PRODUCTION AND CHARACTERISATION OF BIODEGRADABLE BIOFILM FROM PLANTAIN PEEL, EGGSHELL AND CASSAVA STARCH

By

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CERTIFICATION

This is to certify that the thesis titled "**Production and characterisation of biofilm from plantain peel, eggshell and cassava starch**". By Okeyahweh, **Chinyere Simcha (20194272938)** meets the regulations governing the award of the degree of Doctor of Philosophy in Biochemistry, Federal University of Technology, Owerri, and has not been submitted to any university or other organization as a component of any exam, and all published materials utilized in my report have been properly acknowledged.

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DEDICATION

This research report is dedicated to God Almighty

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Figure **Page**

LIST OF PLATES

LIST OF APPENDICES

ABSTRACT

Biodegradable films are made from natural polymeric materials such as starch, vegetable oil, cellulose, lignin, and also materials derived from animals such as proteins and lipids. They are easily degraded by microbes, and the degradation process does not take a long time. The present study investigated the use of ripe plantain peels as a source material for producing biodegradable biofilms; using powdered ripe plantain peels, eggshell powder, acetic acid – vinegar - and cassava starch produced in the study. The biofilms synthesized were plantain peel-based biodegradable biofilms: P-BF, and non-plantain peel biofilm: N-BF, which served as control. The synthesized biofilms were characterized using FTIR, GC-MS, morphology test, water absorption property, biodegradation test, solubility test and swelling test and mechanical test; ultimate tensile test, flexural, hardness test, % elongation and thermo gravimetric analysis. The results of the FTIR analysis of the P-BF and N-BF showed 8 functional groups: ether, ethene, amine, carboxylic acid, nitriles, methylene, cyclic ester, primary, secondary and tertiary alcohols common to hydrocarbons. The results of the biodegradability test showed that both P-BF and N-BF biofilms degraded completely on the 12^{th} day. The results of the thermo gravimetric analysis showed that the P-BF biofilm decomposed at the temperature of 29.92^{0}C-500°C leaving 44.19% residue and compared to the N-BF biofilm which decomposed at the temperature of 22.17^{0}C - 500°C leaving 11.9% residue. This implies that, P-BF matrices, started degrading at 130 °C, while N-PF fibers start degrading at 139 °C. Therefore, the processing temperature for ripe plantain peels was set to below 137 °C to avoid unwanted degradation of the material. Furthermore, the results of the mechanical tests for P-BF showed ultimate tensile test, flexural test, hardness test, and % elongation; 2.87±0.02 Nmm$^{\wedge 2}$, 0.41±0.01 Nmm$^{\wedge 2}$, 22.00±1.78 Nmm$^{\wedge 2}$ and 6.29±0.01 Nmm$^{\wedge 2}$ respectively. while, the result of the mechanical test of N-BF showed ultimate tensile test, flexural test, hardness test and % elongation; 5.45±0.02 Nmm$^{\wedge 2}$, 0.41±0.01 Nmm$^{\wedge 2}$, 49.00±1.78 Nmm$^{\wedge 2}$ and 13.85±0.03 Nmm$^{\wedge 2}$ respectively. The solubility test of P-BF showed partial solubility in acetone, sulfuric acid and ethyl alcohol while the N-BF were completely insoluble in ethyl alcohol but partially soluble in sulfuric acid and acetone. The result showed that the biofilm was not completely soluble. The results of the swelling test for P-BF showed a mean of 0.04±0.00 when soaked in chloroform; and N-BF; 0.02±0.00. When soaked in methanol, P-BF showed a mean of 0.10±0.00 while N-BF showed a mean of 0.10± 0.01.The water absorption analysis showed that P-BF had 31% engorgement while N-PF had 46 % engorgement. The P-BF have high affinity for water due to the hydrophilic nature of the cellulosic fibers in the plantain peel as well as the water absorbing property of glycerol. The biodegradability test showed the plantain based biofilm degraded completely with no toxic effect on the soil implying that the biomass may serve as a potential material for production of an ecofriendly biofilm with good mechanical properties. The study showed that biodegradable biofilms can be synthesized from plantain peel at 3 % plantain peel, 1 % acetic acid, 3 % cassava starch, 1 % eggshell and 3 % glycerol. In conclusion, the study reported that powdered plantain peel-based biofilm with good mechanical properties can be produced.

