruit and vegetable peels, starchy residues, bakery waste, and dairy by-products are examples of common sources (Sadh *et al.,* 2018). Fermentable sugars are abundant in fruit peels, including those from oranges, bananas, and pineapples. While lignocellulosic food residues need to be pretreated in order to release fermentable sugars, starchy wastes, such as the peels from potatoes and yams, can hydrolyze into simple sugars (Kiran *et al.,* 2014). Fruit wastes are low in lignin, which is primarily positioned loosely between cellulose and hemicelluloses, and high in cellulose and hemicellulose. Because of this, these wastes are intriguing for the production of bioethanol. Since lignocellulosic raw materials are regarded as renewable energy sources, using them to produce bioethanol could also help reduce

CO2. The FAO estimates that 40–50% of the world's food waste originates from fruits, vegetables, and root crops. In Asia, 37% of all agricultural waste is made up of fruit and vegetable waste alone. It is estimated that between 35 and 40 percent of fruits and vegetables are wasted each year. There is a problem with managing the large amount of fruit waste that is produced even after consumption, storage, and industrial processing. Although three classes of carbohydrates which are simple sugar, starch, and lignocellulose material can be used to produce bioethanol, starch that is, grains like corn or sorghum or tubers like cassava is currently the source of 95% of ethanol produced. Because it affects the cost of food and feed, the use of this kind of biomass has become more and more controversial (Sanchez and Cardona, 2008)). The first ethanol was produced in nations like the United States and Brazil, and it has been around for a very long time (Jackson, 2008). A total of 51,000 million liters of ethanol can now be produced (RFA 2007). The conversion of biomass into fuel ethanol has received attention lately because it is predicted that fossil fuels will run out within the next few decades. Many agricultural raw materials rich in fermentable carbohydrates have been tested globally for bioconversion from sugar to ethanol, but the cost of carbohydrate raw materials has been a limiting factor for large-scale production by the industries employing fermentation processes (Campo *et al.,* 2006). Research efforts are concentrated on designing and improving a process that would produce a sustainable transportation fuel using low-cost feed stock. In the current energy crisis, the only way to meet the high demand for ethanol is to use efficient fermentative microorganisms to produce it from relatively cheaper sources of raw materials

(Pramanik and Rao, 2005).

The overuse of fossil fuels has caused climate change and global warming. Consequently, efforts are being made to switch from fossil fuels to cleaner, renewable fuels like bioethanol and biodiesel. Furthermore, the development of an alternative renewable energy source, such as bioethanol, is urgently needed for the country's energy security because of the conventional fossil fuels' rapid consumption and their unpredictable price fluctuations. Bioethanol can be made from lignocellulosic raw materials. Natural sugars can be found in abundance in fruit waste. Around the world, a lot of fruits are eaten as functional foods and even as health supplements. Two important fruits that are prone to spoiling during harvest, storage, marketing, or processing are pineapple and banana, which leads to waste. It is estimated that 20 to 30 percent of production is lost due to agricultural waste. Out of the 16.8 million tons of fruit produced, the Food and Agriculture Organization (FAO) estimated that 3.36 million tons of waste were produced. Microorganisms may use the solid waste produced by the fruit processing industries as raw materials to produce secondary metabolites with industrial value. The main by-products of processing different fruits are their peels, which have been found to be a good source of different bioactive compounds with a variety of positive effects. However, the processing industries throw away large amounts of fruit peels as waste, which is a serious environmental issue (Zang *et al.,* 2005). In addition to being a viable option for getting rid of the polluting residues, these fruit processing wastes may be utilized as a feedstock for the production of bioethanol (Wyman, 2011). There are a lot of nutrients and organic matter in food waste. Food waste is therefore a perfect raw material for biofuel production. There is a wide range of uses for biomass, such as the biorefinery sector, which produces pulp and lignosulfonates and other biomass-based products. The production of liquid fuels is one possible use, and the fermentation of sugars to ethanol has long been regarded as one of the most significant processes. Alternative energy sources are becoming more and more popular worldwide as a result of the world's energy supply rapidly running out (Lin and Tanaka, 2006).

One of the main causes of the rise in environmental issues is the combustion of fossil fuels derived from petroleum in machinery and automobiles, which also produces harmful gases. This necessitates the use of an alternative fuel source, which can be achieved by emphasizing greener technologies in the production of bio-fuel from a variety of renewable resources. One of the most notable aspects of the 20th century that has significantly transitioned into the 21st is the production of bio-fuel for both internal and external uses after biotechnology advancements (Mushimiyimana and Tallapragada 2015). In contrast to fossil fuels, bio-ethanol is non-toxic, biodegradable, and does not pollute the environment (Hossain *et al.,* 2010). Following fermentation technology, carbon-based feedstock is converted to bio-ethanol. Because agricultural waste feedstocks use photosynthesis to obtain energy from the sun, they are regarded as renewable components (Hossain *et al.,* 2011). Concern over climate change and the potential of biofuels to lower greenhouse gas (GHG) emissions are two of the main factors propelling the global promotion of bioethanol (Micic and Jotanovic, 2015). *Saccharomyces cerevisiae* and *Aspergillus Niger* can be used to produce bioethanol in greater quantities. The best source of microorganisms for fermentation research in the extraction of bioethanol from pineapple wastes are these two. The fermentation process using these microorganisms has improved as a result of recent developments in the field of microbiology.

The idea of valorization, or turning waste materials into useful products, has drawn interest recently as a sustainable waste management technique. Food waste has a lot of potential as a feedstock for bio-based products, especially bioethanol, because it is high in organic matter, carbohydrates, and nutrients (Kiran *et al.,* 2014). One renewable biofuel that can lessen reliance on fossil fuels and slow down global warming is bioethanol. Other value-added products like biogas, animal feed, compost, and bioplastics can be produced from food waste in addition to bioethanol (Mohapatra *et al.,* 2010). With an emphasis on the procedures, difficulties, and advantages of such methods, this study investigates the possibility of using food waste to produce bioethanol and other valuable products.

1.2 Aim and Objectives

The primary aim of this study is to assess the valorization potential of food wastes for bioethanol production and other valuable products.

The specific objectives of this study are to:

1: To examine the types of food wastes suitable for bioethanol production.

2: To evaluate the processes involved in converting food wastes into bioethanol.

3: To identify other valuable products that can be derived from food wastes.

4: To highlight the challenges and benefits associated with food waste valorization.

1.3 Significance of the Study

This study provides insights into sustainable waste management and renewable energy production. By demonstrating the potential of food wastes as a resource, it contributes to environmental conservation, energy security, and economic development. Policymakers, researchers, and industries can benefit from the findings to develop strategies for implementing food waste valorization projects.

1.4 Problem statement

The increasing generation of food wastes and inadequate waste management practices have led to environmental degradation and economic losses. At the same time, the global demand for renewable energy and sustainable materials is rising. However, the potential of food wastes as a resource for producing bioethanol and other products remains underutilized in many regions. This gap necessitates research into effective valorization techniques that can help address energy needs, reduce waste, and promote a circular economy.

CHAPTER TWO

LITERATURE REVIEW

2.1 BIOFUEL – AN INVESTMENT FOR THE FUTURE

Global energy demand and greenhouse gas (GHG) emissions are rising annually as a result of population growth, which also raises the need for food and energy. The transportation sector is the most energy-intensive industry that emits and will continue to emit significant amounts of greenhouse gases. (Intergovernmental Panel on Climate Change, 2012). This implies that in order to improve the environment and secure a brighter future for our world, GHG emissions must be effectively decreased in this area. Carbon dioxide (CO_2), methane (CH_4), and water vapor (H_2O) are the most prevalent greenhouse gases (Center for Climate and Energy Solutions, 2017). For a number of reasons, the alternative energy source must be sustainable and renewable. First, to reduce greenhouse gas emissions. Second, in order to prevent the energy source from competing with the production of food and arable land (Joelsson, 2015).

Biomass-based transportation fuels are an alternative to fossil fuels. According to photosynthesis, the living plant uses CO_2 throughout its existence. However, when plants are used to produce biofuel, the yeast's cellular respiration releases CO_2. This is an illustration of the carbon cycle, which, in a perfect world, would result in the release of zero CO2 (Erdei, 2013). To guarantee that energy and climate goals are fulfilled by 2020, the European Union has enacted legally binding laws. The three main goals are to reduce greenhouse gas emissions by 20%, increase energy efficiency by 20%, and have 20% of the energy in the EU come from renewable sources. Transportation, waste management, housing, and agriculture are all included in the national emission reduction targets (European Commission, 2017). Sweden has set a goal that by the year 2050, a more sustainable and resource-efficient energy supply without generating any net GHG emission needs to be achieved (SOU, 2013).

2.2 CONCEPT OF FOOD WASTE

Food waste is the loss or disposal of edible food at different points in the food supply chain, such as during production, processing, distribution, and consumption. It is a complicated and multidimensional problem (Gustavsson *et al.,* 2011). Similarly, food waste is the term used to describe the perishable and biodegradable waste generated by food processing, consumption, and cleaning activities in various contexts (Zhang 2017). High concentrations of starch, cellulose, hemicellulose, sugars, proteins, and lipids are commonly found in food waste (Wang, 2005). These components make food waste a suitable substrate for microbial fermentation, offering opportunities for valorization and resource recovery.

A significant portion of household waste is made up of food waste, which also has a higher potential to seriously pollute the environment. Food waste can easily harbor a lot of microorganisms and pathogens due to its high water content, organic matter, and microelements. Food waste has a low calorific value, which indicates that it releases little heat when it burns. Calorific value is frequently used to quantify or convey the worth and potential of trash as fuel. Food waste has a low calorific value per unit mass of roughly 2100–3100 kJ/kg due to its extremely high moisture content. Waste heat can be utilized when the average low calorific value of the waste exceeds 3350 kJ/kg; otherwise, no combustion-supporting agent is added during the incineration process. Coal or fuel oil must be added to power generation in order to facilitate combustion, which raises costs. Simultaneously, temperature control is required to stop the production of dust and other dangerous materials (Zhang *et al.,* 2013).

Food waste has a high utilization value despite being extremely detrimental to both humans and the environment. Food waste has a high nutritional element content, with protein and oil being the primary constituents. Dry materials that can be used to make biodiesel contain roughly 30% crude fat and 20% crude protein. Composting solid food waste materials can create organic fertilizer, which can be applied as fertilizer and for crop growth. Unfortunately, in certain nations, trash is not sorted and is instead mixed together, which wastes a lot of nutrients (Gao 2019).

Food waste sources can be divided into three categories: the first is food products lost in the production phase, the second is the inevitable waste of food, and the third is avoidable food waste. According to each of the 4 stages of the food supply chain, Gustavsson divides food waste into five sources: agricultural production, postharvest handling and storage, processing and packaging, distribution consumption (Jenny and Christel 2011). Large amounts of perishable materials found in food waste make it unsuitable for the current garbage collector and transportation vehicles. Food waste is extremely difficult to transport in sealed bottles because of its high water content. Waste liquid will spill out of the bottles if you are a little careless. Using a sealed tank car to load, collect, and unload garbage will be extremely challenging. If the road in the transportation project is not smooth, the waste liquid may leak out (Bian and Chen 2018). New secondary pollution will be created as the waste liquid seeps into the surrounding soil and contaminates the groundwater and nearby water source. Because food waste is perishable, it will release an unpleasant and repulsive gas that not only contaminates the air around it but also harbors dangerous bacteria, draws pests like cockroaches, flies, and mosquitoes, and spreads a number of diseases. Furthermore, environmental protection is neglected in some places. Food scraps are frequently used as animal feed. Through livestock, certain bacteria and heavy metals in waste can be harmful to human health (Bian and Chen 2018).

2.2.2. Methods of Conversion of Food Waste

Biomass can be converted into biofuels through mechanical, thermochemical, and biological processes

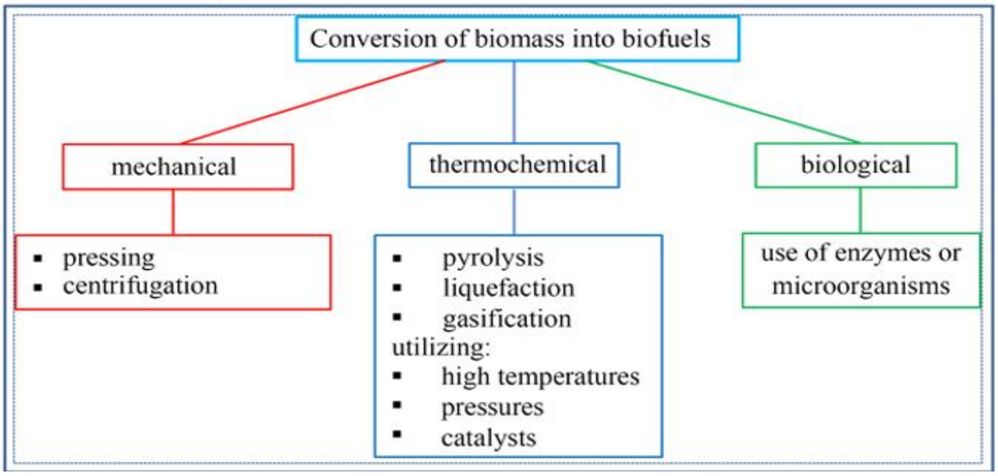

Figure 2.1: Schematic representation of various methods for converting biomass into biofuels.

Pressing and centrifugation are two of the main mechanical methods used to extract bioactive compounds, including biofuels, from materials like algae and oilseed crops. Pyrolysis, liquefaction, and gasification are examples of thermochemical conversion processes that use high temperatures, pressures, and catalysts to convert biomass into different types of solid, liquid, and gaseous biofuels. In order to create bio char and bio-oil, which can then be further processed into transportation fuels, pyrolysis heats biomass in an oxygen-free environment. Biomass is converted by gasification into synthesis gas, which can be utilized to create synthetic liquids and fuels like hydrogen. By breaking down organic compounds in biomass using catalysts and solvents, liquefaction produces bio-crude oil with fuel qualities akin to those of liquid transportation fuels. Biofuels and other biochemical products, including bioethanol, bio butanol, bio hydrogen, and bio methane, as well as organic acids like acetic, butyric, and lactic acids, can be produced through fermentation and anaerobic digestion using biological conversion, which uses enzymes or microorganisms to break down biomass components like cellulose and hemicellulose into simple sugars (pentoses and hexoses) (Nanda *et al.,* 2023).

2.3 CONCEPT OF VALORIZATION

Through waste reduction and resource efficiency, the concept of valorization embraces not only the economic aspect but also the environmental and social dimensions (Kumar *et al.,* 2020). The sustainable practice of turning excess, discarded, or spoiled food materials into valuable products, like bioethanol, as opposed to throwing them in landfills or incinerators is known as "valuation of food waste." This procedure aims to minimize waste and maximize the value of biomass resources, which is consistent with the circular economy's tenets (Kumar *et al.,* 2020). The processes of collection, pretreatment (mechanical, thermal, or enzymatic), hydrolysis, fermentation, and ethanol recovery are all part of the value-adding process. Every stage is designed to maximize the conversion of organic material in food waste into fermentable sugars, which microbes such as *Saccharomyces cerevisiae* use to make ethanol (Sadh *et al.,* 2018). Examples of food waste valorized into bioethanol are banana peels, bread waste, pineapple peels. Potato peels etc.

2.3.1. Food Waste Valorization

Animal feed, composting, incineration, waste conversion to energy (such as anaerobic digestion), and landfilling are examples of traditional waste management techniques for handling food waste. Problems with disposal techniques like landfilling and incineration have raised interest in developing innovative substitutes to lessen the harm these techniques do to the environment in recent years. Food wastes can be transformed into a wide range of products with added value, making them intriguing renewable resources. Waste valorization, the process of turning food waste into more valuable and useful products, is an alluring strategy. The practice of valuing food waste components has been around for a while and is typically connected to waste management procedures, but it is becoming more popular because it can significantly influence the development of economical and sustainable processes for creating high-value goods. Given the high demand for biofuels, enzymes, medications, solvents, and surfactants worldwide, waste valorization is especially crucial today. Many nations have developed plans for the construction of extensive facilities for turning various food waste streams into a range of valorized products as a result of this high demand (Snyder, 2015). For instance, by 2030, it is anticipated that materials from crop sources will account for about 25% of the chemical feedstock used in the United States (Sengupta and Pike, 2012). Present bioenergy studies have shown that anaerobic digestion can be used on a wide range of food and grain wastes to produce bioethanol, biodiesel and biogas.