KEY WORDS; Cassava starch, Biodegradable biofilm, powdered plantain peel-based, eggshells.

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND INFORMATION

Plastics are engineered natural polymers basically made from petroleum derivative-based synthetics (petrochemicals), which can be utilized in packaging, healthcare industries (medical instruments) and electronic industries (electronic goods).

There are around 8.3 billion tons of plastics that have been generated since the 1950s, and around 79% of them are not recycled, but left as waste in the environment (Applications Sectors—European, 2023). Deterioration of plastics in the climate requires several years, and, consequently, leaving plastic waste in the climate or land filling, making harmful contamination to the earth. Yearly, roughly 13 million tons of plastic waste have been tossed by people into the sea, which then hurts marine lives (Okunola, Kehinde, Oluwaseun and Olufiropo, 2019).

Endeavors have been made to deal with plastic waste issues. Techniques have been applied from the recycling process to the most extreme process (i.e., burning process) (Ezeoha and Ezenwanne, 2013). Recycling or reusing is one of the methodologies used to defeat the issues brought by conventional plastics, as the wastes could be recycled to be used as sustainable polymers (Yang, Zhang, Li and Wang, 2020). The ultimate aim of recycling polymer is to develop eco-friendly recycled plastics, which meet performance and quality requirements (Vieyra, Molina-Romero, Calderon-Najera and Santana-Diaz, 2020). Through recycling polymer, it can slow down the consumption of fossil-fuel based chemicals as the production of virgin plastics is slowed down. Nonetheless, the reused polymer items are worse contrasted with virgin plastics because of the synthetic pollutant during reusing of the plastic wastes (Faraca, 2019). In addition, the process of reproducing the plastics involves the grading of polymers, washing, grounding, and extruding, which will lead to a certain degree of degradation (Tang and Chen, 2019).

Likewise, recycled plastic can cause secondary effects on sullying the item, particularly when it is utilized as a food packaging material because of the presence of possibly cancer-causing substances (Jaramillo *et al.*, 2016). Burning plastic waste can contaminate the air on the grounds that the smoke created contains dangerous synthetic compounds like dioxin (Ezeoha and

Ezenwanne, 2013). Furthermore, Polystyrene, the major component in plastics can leach into water bodies such as oceans, seas and increase the toxicity in water. Aquatic organisms also can mistake the plastic floating on the surface of water for food which can result to death (Pawan and Malik, 2013).

Since plastic cannot be replaced by other materials, some researchers suggested developing bioplastics that are certainly more eco- friendly (Wu, 2009; Alves, Mali, Beléia and Grossmann, 2010). Bioplastics were developed as an approach to overcome the issue in the early 21st century (Rudin and Choi, 2013). Rudin and Choi, (2013) defined bioplastics as commercial polymer products produced by renewable resources or natural sources. Under reasonable circumstances, some of the bioplastics are biodegradable, and some renewable resources made of bioplastics could be recycled through biological processes; The common natural and renewable resources used for the synthesis of bioplastics are vegetable oil, starch, cellulose, protein, etc. (Harnkarnsujarit, Wongphan, Chatkitanan, Laorenza, and Srisa, 2021). Utilization of bioplastics has partly replaced the use of conventional plastics in various industrial applications, including packaging for food and others, medical instruments, hygiene, and agriculture. In recent developments, bioplastics can be applied in human bodies for medical usage, such as controlled drug delivery systems and therapeutic devices implantation (Narancic, Cerrone, Beagan and O'connor, 2020).

The European Bioplastic has reported that the production capacity of bioplastics in the year 2022 is 1,075 in 1000 tons, which is expected to increase to approximately 2453 in 1000 tons in the year 2024 (Europe, 2020). As time passed by, awareness towards preserving and protecting the environment increases, and there are more demands for replacing conventional plastics with bioplastics. In the world market, around 1% of plastics produced annually are biodegradable plastics, and the current trend shows that the bioplastics market has a continuous increment in various industries (Siracusa and Blanco, 2020).

Plantain peels are in many cases disposed of as, wastes and in street sides bringing about environmental pollution (Auta and Kumurya, 2015). The plantain peels make up around 40% of the complete organic product weight and have been viewed as a potential unrefined components in modern applications particularly agro-based enterprises (Gilver and Lilian, 2017).

The peels have also been utilized in the chemical industry for the development of ethanol. It has likewise been investigated in the food industry for making flour as well as to improve wheat flour (Arun et al., 2015). Notwithstanding, many research focused on synthesis of bioplastics from banana peels, cornstarch and so on. There is scanty or no exploration on blend of bioplastics from plantain peels or strips.

In light of the above, the current study was conducted to synthesize a biodegradable plastic from ripe plantain peels and characterization of the synthesized biodegradable plastics by FTIR, GC-MS and GC-FID, mechanical and chemical test.

1.2. STATEMENT OF THE PROBLEM

Synthetic polymer take a long time to be degraded, hence it leads to worsening of environmental pollution. Plastic waste has properties that are very difficult to be degraded by microbes in the environment since most plastics are made from synthetic or semi-synthetic materials such as polypropylene, polystyrene, and poly (vinyl chloride). The major component of plastic which is a polymer (Polypropylene/ Polystyrene) can leach into water and increase the toxicity in water. The plastic materials floating on the surface of water can be mistaken as food by aquatic organisms and eaten which may eventually leads to choking or even death (Pawan and Malik, 2013). Indeed, in nature, the degradation of plastics prepared from these raw materials takes a long time; hundreds or even thousands of years to break the carbon chain. (Wu *et al*., 2009; Jaramillo *et al*., 2016). In order to replace the use of plastics with another material that is more eco-friendly, the research will focus on renewable sources such as plantain peels, egg shells and cassava starch for the production of biodegradable plastic.

1.3. OBJECTIVES OF THE STUDY

The main objectives of the study was to produce a biofilm from renewable sources such as plantain peel, eggshells and cassava starch that will serve as an alternative to petrochemical based plastics.

The specific objectives of the study include; To:

a. Produce the raw materials for the production of biodegradable plastic such as glycerol, acetic acid (vinegar) and cassava starch.

b. Carry out pretreatment on some of the raw materials for the production of biodegradable plastic such as ripe plantain peels and raw egg shells.

c. Carry out confirmatory test on synthesized raw materials (starch test, acrolein test and ester test).

d. Characterize the synthesized raw materials by using FTIR (Fourier transform infrared spectroscopy) analysis and GC-MS analysis.

e. Produce biodegradable biofilm from the raw materials mentioned in (a) and (b).

f. Characterize the synthesized biodegradable plastic by conducting FTIR analysis, thermogravimetric analysis, biodegradation test, morphology study, chemical test (solubility and swelling test) and mechanical test.

1.4. JUSTIFICATION OF THE STUDY

Petroleum based plastics have become a major problem for the environment because it degrades slowly and contributes to environmental pollution. These will affect lands, waterways and ocean as well as with the marine organisms. Exposure of the chemicals to human is detrimental to health due to the carcinogenic potential of materials used in plastic production (Yaradoddi *et al*., 2016).

However, Biodegradable plastic is the best alternative to minimize the cost of solid waste management in Nigeria. Due to the availability of the waste biomass, it reduces cost of production. The production of biodegradable plastic can promote a sustainable solution to reduce plastic waste together with food waste in long term. Biodegradable plastic shows a great potential to overcome environmental pollution since it is biodegradable. Biodegradable plastic is potentially suitable to replace the plastic materials from petroleum based. Hence, the biodegradable films that shall be produced will be environmentally friendly and serve as an alternative to the conventional plastics (Yaradoddi, Patil, Ganachari, Banapurmath, Hunashyal, and Shettar, 2016).

1.5 SCOPE OF THE STUDY

This research covers laboratory scale synthesis of biodegradable plastics from plantain peels, cassava starch and eggshells, it includes:Collection of plantain peels and eggshells from restaurants, pre- treatment and processing of plantain peels, production of cassava starch, production of acetic acid(vinegar) from pineapple peels, production of crude glycerol from palm oil, pretreatment of eggshells, confirmatory test on the synthesized raw materials, characterization of synthesized biofilms based on their mechanical and chemical properties.