2.4 VALORIZATION OF FOOD WASTES FOR SUSTAINABLE BIOETHANOL PRODUCTION

2.4.1 Overview of Biofuels

A biofuel is any fuel derived from forestry and agricultural products or their byproducts, household and industrial waste, and other biological organisms and their metabolic waste. Biomass is a ubiquitous resource that is used to make biofuels. Thus, the advancement of biofuels is important. Liquid biofuels like bioethanol, methanol, and biodiesel are typically referred to as "biofuels." because products made from liquid biofuel are thought to be the greatest supplement or substitute for petrochemicals and gasoline. Due to growing concerns about energy security and environmental pollution, the development of biofuels has attracted worldwide attention (Moioli *et al.,* 2018).

While fossil fuel resources are relatively limited, the primary goal of biofuels is to create new fuels to mitigate the energy crisis. The conflict between oil supply and demand has grown to be a significant issue in the evolution of modern society. Furthermore, biofuels are a clean energy source that can enhance the atmosphere and lessen pollution. Car exhaust contributes significantly to the pollution of urban air. In addition to resolving these issues, using fuel alcohol has several advantages for the car itself. Nevertheless, there are still certain issues with the way biofuels are produced. Biofuels have a single raw material structure and a limited supply. Although some crops are used as biofuel raw materials, there won't be enough food available if these crops are used as biofuel raw materials. This issue can be effectively resolved by using food waste. The second issue is that production costs are prohibitively high, although they will continue to decline as technology advances (Moioli *et al.,* 2018). The most promising alternative fuels are now biofuels, with large-scale development of fuel ethanol and biodiesel technologies among them. In 2017, 79.81 million tons of biofuel ethanol were produced worldwide, with the United States and Brazil producing the most fuel ethanol (44.1 million tons and 21.28 million tons, respectively) (Ma *et al.,* 2019). Due to the current global resource scarcity, high energy demands, and severe environmental pollution, many waste treatment techniques are unable to meet the demands of resource and emission reduction.

2.4.2 Introduction to Bioethanol

Bioethanol is extremely adaptable and has a wide range of uses. For instance, it can be utilized as a chemical that serves as a precursor for numerous reactions, as an additive in gasoline, or directly as fuel (Makkee *et al.,* 2013). One popular biofuel is bioethanol. Bioenergy, which is bio liquid fuel made by processing and converting biomass raw materials, includes bioethanol. Bioethanol is safe for the environment and free of sulfur. Compared to comparable gasoline-powered vehicles, pure ethanol vehicles emit significantly less carbon (CO_2) (Mannberg *et al.,* 2014). Bioethanol is a renewable energy source at the same time. The world's fossil fuel supplies are currently running low, pressure is mounting to reduce carbon emissions worldwide, and the situation regarding oil security is getting worse. For nations to successfully make the switch from conventional fuel to clean, low-carbon fuel, bioethanol emerges as the top option. The production of ethanol for biofuels has expanded globally in the twenty-first century Currently, the United States, Brazil, the European Union, China, and Canada are the largest nations and regions in the world that support the use of biofuel ethanol. Brazil and the United States have the biggest biofuel ethanol industries among them (Adewuyi 2020).

2.4.3 Bioethanol Production from Food Waste

There are primarily two methods for producing ethanol. Thermochemical production is one possibility. The feedstock is then gasified to create syngas as part of the process steps. In this process, the material reacts with oxygen and steam at temperatures higher than 700°C. A combination of carbon monoxide, hydrogen, carbon dioxide, methane, and nitrogen make up the final product. Later, tar and ash are eliminated from the syngas through purification (Dwidevi *et al.,* 2009). Either adjusting the gasifier's parameters or cleaning the gasifier with hot gas can accomplish this. Filters, revolving particle separators, or chemical techniques like thermal or catalytic cracking can all be used to clean hot gases. Finally, catalytic synthesis or fermentation can be used to produce ethanol (Makkee *et al.,* 2013).

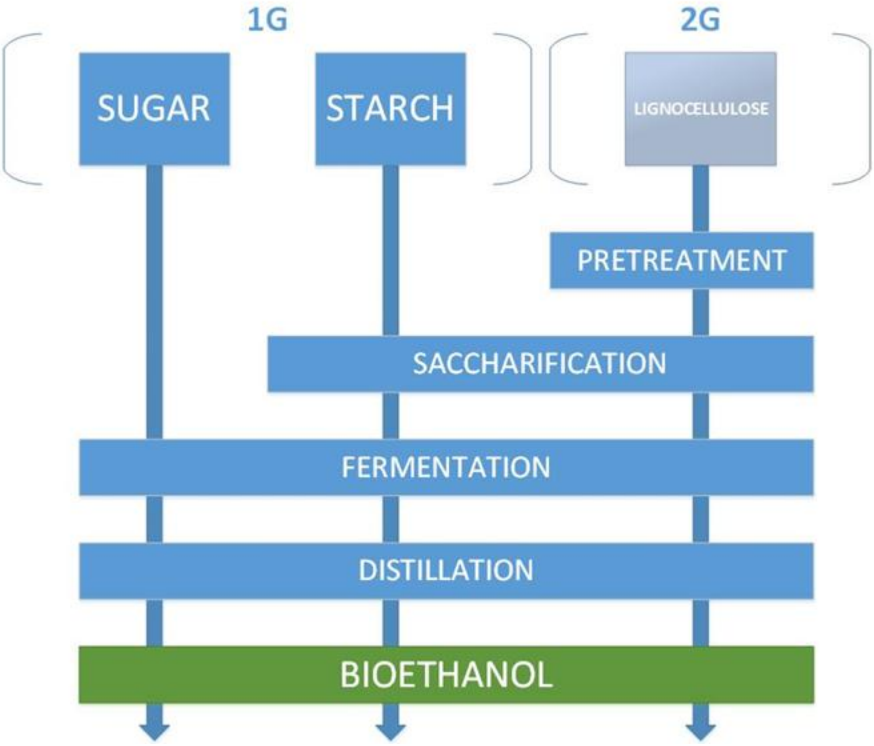

Figure 2.2: Process pathways for 1G and 2G ethanol production respectively

Refined ethanol can be added to fuel oil as fuel ethanol, which is of great significance to alleviate the energy crisis. Figure 2.3s is the flow chart of ethanol production from food waste (Wang 2015). Food waste is first divided into liquid and solid categories. The liquid is then separated into wastewater and lipid. After a solid component is crushed, it can be converted into ethanol in two different ways. The first technique involves hydrolyzing the solid ingredients in a low-acid environment to bring the pH down to roughly 5.5, after which yeast powder is added to start the fermentation process. The second method involves adding yeast powder to ferment after sterilizing solids in a microwave reactor to control ph. Liquid separation is used to recover the oil layer following fermentation. Crude ethanol is obtained by distilling the fermentation broth.

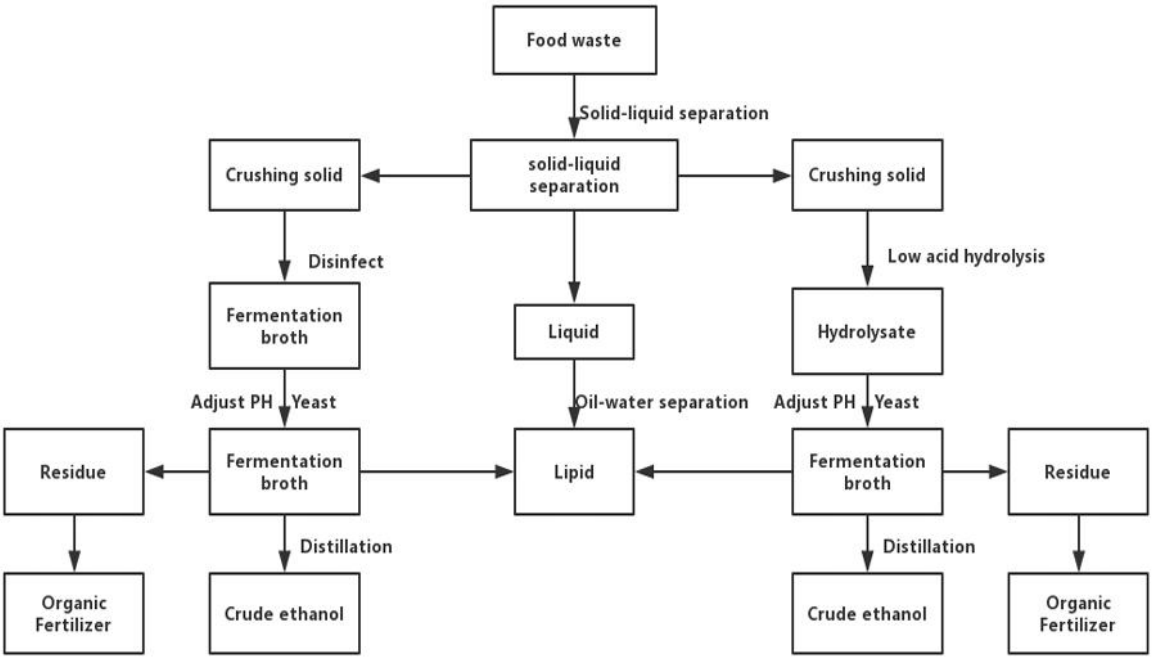

Figure 2.3: The process of preparation of ethanol from food waste.

Source: Wang 2015

The industrial synthesis of ethanol is mainly ethylene direct water method, which is divided into liquid phase method and gas-phase method. At present, most of ethanol is produced by gas phase method. The principle is that ethylene gas reacts with water under the action of phosphoric acid and solid catalyst.

The reaction equation is shown in the (1). (Guo 2015)

$$CH2 = CH2 + H2O \rightarrow CH3CH2OH \qquad \text{(Phosphoric acid)} \qquad (1)$$

The three primary methods of extracting sugar from biomass are concentrated acid hydrolysis, diluted acid hydrolysis, and enzymatic hydrolysis. Biomass waste contains a complex mixture of carbohydrate polymers from cellulose, hemicellulose, and lignin from plant cell walls. To produce sugar from biomass, the biomass is pretreated with acid or enzyme to reduce the size of raw material and decompose the raw material preliminarily. A portion of cellulose and hemicellulose are hydrolyzed to sucrose by enzymes or diluted acid, and fermentation produces ethanol (Chakraborty *et al.,* 2016)

Using sugar as a starting material and yeast's anaerobic respiration to turn it into ethanol is the fermentation process. Bioethanol is made from food waste using this process. There is a lot of sugar, starch, and fiber in food waste. There are many different reasons why food is wasted. First, food waste is separated into liquid and solid, and the solid is either hydrolyzed or disinfected after fermentation. Following that, either in an acidic environment or with the aid of an enzyme, starch and cellulose were hydrolyzed to glucose. Following sterilization, bioethanol underwent additional fermentation to yield bioethanol (Guo 2015).

2.4.4 Necessity to Increase the Scale of Bioethanol Production

The advantages of bioethanol over traditional fuels are substantial. It is safer to use and has a higher octane rating. Its clean and appropriate burning quality will enhance the quality of the air. These national policies outline how various nations plan to blend 20–30% ethanol into gasoline by 2030. However, there are still issues with ethanol availability. The global consumption of oil is actually much higher, at 95 million barrels per day. It is suggested that renewable biofuels like bioethanol and biodiesel be used in place of oil. But, given the availability of feedstock's at such large scales, producing these fuels from crops like oil seeds, wheat or rice starch, sugar cane, etc., at such a scale would present additional difficulties. In addition, the utilization of these traditional feedstock's may result in disputes between food and fuel. Utilizing forest residues, MSW, agro residues, etc., would likewise only produce a small quantity of bioethanol. Therefore, there is a need to explore the use of other wastes such as, fruit wastes or vegetable wastes which are consumed at huge scales. In fact, fruits are rich in sugars therefore fruit wastes could be a good source of fermentable sugars and bioethanol.

Every fruit generates 50% of its weight as a waste after its consumption, which is a huge amount and its utilization to generate bioethanol would help in not only solving the problem of energy security but this may also help in solving the problem of waste management (Hu *et al.,* 2011)

2.5 BENEFITS AND APPLICATIONS OF BIOETHANOL UTILIZATION IN ENERGY AND INDUSTRY

2.5.1 Benefits of bioethanol

Compared to traditional fuels, bioethanol offers numerous benefits. It is made from renewable resources and absorbs CO2 during crop growth, so virtually no new CO2 is released into the atmosphere. Bioethanol is therefore a new energy source that is good for the environment. Vehicle emissions during transportation contribute significantly to the atmosphere's concentration of greenhouse gases. These days, greenhouse gas emissions can be significantly decreased by using bioethanol.

In addition to being less harmful than fossil fuels, bioethanol is biodegradable. Third, the use of gasoline blends and bioethanol can lower oil consumption worldwide, improving fuel safety and preventing reliance on foreign fuel supplies. The increased demand for vital crops for the production of bioethanol will also help rural areas (Małgorzata *et al.,* 2019).

Air quality is enhanced by bioethanol's ability to lower carbon monoxide emissions from used automobile engines. The ease with which bioethanol can be integrated into the current fuel system for road transportation is another significant benefit. Adding bioethanol to gasoline is an efficient way to raise its octane number. It is simple to combine bioethanol with traditional fuels without modifying the engine.

2.5.2 Applications of Bioethanol

It is possible to use bioethanol in place of gasoline engines. Almost any percentage of gasoline can be mixed with it. Up to 15% bioethanol and oil are combined in the majority of gasoline engines currently in use. Compared with gasoline without ethanol, bioethanol has a higher octane number, which can improve the compression ratio of the engine and improve the thermal efficiency. Additionally, it serves as fuel for fireplaces that burn bioethanol. It is ideal for residential use because it emits no smoke and doesn't require a chimney. Additional significant uses of bioethanol include as a fuel for thermal combustion power generation, a fuel for cogeneration systems, a raw material for the chemical industry, and a fuel for cells via thermochemical reactions (Shi *et al.,* 2017).

2.8 PROCESSES OF BIOETHANOL PRODUCTION

1. Pretreatment

The complex chemical structure of lignocellulosic biomass necessitates pretreating the material to increase its digestibility (Galbe and Zacchi, 2012) (Hendriks and Zeeman, 2009). Pretreatment is required for the production of bioethanol from biomass because it is preferable to obtain a high yield of released sugars during the hydrolysis step, which will increase the production rate. Put another way, polymeric sugar chains must be readily available for microbial or enzymatic degradation of biomass to proceed at a high enough rate. Biological, physical, and chemical pretreatment are alternatives to conventional pretreatment techniques. An overview of degradation of biomass is presented in figure 2.

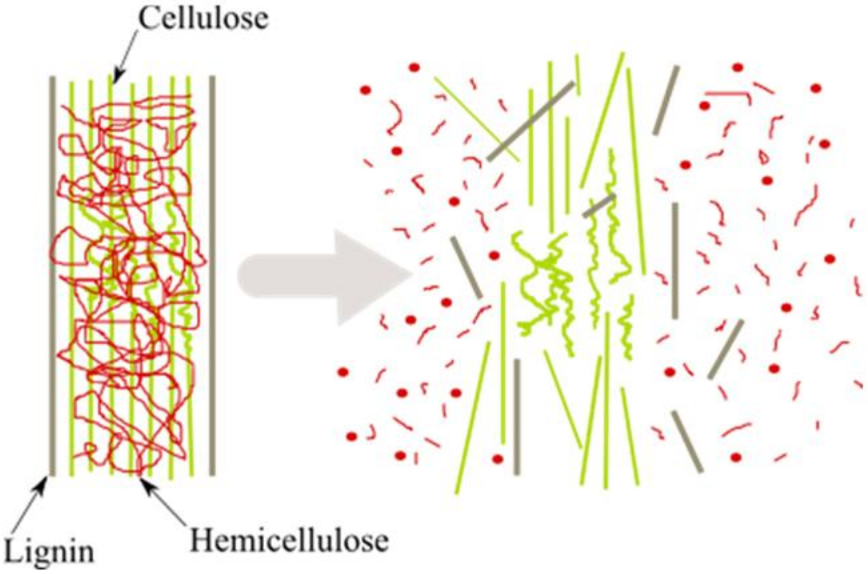

Figure 2.4: Overview of the degradation of biomass after steam pretreatment.