CHAPTER TWO

LITERATURE REVIEW

2.1. Plastics and the Environment

The worldwide utilization of plastics has expanded throughout the long term, especially on the grounds that they are lightweight, tough, moderately low-priced, and durable. The plastic industry produces around 300 million tons of plastics every year, which are utilized once and disposed of after use (Muller, Townsend and Matschullat, 2012).

Discarded plastic waste, owing to the durability and low degradability of these polymers, may take hundreds to thousands of years to decompose (Muller et al., 2012). Besides, of the total produced quantity of plastics, only 7% is recycled, while about 8% is incinerated and the residual and filled (Curia, Dautle, Satterfield, Yorke, Cranley and Dobson, 2019).

The Public Foundation of Sciences in 1975 evaluated that 14 billion pounds of trash was unloaded consistently, either covered underground or covered in the seas. Thus, seas and body of land are swarmed with plastics. As a matter of fact, in excess of 10 million tons of plastic waste is unloaded in the seas alone, so most of anthropogenic garbage littering the seas is made out of human-made plastics. Reports propose that plastics can now be utilized as a geographical stratigraphic sign of the Anthropocene period (Rochman, Tahir, Williams, Baxa, Lam, and Miller, 2015; Curia et al., 2019). This anthropogenic debris threatens ocean safety, integrity, and sustainability (Lewis and Hayes, 2019). Overall, plastic waste contributes to a pressing environmental problem is as yet unsolved.

2.2. Bioplastics

The ecological issues brought about by discarded plastics have made ready for the quest for substitutes. Bioplastics, which are both practically like manufactured plastics and earth maintainable, are promoted as promising new materials to resolve these issues.

Bioplastics are plastics integrated from inexhaustible biomass like cellulose, starch, chitosan and so forth. These plastics can be biodegradable like polycaprolactone or poly butylene succinate (PLC or PLB) or on the other hand might be non-degradable such as bio-polyproplene (bio-pp), bio-polyamide (bio-PA). They are non-degradable because they are fabricated partially from

renewable biomass and synthetic materials (Brydson, 1999; Gervet, 2007).The degradability of bioplastics is dependent on factors such as composition, degree of crystallinity and environmental conditions which in turn influence degradation times which ranges from seven days to several years (Mozaffari and Kholdebarin, 2019).

Biodegradable plastics are classified into two categories based on their degradation mechanism which includes: oxo-biodegradable plastics and hydrodegradable plastics (Iwata, 2015). Biodegradable plastics synthesized from petrochemicals in conjunction with a pro-oxidant are termed oxo-biodegradable plastics (Thomas, Clarke, McLauchlin and Patrick, 2012). The pro-degradant (a metal salt such as iron or mangnese salt) serve as a catalyst enhancing the degradation process aerobically (Da Luz, Paes, Nunes, Da Silva and Kasuya, 2013). Recently, oxo-biodegradable plastics are primarily synthesized from Naptha (a byproduct of oil) (Otaigbe *et al*., 1999). The time of degradation of oxo-biodegradable plastics ranges from months to years (Da Luz et al., 2013). However, the time of degradation can be programmed by the manufacturer (Thomas et al., 2012).

On the other hand, hydro-biodegradable plastics are synthesized from plant sources (such as cellulose, starch, polylactic acid etc). The degradation time of hydro-biodegradable plastics occurs at a much faster rate than oxo-biodegradable plastics. Additionally, they can be converted to synthetic fertilizers. Examples include bioplastics produced from plant sources (such as starch), and polylactic acid (PLA) (Ghada, Abanoub, Christopher, Joseph, 2021).