Source: Bondesson, 2016

2. Enzymatic Hydrolysis

Before fermentation, a significant amount of undigested polysaccharides from the pretreated biomass must break down into monomeric sugars. This is accomplished through a process step known as enzymatic hydrolysis, which uses various enzymes. The cellulose is broken down into oligosaccharides by the enzyme group cellulase, and the oligosaccharides are then hydrolyzed into monomeric sugars by the enzyme βglucosidase. When water and the right enzymes are added to the polysaccharide, the hydrolysis process is completed.

$$[C_5H_{10}O]_n + H_2O \xrightarrow{Cellulase} [C_5H_{10}O]_{n-2} + C_{12}H_{22}O_{11}$$

$$C_{12}H_{22}O_{11} + H_2O \xrightarrow{\beta-glucosidase} 2\ C_6H_{12}O_6$$

However, β-glucosidase is product inhibited, thus, when too much glucose is formed, the glucose formation will be affected negatively (Galbe and Zacchi, 2002). To break down the polymeric sugar molecules, lignocellulosic material's complex chemical makeup necessitates the use of multiple enzymes. Enzymatic hydrolysis, on the other hand, is a comparatively costly procedure. Acidic hydrolysis can therefore be an alternative because of its lower cost and greater efficacy; however, the equipment may be too expensive due to the acid's corrosive action. Furthermore, a problem is the development of toxic substances or inhibitors (Ismail *et al.,* 2012).

3. Fermentation

Hydrolyzed biomass sugars in the liquid phase can be fermented to produce ethanol using bacteria or yeast. In addition to lignin and its derivatives, the hydrolyzed lignocellulosic material contains inhibitory compounds and the sugars pentose (C5) and hexose (C6). When the hydrolysate ferments, the primary sugars that are produced are glucose (C6) and xylose (C5). A common ethanol-producing yeast, *Saccharomyces cerevisiae (S. cerevisiae*), can be used to ferment C6 sugars, but a genetically modified yeast, such as the strain *S. cerevisiae* KE6-12B, is needed to ferment both C5 and C6 sugars (Erdei, 2013; Bondesson, 2016). When lignocellulosic material is pretreated, *S. cerevisiae* can produce a variety of by-products and has a high tolerance to ethanol, making it suitable for both starch and lignocellulosic-based biomass. A good yeast should also be able to withstand low pH in addition to these characteristics. Contamination is less likely when the pH is lowered (Gírio, *et al.,* 2010). For fermentation with lignocellulosic material, the optimal operating temperature is approximately 30°C and a favorable pH is between 4.5 and 5.5 (Erdei, 2013). The maximum ethanol yield that can be achieved from fermentation is based on glucose to ethanol conversion. The chemical reaction for glucose to ethanol is as followed:

$$C_6H_{12}O_6\ (180.16\ g/mol) \rightarrow\ 2\ C_2H_5OH\ (46.07g/mol)\ +2\ CO_2\ (44.01g/mol).$$

The maximum theoretical ethanol yield, YT, that can be produced from glucose is determined from the following equation:

$$Y_T = \frac{2 \cdot 46.07}{180.16} = 0.51 \frac{g\ EtOH}{g\ glucose}$$

4. Distillation

Product recovery is required after ethanol fermentation because the product's concentration is low and byproducts must be eliminated. Distillation and solid-liquid separation are used in this step. Strippers and rectification columns are both used in the distillation process. To raise the ethanol concentration, the first column extracts the ethanol from the solid and non-volatile substances and eliminates water. In the rectification column, the ethanol is subsequently concentrated near the azeotropic point. The total cost of production is significantly impacted by the recovery of ethanol (Galbe, *et al.,* 2011)

2.9 OTHER VALUABLE PRODUCTS FROM FOOD WASTE

Besides bioethanol, food waste can be valorized into a range of other valuable products:

1: Biogas (methane) by anaerobic digestion of organic residues producing heat and power

2: Citric acid from microbial fermentation of food wastes such as fruit peels (e.g. Orange, Pineapple, or Banana Peels).

3: Organic acids, enzymes, pre-biotic compounds and bioactive compounds by fermentation of food and kitchen wastes

4: Thermal conversion products such as bio-oil, syngas, hydro char from pyrolysis/hydrothermal methods.

5: Animal feed, compost or fertilizer from processed residual biomass.

2.10 ENVIRONMENTAL AND ECONOMIC BENEFITS OF VALORIZATION OF FOOD WASTES

Valorization of food waste offers major benefits on both the environmental and economic fronts. In a circular-economy framework, it transforms what would be waste into useful resources, thereby reducing negative environmental impacts, recovering value, and creating economic opportunity.

2.10.1 Environmental Benefits

1. Waste diversion and reduction of greenhouse gas (GHG) emissions

When food waste is sent to landfill, it often decomposes anaerobically and emits methane a potent GHG. Proper valorization (e.g., thorough fermentation, anaerobic digestion or bio refining) helps divert organic material from landfill, thereby reducing methane and other GHGs. For example, one review indicates that improper management of food waste is associated with about 3.3 billion tonnes of CO_2-equivalent emissions annually. Another study emphasizes that using food waste as a feedstock recovers embedded nutrients and energy (which would otherwise be lost), thus improving resource efficiency.

2. Resource efficiency and reduction of virgin material use

Food waste valorization recovers the energy, water, land and nutrients already invested in producing the food. The use of this “waste biomass” means less need to extract or produce virgin feedstock’s for fuels, chemicals or materials. For instance, turning food waste into bio-based materials (bioplastics, biofertilisers) reduces reliance on petrochemical or mineral-fertilizer inputs. Furthermore, diverting food waste from disposal also reduces problems such as landfill leachate, soil and water contamination, and inefficient land use.

3. Promotion of a circular economy and sustainability goals

Valorization supports a shift from a linear “take-make-dispose” model to one where waste becomes feedstock for new products. This aligns with global sustainability agendas (e.g., Food and Agriculture Organization (FAO)’s emphasis and the UN Sustainable Development Goals) (FAO 2019). By establishing integrated bio refinery models, food-waste valorization facilities can maximize resource recovery, reduce emissions and create diverse value streams rather than a single-product output.

2.10.2 Economic Benefits

1. Creation of additional revenue streams

Food waste, once regarded purely as a disposal cost, becomes a feedstock for valuable products such as bioethanol, biogas, bio-fertilizers, bioplastics, feed ingredients or extraction of high-value bioactive compounds. This opens up new markets and business models.

2. Reduction in waste disposal and treatment costs

Municipalities and industries spend significant sums on waste collection, transport, landfill tipping fees, and environmental mitigation. Valorizing food waste reduces the volume of waste requiring disposal and can reduce these costs. In addition, by converting a liability (waste) into an asset (feedstock), organizations can improve their bottom line and reduce risk associated with disposal.

3. Job creation and industrial development

Valorization processes (collection, sorting, pretreatment, conversion, downstream product recovery) require labor, infrastructure and logistics, thereby creating employment opportunities. Over time, this can stimulate rural or urban economies, especially in waste-rich regions. The establishment of bio refineries or modular waste-conversion plants can attract investment and foster innovation in local communities.

4. Energy security and fossil fuel substitution

By converting food waste into biofuels (e.g., bioethanol) the dependency on imported or fossil-based fuels is reduced, improving energy security and potentially stabilizing energy costs. This substitution also has secondary economic benefits: less exposure to fossil-fuel price volatility, improved trade balances, and alignment with decarbonisation incentives (tax credits, carbon markets).

CHAPTER THREE

MATERIALS AND METHODS

3.1 MATERIALS

Table3.1: List of Materials Used

MATERIALS	PURPOSE	SOURCES
Food wastes (e.g. pineapple peels and banana peels}	The primary substrate, rich in carbohydrates and sugars for bioethanol production	Ekosodin local fruit sellers, Benin city, Edo state
Distilled Water	Used to prepare media and solutions, ensuring purity and preventing contamination during bioethanol production	Department of Science Laboratory Technology, University of Benin
Aspergillus Niger	Used for hydrolysis and fermentation. It breaks downs complex sugars into fermentable sugars for bioethanol production.	Department of microbiology, University of Benin

3.2 Lists of Apparatus and Equipment Used

APPARTUS	USES
pH meter	Used to check the ph. of the fermentation media daily.
Rotary Evaporator	Used to separate and concentrate ethanol from the fermentation mixture by gentle heating and evaporation
Oven	Used to dry the food waste before use.
Measuring Cylinder	Used to measure volumes of liquids accurately.
Autoclave	Used to sterile media, equipment and substrates to eliminate contaminants before fermentation.
Weighing balance	Used to measure the Mass of food waste samples.
Blender	For blending the food waste
Fermenter	Serves as the vessel where fermentation takes place
Personal Protective Equipment (PPE)	Protect individuals from hazards in the workplace or laboratory.
Pipette	For transferring small amounts of liquids from one container to another.
Filter paper	To filter or remove solid particles from liquids
Spectrophotometer	For measuring the absorbance to estimate sugar concentration or cell growth during fermentation.

3.2 METHOD

3.2.1 Sample Collection and Preparation

Banana and pineapple peels were obtained from neighboring fruit sellers in ekosodin Edo State. They were brought to the laboratory where they were properly washed with water to remove dirt and unwanted materials. The peels of all Samples were individually cut into small pieces and were oven dried at 70°C until constant weight was achieved. The dried samples were ground into fine powder using a blender and stored in airtight containers**.**

3.2.2 Proximate Composition Analysis

The proximate composition of each sample was determined on a dry weight basis. Parameters analyzed included moisture content, ash content, crude protein, crude fat, crude fiber, and carbohydrate (by difference). These analysis helped to evaluate the nutritional composition and fermentable potential of the food wastes prior to fermentation.

3.2.3 Inoculum Preparation

The microorganism *Aspergillus Niger* was used for hydrolysis and fermentation, respectively. The fungal culture of was grown on potato dextrose agar and incubated for several days to obtain 1×10^6 spores/mL spores.

3.2.4 Fermentation Process

For each substrate (banana, pineapple, banana + pineapple, and control), 50 mL of substrate solution was prepared in 250 mL conical flasks. The concentration was adjusted to achieve 10 g/L total solids. Each flask was sterilized at 121°C for 15 minutes, cooled, and then inoculated with 5 mL of *A. Niger* suspension under aseptic conditions. Fermentation was carried out for 21 days at 30 ± 2°C under aerobic conditions, with gentle agitation every 24 hours to ensure homogeneity. Samples were withdrawn every 7 days (Day 0, 7, 14, and 21) for analysis. During this period, the fungi converted sugars into ethanol and carbon dioxide. Throughout the fermentation period, which spanned 21 days, various parameters were monitored. These included pH, reducing sugar concentration and cell biomass. These assessments were conducted at intervals of 7 days. All recorded data were documented for subsequent analysis.

Plate 3.1: Fermentation of banana and Pineapple Peels

3.2.5 Distillation and Ethanol Recovery

Bioethanol was recovered from the fermented broth through distillation at 78°C after 21 days. The ethanol produced was collected as the distillate. The distillation was carried out at a temperature close to 78°C, which is the boiling point of ethanol.

3.2.6 Analysis

All experiments were carried out in a single replicate and several parameters where assessed which are the determination of dried cell biomass, the measurement of the pH level of each medium, the calculation of the reducing sugar in each medium.

3.2.7 Determination of Ethanol Content

The ethanol content of the distillate was determined using a hydrometer and a spectrophotometer. The hydrometer measured the alcohol percentage based on density, while the spectrophotometer helped confirm ethanol concentration by measuring light absorbance**.**

3.2.8 Determination of Reducing Sugars using DNS method/assay

Dinitrosalicyclic acid(DNS) colours a solution on heating when reacting with a sugar. The colour might not be seen physically but with a spectrophotometer. To test for reducing sugars in the samples, 1ml of the sample,1ml of distilled water and 3ml of warm DNS solution were put into test tubes. The mixture was put into boiling water for 5mins at 100 degrees. The test tubes are then cooled in water. After cooling, 2ml of potassium sodium tartrate solution was added to the test tube to stop the reaction immediately. The mixture is then checked for reducing sugar using the spectrophotometer with a wavelength of 540nm.

CHAPTER FOUR

RESULT

Table 4.1 shows the proximate composition of food waste samples on a dry weight basis. The results revealed that banana waste recorded the highest moisture content (72.15 ± 0.42%), followed by banana + pineapple (70.40 ± 0.38%) and pineapple waste (68.74 ± 0.31%), while the control sample had the lowest moisture content (8.92 ± 0.15%). In contrast, carbohydrate content was highest in the control (88.26 ± 0.55%), followed by pineapple waste (21.29 ± 0.45%), banana + pineapple (19.41 ± 0.42%), and banana waste (17.50 ± 0.37%).

For crude protein, banana waste (4.26 ± 0.25%) had the highest value, followed closely by banana + pineapple (4.10 ± 0.24%), pineapple waste (3.85 ± 0.21%), while the control sample had the lowest protein content (0.96 ± 0.05%). The ash content was highest in pineapple waste (2.05 ± 0.09%), followed by banana + pineapple (1.96 ± 0.10%), banana waste (1.89 ± 0.12%), and least in the control (0.85 ± 0.03%).
Crude fiber values ranged from 2.75 ± 0.13% in pineapple waste to 0.58 ± 0.03% in the control, while crude fat was highest in banana waste (1.57 ± 0.08%) and lowest in the control (0.43 ± 0.02%).

Table 4.2 presents the physicochemical properties of the food waste samples before fermentation. The results show that pH values ranged from 4.55 ± 0.03 in pineapple waste to 6.80 ± 0.01 in the control. Banana waste (4.82 ± 0.02) had the highest pH among the fruit wastes, followed by banana + pineapple (4.68 ± 0.02).

Total solids (TS) were highest in banana waste (10.34 ± 0.24 g/L), followed by banana + pineapple (10.10 ± 0.30 g/L), pineapple waste (9.78 ± 0.27 g/L), and least in the control (8.00 ± 0.18 g/L). Total dissolved solids (TDS) followed a similar trend, with banana waste (612.5 ± 5.10 mg/L) recording the highest and the control (550.0 ± 5.20 mg/L) the lowest. Specific gravity values ranged narrowly between 1.020 ± 0.01 for banana waste and 1.010 ± 0.00 for the control.

Table 4.3 shows the changes in pH of the food waste samples during the 21-day fermentation period. The results revealed a steady decline in pH values across all samples as fermentation progressed. Banana waste, which initially had the highest pH (4.82 ± 0.02), decreased to 3.50 ± 0.02 by day 21. Similarly, banana + pineapple reduced from 4.68 ± 0.02 to 3.48 ± 0.02, and pineapple waste from 4.55 ± 0.03 to 3.45 ± 0.02. The control sample, however, maintained higher pH levels throughout, dropping from 6.80 ± 0.01 at day 0 to 5.50 ± 0.01 at day 21.

Table 4.4 presents the reducing sugar concentration of the food waste samples during fermentation. The results show that at day 0, the control had the highest reducing sugar content (10.00 ± 0.16 mg/mL), followed by pineapple waste (9.10 ± 0.15 mg/mL), banana + pineapple (8.70 ± 0.14 mg/mL), and banana waste (8.20 ± 0.12 mg/mL). By day 21, banana waste recorded the lowest residual sugar (2.80 ± 0.05 mg/mL), followed by banana + pineapple (2.90 ± 0.05 mg/mL), pineapple waste (3.00

± 0.06 mg/mL), while the control retained the highest unutilized sugar (7.10 ± 0.10 mg/mL).

Table 4.5 shows the changes in wet and dry cell biomass during the fermentation period. The results indicated a steady increase in microbial biomass with time. After 21 days, the banana + pineapple mixture exhibited the highest wet biomass (5.30 ± 0.13 g/L) and dry biomass (2.20 ± 0.06 g/L), followed by banana waste (5.10 ± 0.12 g/L; 2.10 ± 0.06 g/L), pineapple waste (4.70 ± 0.11 g/L; 1.95 ± 0.05 g/L), while the control had the lowest biomass (2.00 ± 0.05 g/L; 0.85 ± 0.02 g/L). The increase in biomass correlates with efficient microbial growth and substrate utilization, particularly in the mixed fruit substrate.

Table 4.6 presents the bioethanol yield obtained during fermentation. **The** banana + pineapple mixture produced the highest bioethanol yield throughout the study, reaching 7.60 ± 0.16 mL/100 mL at day 21. This was followed by banana waste (7.20 ± 0.15 mL/100 mL), pineapple waste (6.80 ± 0.14 mL/100 mL), and the control, which had the lowest yield (1.50 ± 0.05 mL/100 mL). Ethanol yield increased steadily from day 7 to day 21, corresponding with the observed reduction in sugar levels and increase in microbial biomass, confirming efficient sugar conversion by *Aspergillus Niger*.