2.3 Sources of biodegradable plastics

2.3.1 Cellulose

Biodegradable plastics are integrated from cellulose rich materials like flax, hemp, bamboo, sisal and jute (Rahman and Baltimore, 2021).Cellulose is a polymer made out of glucose units consolidated by glycosidic bonds (Jamshidi, Hyon and Ikada, 1998). lt is segregated from its glasslike starch in microfibrils. It is soluble in methylmorpholine N-oxide. In any case, it doesn't make a good packaging material due to its poor solubility nature and highly crystalline structure. Additionally, the crystalline nature of cellulose makes cellulose based materials to exhibit poor flexibility, poor tensile strength as well as brittleness (Jamshidi et al., 1998).

The alternating hydroxyl side chains in cellulose contribute to the poor moisture barrier of cellulose (Jamshidi et al., 1998).

2.3.2 Cellulose derivatives

Biodegradable plastics are integrated from cellulose derivatives as a substitute for cellulose. These incorporate methyl cellulose, propylene cellulose, and carboxylicmethyl cellulose and hydroxyl propyl. They can shape gel on heating and hold their consistency when cooled (thermogelation) (Murray, Philips, and Williams, 2002). However, due to hydrophilic nature of the molecules, cellulose derivatives exhibit poor moisture barrier as well as poor mechanical properties (Gennadios and Weller, 1993).On the other hand, the use of hydrophobic compound such as fatty acid can enhance or improve the moisture barrier (Morrillon, Debeaufort, Blond, Capelle, and Voilley, 2002).

2.3.3 Starch

Biodegradable plastics are likewise blended from starch rich materials, for example, corn, potatoes and cassava. Starch is a polymer made out of amylose and amylopectin combined by glycosidic bonds (Jariyasakoolroj, Leelaphiwat and Harnkarnsujarit, 2018). It is insoluble in cool water, liquor and other solvent (Jane, 1995). It is utilized in food industry, paper, and material as well as drug ventures as a limiting specialist (Laycock and Halley, 2014).

2.3.4 Chitin/chitosan

Biodegradable plastics are also blended from chitin or chiosan. Chitin is a polysaccharide tracked down in the exoskeleton of arthropods and furthermore fish waste, egg shells and crabs (Shahidi, Arachchi, Jeon, 1999). It is ubiquitous in nature. Chitosan is a derivatives of Chitin or a deacetylated Chitin. It has excellent properties, for example, mechanical and microbial, good oxygen and carbon dioxide porousness, forming gel without added substances makes it reasonable for biodegradable plastic (Park, Li, Jin, and Cho, 2002).

2.3.5 Poly-beta-hydroxyalkanoates (PHB)

Poly-beta-hydroxyalkanoates (PHB), degrades in the presence of microorganism that comes in contact with the polymer, secrete enzymes and breaks down into smaller parts. properties of Poly-beta-hydroxyalkanoates are 100% resistance to water , 100% biodegradability and has the

ability to process thermoplastic (Chisti, 2014; Kumar, Shukla, Singh, Prabhakaran, and Tanwar, 2014 and Gadhave, 2018).

2.3.6 Polylactide acid (PLA) plastics

2-hydroxy propionic acid (PLA) is made up of lactic acid and contains methyl group on alpha C atoms (Hakola, 1997). Due to its biocompatibility, biodegradable and processing ability, it is an excellent material for packaging. It is synthesized by injection molding, blow molding, thermoforming and extrusion. Commercially, it was the first biobased polymer produced on a large scale, which could be shaped into various objects and films. It has replaced high density polyethylene (HDPE), low density polyethylene (LDPE), polyethylene terephthalate (PET) and polystyrene (PS) as packaging material (Rasal, Janorkar and Hirt, 2010).

2.3.7 Bioplastic from non-edible substances

In this day and age, where food is a scare resource, we can produce bioplastics from non-edible portions too. Things such as orange peel, pomegranate peel, banana peel, potato peel are used for the production of bioplastic. In the latest trend bioplastic films from polysaccharide residue feedstock is in great demand. Cellulose, hemicelluloses, starch, pectin make these lignocellulosic feedstocks useful for the production of bioplastic (Manali, 2021).

2.3.7.1. Pomegranate strips/peels

The strips of pomegranate have been viewed as a rich wellspring of bioactive mixtures (Malgorzata, Artur, and Barbara, 2016). It comprises of lignin-5.7% and hemicelluloses-10.8%, cellulose-26.2% and gelatin 27%. On acid hydrolysis, the polysaccharides present in strip are hydrolyzed to monosaccharides which can separate into cellulose, hemicelluloses and lignin parts. These parts further are utilized to create bioplastic (Chozhavendhan, Usha, Sowmiya, and Rohini, 2020).