Table 4.7 shows the comparative yield of valuable products produced by *Aspergillus Niger* after 21 days of fermentation. The results revealed that the banana + pineapple mixture recorded the highest yield for all metabolites measured: bioethanol (7.60 ±

0.16 mL/100 mL), citric acid (4.30 ± 0.13 mg/mL), gluconic acid (2.60 ± 0.08 mg/mL), and carbon dioxide (4.50 ± 0.13 g/L). This was followed by banana waste, with bioethanol (7.20 ± 0.15 mL/100 mL), citric acid (4.10 ± 0.12 mg/mL), gluconic acid (2.45 ± 0.07 mg/mL), and CO_2 (4.20 ± 0.12 g/L). Pineapple waste ranked third, while the control recorded the lowest values across all parameters, with bioethanol (1.50 ± 0.05 mL/100 mL), citric acid (0.95 ± 0.04 mg/mL), gluconic acid (0.70 ± 0.03 mg/mL), and CO_2 (1.10 ± 0.04 g/L).

Table 4.1: Proximate Composition of Food Waste Samples (% Dry Weight Basis)

Sample	**Moisture (%)**	**Ash (%)**	**Crude Protein (%)**	**Crude Fat (%)**	**Crude Fibre (%)**	**Carbohydrate (%)**
Banana Waste	72.15 ± 0.42	1.89 ± 0.12	4.26 ± 0.25	1.57 ± 0.08	2.63 ± 0.11	17.50 ± 0.37
Pineapple Waste	68.74 ± 0.31	2.05 ± 0.09	3.85 ± 0.21	1.32 ± 0.07	2.75 ± 0.13	21.29 ± 0.45
Banana + Pineapple	70.40 ± 0.38	1.96 ± 0.10	4.10 ± 0.24	1.45 ± 0.06	2.68 ± 0.10	19.41 ± 0.42
Control	8.92 ± 0.15	0.85 ± 0.03	0.96 ± 0.05	0.43 ± 0.02	0.58 ± 0.03	88.26 ± 0.55

Table 4.2: Physicochemical Properties of Food Waste Samples (Before Fermentation)

Sample	**pH**	**Total Solids (g/L)**	**Total Dissolved Solids (mg/L)**	**Specific Gravity**
Banana Waste	4.82 ± 0.02	10.34 ± 0.24	612.5 ± 5.10	1.020 ± 0.01
Pineapple Waste	4.55 ± 0.03	9.78 ± 0.27	587.3 ± 6.40	1.018 ± 0.01
Banana + Pineapple	4.68 ± 0.02	10.10 ± 0.30	605.2 ± 4.60	1.019 ± 0.01
Control	6.80 ± 0.01	8.00 ± 0.18	550.0 ± 5.20	1.010 ± 0.00

Table 4.3: Changes in pH During 21-Day Fermentation

Sample	**Day 0**	**Day 7**	**Day 14**	**Day 21**
Banana Waste	4.82 ± 0.02	4.10 ± 0.03	3.85 ± 0.04	3.50 ± 0.02
Pineapple Waste	4.55 ± 0.03	4.00 ± 0.04	3.72 ± 0.03	3.45 ± 0.02
Banana + Pineapple	4.68 ± 0.02	4.05 ± 0.02	3.80 ± 0.03	3.48 ± 0.02
Control	6.80 ± 0.01	6.10 ± 0.02	5.80 ± 0.02	5.50 ± 0.01

Table 4.4: Reducing Sugar Concentration (mg/mL) During Fermentation

Sample	Day 0	Day 7	Day 14	Day 21
Banana Waste	8.20 ± 0.12	6.45 ± 0.10	4.10 ± 0.08	2.80 ± 0.05
Pineapple Waste	9.10 ± 0.15	7.30 ± 0.12	5.20 ± 0.09	3.00 ± 0.06
Banana + Pineapple	8.70 ± 0.14	6.80 ± 0.10	4.80 ± 0.08	2.90 ± 0.05
Control	10.00 ± 0.16	9.00 ± 0.14	8.00 ± 0.12	7.10 ± 0.10

Table 4.5: Changes in Wet and Dry Cell Biomass During Fermentation

Sample	**Parameter**	**Day 7 (g/L)**	**Day 14 (g/L)**	**Day 21 (g/L)**
Banana Waste	Wet Weight	3.10 ± 0.08	4.25 ± 0.10	5.10 ± 0.12
	Dry Weight	1.25 ± 0.04	1.80 ± 0.05	2.10 ± 0.06
Pineapple Waste	Wet Weight	2.90 ± 0.07	3.90 ± 0.09	4.70 ± 0.11
	Dry Weight	1.10 ± 0.03	1.60 ± 0.04	1.95 ± 0.05
Banana + Pineapple	Wet Weight	3.40 ± 0.09	4.50 ± 0.11	5.30 ± 0.13
	Dry Weight	1.30 ± 0.04	1.90 ± 0.05	2.20 ± 0.06
Control	Wet Weight	1.50 ± 0.05	1.80 ± 0.06	2.00 ± 0.05
	Dry Weight	0.60 ± 0.02	0.70 ± 0.03	0.85 ± 0.02

Table 4.6: Bioethanol Yield During Fermentation

Sample	Day 7 (mL/100 mL)	Day 14 (mL/100 mL)	Day 21 (mL/100 mL)
Banana Waste	3.40 ± 0.10	5.80 ± 0.14	7.20 ± 0.15
Pineapple Waste	3.10 ± 0.09	5.40 ± 0.12	6.80 ± 0.14
Banana + Pineapple	3.60 ± 0.10	6.20 ± 0.15	7.60 ± 0.16
Control	1.00 ± 0.03	1.30 ± 0.05	1.50 ± 0.05

Table 4.7: Comparative Yield of Valuable Products Produced by *Aspergillus Niger* After 21 Days

Sample	Bioethanol (mL/100 mL)	Citric Acid (mg/mL)	Gluconic Acid (mg/mL)	CO_2 (g/L)
Banana Waste	7.20 ± 0.15	4.10 ± 0.12	2.45 ± 0.07	4.20 ± 0.12
Pineapple Waste	6.80 ± 0.14	3.85 ± 0.10	2.20 ± 0.06	3.90 ± 0.11
Banana + Pineapple	7.60 ± 0.16	4.30 ± 0.13	2.60 ± 0.08	4.50 ± 0.13
Control	1.50 ± 0.05	0.95 ± 0.04	0.70 ± 0.03	1.10 ± 0.04

CHAPTER FIVE

DISCUSSION, CONCLUSION AND RECOMMENDATION

5.1 DISCUSSION

Bioethanol, a renewable biofuel derived from the fermentation of sugars or starches, represents a sustainable alternative to fossil fuels, contributing to reduced greenhouse gas emissions and enhanced energy security (Von Blottnitz and Curran, 2007). Its production from lignocellulosic and food waste substrates has gained significant traction due to the abundance of agricultural and domestic residues, which otherwise pose environmental disposal challenges (Panahi *et al.,* 2022). Valorization of food wastes particularly fruit peels and by-products rich in fermentable carbohydrates offers a dual benefit: waste minimization and the generation of value-added products such as bioethanol, organic acids, and microbial biomass. This approach aligns with circular economy principles, transforming low-value waste streams into economically viable bio-based commodities (John *et al.,* 2019; Subramaniyan *et al.,* 2019; Singh *et al.,* 2025). This study seeks to evaluate the potential of banana waste, pineapple waste, and their mixture as substrates for bioethanol production and co-generation of valuable metabolites (citric acid, gluconic acid, and carbon dioxide) using *Aspergillus Niger*.

The proximate analysis revealed that banana waste had the highest moisture content (72.15%), followed closely by the mixed banana–pineapple sample (70.40%) and pineapple waste (68.74%). High moisture content is favorable for microbial growth and enzymatic hydrolysis, as it enhances substrate solubility and nutrient availability. The result aligns with findings by Singh *et al.* (2016) and John *et al.* (2019), who reported similar moisture characteristics in banana peels used for bioethanol production.

Carbohydrate content, which represents the fermentable sugar potential, was highest in pineapple waste (21.29%) compared to banana (17.50%), while the mixed substrate had 19.41%. This indicates that pineapple waste contained more polysaccharides (cellulose, hemicellulose, and residual sucrose) available for hydrolysis. These findings are consistent with Casabar *et al.* (2019), who emphasized pineapple peel as a carbohydrate-rich substrate suitable for fermentation.