2.3.7.2 Orange peel

The peel contains carbohydrates which can be used for the production of biomolecules. It is readily disposed off as waste. It is recommended to collect the waste and convert it to biodegradable plastics (Chozhavendhan et al., 2020).

2.3.8 Plasticizers used for the production of biodegradable plastic

Organic molecules known as plasticizers are added to polymers in order to increase toughness, durability, and minimize brittleness as well as lower melting temperatures (De Groote, Devaux and Godard, 2002). These lessen the interaction between polymers, which in turn lessens the stiffness of the three-dimensional structures and permits deformation without rupture (Mekonnen, Mussone, Khalil, and Bressler, 2013). Several plasticizers, such as fatty acids like palmitate or myristate and polyols like glycol, glycerol, sorbitol, fructose, sucrose, and mannose, are used in the manufacturing of biodegradable plastic. Due to its high boiling point (292°C), low cost, and non-toxicity, glycerol is the most widely used plasticizer (Forssell, Mikkila, Moates, and Parker, 1997).

2.4 Raw materials for the synthesis of biofilm

2.4.1 Ripe plantain peels

Plantain (*Musca parasidiaca*) is a fruit cultivated in the tropical regions such as Africa and South Africa. The plantain peels make up about 40% of the total fruit weight (Gilver and Lilian, 2017) and have been found to be a potential raw materials in industrial applications especially agro-based industries. Also, the peels have been employed in the chemical industry for the production of ethanol. It has also been explored in the food industry for the production of flour as well as to enrich wheat flour However, plantain peels have not been explored for synthesis of biodegradable plastics (Arun et al., 2015).

2.4.2 Acetic acid (vinegar)

Vinegar is formed when acetic acid bacteria is added to alcoholic beverages. In this process, oxidative fermentation takes place that creates vinegar as a by-product (Thokchom and Joshi, 2012). Vinegar is obtained by biotechnological process of double fermentation, alcoholic and acetic. Prior to the start of the acetic fermentation the fruit wine is subjected to alcoholic fermentation which is carried out with yeast, thus obtaining the amount of necessary alcohol to produce acetic acid (Dan, 2000; Rocio, Carmen, Torres, and Sanz, 2010).

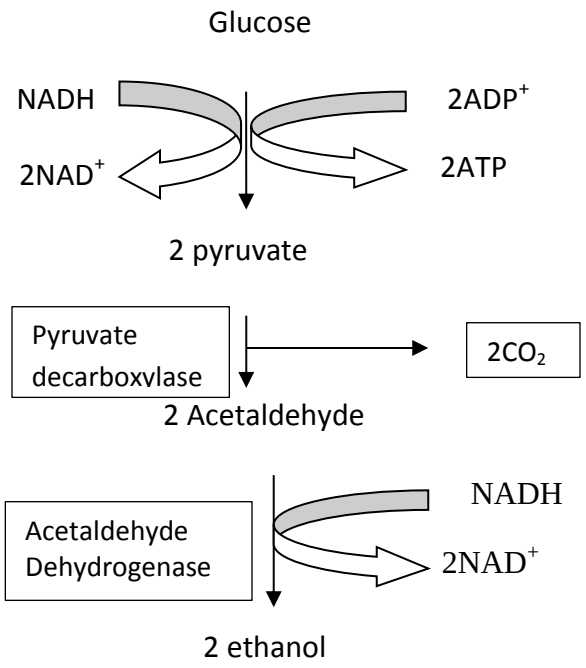
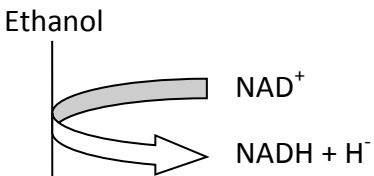


Figure 2.1: Pathway for alcohol fermentation



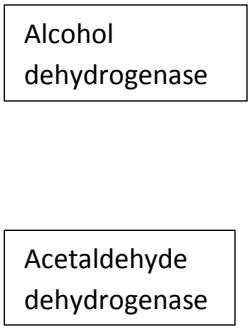


Figure 2.2: Pathway for Acetic acid fermentation

2.4.3 Cassava starch

Cassava (*Manihot esculenta frantz*) is widely cultivated for its various purposes such as food, feed, and raw materials for several industrial purposes owing to its high starch content (Mestress, Bangou, Zakhia, Rouau and Faure, 1996).

Cassava starch presents physicochemical characteristics of interest both for native starch application as well as for using as raw-material for modifications. Cassava sour starch is an example of a different product obtained by fermentation followed by sun-drying in some South American countries. Mainly in Brazil and Colombia, known as polvilho azedo and almidón agrio, respectively, is very valued and unique for production of gluten-free biscuits and bread-like foods (Mestress et al., 1996).

Cassava starch is obtained after processing cassava. It is used in sugar production and other industrial purposes due to its characteristic high freeze thaw, stability, high paste clarity, high paste viscosity etc. It is used majorly in the formation of matrix in biodegradable plastics.

2.4.4 Eggshells

Eggshell (ES) waste has been explored in recent years because it is economically cheap, abundant in nature and has intrinsic pore structure (Jianwei, Mokgadi, Katlego, Jacob and Maurice, 2012). The shells of an egg make up about 11% of the total weight of the egg. It contains 91% of $CaCO_3$. The generalized eggshell structure, which varies widely among species, is a protein lined with mineral crystals, usually of a calcium compound such as calcium carbonate. These characteristics qualify eggshell as a good candidate for bulk quantity , inexpensive, light-weight and low –load bearing composite applications, such as the automotive industry, trucks, homes, offices, and factories (Hassan, Aigbodion and Patrick, 2012; Stanley, 2016). Eggshells have been utilized as a're-enforcement materials in synthesis of Bioplastics (Syarifah, Syed, Nurul and Talalah, 2018) and biobag applications.

2.4.5 Glycerol

The chemical name for glycerin, often known as glycerol, is propane 1, 2, 3 triol. It is made up of three carbon chains to which a hydroxyl group is joined (Ueoka, 2001).

Additionally, glycerol is a colorless, odorless, thick, sweet-tasting, non-toxic liquid. It is produced as a byproduct of the transesterification, hydrolysis, and saponification reactions. When creating biodegradable polymers, glycerol is used as a plasticizer to make the substance more fluid. Depending on the procedures used and the kinds of materials handled, the crude glycerol obtained from oleochemical or biodiesel plants may contain a variety of contaminants, including soapy, alkali, and greasy components, a salt, or diols (Ueoka, 2001).Typically, it is referred to as crude glycerol or unrefined glycerol.

2.5.1. Properties of Bioplastics

The biopolymers' capacity to make their way to the market is mainly due to their properties that are not at all inferior to those of the conventional petrochemical polymers (Koch and Mihalyi, 2018; Ashok and Rejeesh, 2019 and Gómez and Michel, 2013). Bioplastics are mostly known for their environmentally friendly nature due to their lower persistence when abandoned in the environment (Calabrò and Grosso, 2018). Through the utilization of renewable resources such as organic waste (Jain and Tiwari, 2015) there exist additional benefits by the valorization of solid waste(Song, 2009; Peelman et al., 2013) that would be otherwise landfilled or used for energy recovery (Javierre, Sarasa, Claveria and Fernandez, 2015). Another property of the bioplastics is that they are non-toxic and compostable, which means that they are not harmful to living organisms (Gómez and Michel, 2013; Endres, 2017; Karamanlioglu, Preziosi and Robson, 2017). A crucial point is that biopolymers should have similar mechanical (Song, 2009; Gómez and Michel, 2013 and Peelman, 2013) chemical, and physical properties to conventional plastics(Gómez and Michel, 2013 and Peelman, 2013).These include tensile strength, tensile elongation, elasticity, flexural strength, density, crystallinity, melting point, water vapor and oxygen permeability, and also UV resistance (Peelman et al., 2013; Harmaen, Khalina, Ali, and Azowa, 2015, and Karamanlioglu et al., 2017).