Protein and ash contents were moderate in all fruit wastes, with banana waste showing the highest crude protein (4.26%) and pineapple the highest ash (2.05%). These components provide nitrogen and mineral nutrients necessary for fungal growth and enzyme synthesis. The relatively low fat and fibre contents observed in this study are advantageous, as excessive lipid or lignin fractions can inhibit saccharification and fermentation efficiency.

The initial physicochemical properties also supported fermentation potential. The pH values ranged from 4.55 to 4.82 among the fruit wastes, providing a slightly acidic environment optimal for *A. Niger* metabolism. According to Subramaniyan *et al.* (2019) and Hamdy (2013), acidic media (pH 4–5) enhance organic acid production by *A. Niger*. The total solids (9.78–10.34 g/L) and dissolved solids (587–612 mg/L) were indicative of sufficient organic matter, confirming the substrates' suitability for microbial conversion processes.

A gradual decline in pH was observed in all fermented samples, from an initial range of 4.55–4.82 to final values between 3.45 and 3.50 after 21 days. This acidification signifies active microbial metabolism and accumulation of acidic metabolites such as citric and gluconic acids. A similar trend was reported by Singh *et al.* (2025), who recorded a pH drop from 5.0 to 4.0 during banana peel fermentation using *A. Niger* and *S. cerevisiae*. The pH decline observed here aligns with Hamdy (2013), who demonstrated that *A. Niger* growth and acid production from orange peels were associated with decreasing pH due to organic acid secretion.

The consistent pH reduction across banana, pineapple, and their mixture reflects efficient sugar catabolism and organic acid synthesis. The control sample, which showed a lesser decline (6.80 to 5.50), further confirms that *A. Niger* activity was central to the fermentation process.

The concentration of reducing sugars decreased progressively across all samples, indicating successful sugar assimilation by *A. Niger*. The initial reducing sugar values ranged from 8.20 mg/mL (banana) to 9.10 mg/mL (pineapple) and declined to between 2.80 and 3.00 mg/mL after fermentation. This observation agrees with Casabar *et al.* (2019), who reported steady sugar depletion in pineapple peel fermentation, correlating with ethanol formation.

Similarly, John *et al*. (2019) found that sugar reduction was proportional to ethanol yield during banana peel fermentation, suggesting that carbohydrate hydrolysis and fermentation were synchronized. The efficiency of sugar utilization in the current study was highest in the mixed banana–pineapple substrate, implying that the combination provided balanced nutrients that enhanced enzymatic conversion.

Microbial growth, represented by wet and dry biomass, increased steadily throughout the fermentation period. The highest biomass accumulation was observed in the banana + pineapple mixture (2.20 g/L dry weight), followed by banana waste (2.10 g/L) and pineapple waste (1.95 g/L). The increase in biomass reflects the robust metabolic activity of *A. Niger*, which utilized the available nutrients efficiently.

This observation corroborates the findings of Singh *et al*. (2016), where co-cultured *A. Niger* and *S. cerevisiae* showed significant biomass accumulation due to simultaneous saccharification and fermentation. The mixed-substrate synergy observed here may be attributed to the combined nutritional and carbohydrate profile of the two wastes, fostering higher fungal proliferation.

The production of bioethanol increased progressively from day 7 to day 21, corresponding with the decline in sugar concentration. The highest yield was obtained from the banana + pineapple mixture (7.60 mL/100 mL or 7.6% v/v), followed by banana waste (7.20%) and pineapple waste (6.80%). The control sample showed minimal ethanol formation (1.50%). When compared with earlier studies, the yield from this study was superior. Benjamin *et al*. (2014) obtained a maximum of 6.287%

(v/v) bioethanol from banana peels using co-cultures of *A. Niger* and *S. cerevisiae*, while John *et al*. (2019) achieved 0.424% (v/v) using cellulase-mediated hydrolysis and yeast fermentation. The higher ethanol yield observed in this study can be attributed to prolonged fermentation time (21 days) and the efficiency of *A. Niger* in directly fermenting simple sugars and partially hydrolyzed polysaccharides without requiring yeast co-culture.

Similarly, Casabar *et al*. (2019) reported 5.98 g/L (0.598% v/v) ethanol from pineapple peel within 48 hours using *Trichoderma harzianum*, which is notably lower than the present yield. The lower values in these studies could be due to shorter fermentation durations, differences in substrate composition, and microbial species employed. The results of this thesis demonstrate that *A. Niger* is capable of efficient sugar conversion under acidic conditions and that mixed fruit substrates can enhance ethanol output through synergistic nutrient effects.

In addition to ethanol, *A. Niger* also produced significant amounts of citric acid (4.30 mg/mL) and gluconic acid (2.60 mg/mL), with the banana + pineapple mixture giving the highest yields. This simultaneous production of organic acids alongside ethanol underscores the organism’s versatile metabolic capacity. The results are comparable to those of Subramaniyan *et al*. (2019), who reported 6.2% citric acid yield from pineapple peel and 1.8% from banana peel under submerged fermentation. The relatively lower citric acid yield in the present study may be due to longer fermentation time and absence of methanol supplementation, which typically enhances acidogenesis. Nevertheless, the trend of higher citric acid yield in pineapple-rich substrates is consistent across both studies.

Kannaiyan (2019) similarly obtained 6.061% (w/w) citric acid from pineapple waste using SSF, reinforcing the fruit's suitability for acid production. The lower values here (3.85–4.30 mg/mL) are explained by the submerged fermentation method and natural substrate composition without sucrose enrichment. However, the simultaneous production of ethanol, citric, and gluconic acids in the current work represents a distinct advantage, achieving multi-product valorization in a single process. Furthermore, Hamdy (2013) demonstrated an exceptionally high citric acid yield (64%) using orange peel fortified with molasses. Though the yield far exceeded that in this study, the finding supports the potential of fruit waste as a rich substrate for *A. Niger*. The acidic shift (pH 4.68 to 3.48) in the present study mirrors Hamdy's conditions for citric acid accumulation, confirming that pH regulation is a critical factor for optimizing acid production.

The accumulation of CO_2, ranging from 3.90 to 4.50 g/L, paralleled ethanol production, as both are primary metabolic products of carbohydrate fermentation. The mixed substrate recorded the highest CO_2 value (4.50 g/L), indicating active decarboxylation processes. This aligns with general fermentation stoichiometry, where CO_2 evolution correlates directly with ethanol synthesis.

The outcomes of this research confirm that *Aspergillus Niger* can efficiently convert food wastes such as banana and pineapple into bioethanol and other value-added products. The high ethanol yield and multi-product formation from a single microbial strain and simple substrate combination demonstrate the feasibility of a low-cost, sustainable bioprocess. Moreover, the superior performance of the banana + pineapple mixture emphasizes the advantage of substrate co-fermentation in enhancing microbial metabolism and product yield.

These findings substantiate earlier reports by Benjamin *et al*. (2014), Subramaniyan *et al*. (2019), and Kannaiyan (2019), while extending them by integrating ethanol and organic acid production into one unified valorization scheme. Such integrated bioprocessing supports waste-to-wealth conversion models and contributes to global efforts toward renewable energy and sustainable resource management.

5.2 CONCLUSION

The findings of this study demonstrate that *Aspergillus Niger* effectively utilized banana and pineapple wastes, as well as their mixture, for the production of bioethanol and other valuable metabolites. The observed reduction in pH and sugar concentration, alongside increased biomass formation, indicates efficient fermentation and substrate conversion. Among all substrates, the mixed banana–pineapple waste yielded the highest levels of bioethanol, citric acid, gluconic acid, and carbon dioxide, confirming the synergistic advantage of mixed-substrate fermentation. These results highlight the potential of fruit wastes as inexpensive and sustainable feedstocks for bioethanol and organic acid production. This approach not only reduces environmental waste but also promotes resource recovery and supports the development of a circular bio economy.

5.3 RECOMMENDATIONS

1.Governments should promote policies supporting food waste collection and bioethanol production.

2. Invest in improved technologies to enhance ethanol yield from food wastes.

3. Create public awareness on the benefits of waste valorization.

4. Support small-scale and community-based bioethanol projects.

5.Conduct further research to optimize processes and evaluate environmental impacts.

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