However, the often low mechanical strength of bioplastics is the property that mostly limits their application (Ochi, 2005; Stevens, 2010) and requires the use of synthetic fibers, such as glass or carbon, to increase this property (Yang, Ching and Chuah, 2019). This leads to environmental problems due to reduction of their biodegradability (Yong, Ching, Chuah, and Liou, 2015; Yang et al., 2019). As a replacement of synthetic fibers, environmentally friendly materials, such as lignocellulosic fibers, fillers derived from cellulosic materials and starch (Muhammad et al.,

2019; Jangong, Gareso, Mutmainna and Tahir, 2019) can be added to biopolymers to reinforce the produced bioplastic(Sun, Yang, Lu, and He, 2019; Yang et al., 2019). Nanomaterials, such as nanoclay (Suryanto et al., 2019) or chitosan (Jangong et al., 2019) have been used to increase the thermal stability of bioplastics developed from Starch (Suryanto et al., 2019), which are known to have poor mechanical properties, due to their intra- and intermolecular bonds (Jafar, Mostafa, Esmaeil, Heidari and Rahim, 2011). Moreover, in a study conducted by Masruri, Azhar, Rosyada and Febrianto (2019), the addition of essential oil from kaffir lime to starch from cassava peel waste to produce bioplastic was found to increase the stability in tensile strength and the plastic was able to elongate by 65–85%. The simple addition of glycerol at a concentration of 5% (w/w) produced a bioplastic with a higher tensile strength of 205.52 N mm−2 and 42.69% of elongation (Saiful, Helwati, Saleha and Iqbalsyah, 2018). The addition of PLA (10%) to starch-based bioplastics enhances the general properties of Bioplastic composites (Abdullah, Fikriyyah, Putri, Puspa and Asri, 2019). Polyhydroxyaliphatic acids (PHAs) have mechanical properties similar to synthetic plastics, are insoluble in water and non-toxic, which makes them suitable for biomedical applications (Akinmulewo and Nwinyi, 2019). Another notable property of some bioplastics is the shape memory capability, which is a change of the shape of a material upon application of an external incentive (Javierre et al., 2015). Plasticizers, such as glycerol, glyoxal, or xanthan gum (Saiful et al., 2018; Jiménez-Rosado, Zarate-Ramírez, Romero, Bengoechea, Partal and Guerrero, 2019), are usually added to the starch-based bioplastics to improve their strength, flexibility, and the ability to process it (Abdullah et al., 2019; Jiménez-Rosado et al., 2019). An optimum bioplastics composition was found to be 12% (w/w) starch and 5% (w/w) glycerol (Saiful et al., 2018).

However, when it comes to the disposal or waste management system, with very few exceptions (Mostafa, Farag, Abo-dief and Tayeb, 2018) biopolymers cannot be chemically or mechanically recycled along with petrochemical plastics. For example, PLA can negatively impact the traditional recycling processes of PET (Niaounakis, 2019; Calabrò, Folino, Fazzino and Komilis, 2020).

2.5.2 Degradation of Bioplastics in the Environment

Just like the rest of the products, bioplastics have a life cycle. However, due to the wide variety of bioplastics, the life cycle of each material differs, depending on the type of bioplastic (Harrison, Boardman, O'Callaghan, Delort and Song, 2018), and on the end-of-life scenarios according to the available waste management systems in each country (Gómez and Michel).The degradability of bio-polymers is affected by the chemical and physical structure of the materials (Javierre et al., 2015; Endres, 2017) and not by the origin of their resources or their production process (Javierre et al., 2015; Endres, 2017). According to Endres (2017) "degradability is a functional property or a disposal option at the end of the material's life cycle". The degradation process depends on a combination of abiotic (UV, temperature, moisture, pH) and biotic processes and parameters (microbial activity) (Harrison, et al., 2018; Karamanlioglu et al., 2018).

It is worth mentioning that degradation differs from biodegradation. Degradation is a process of decomposition that stops at the fragmentation of polymers through the effect of heat, moisture, sunlight, and/or enzymes, which results in weakening the polymers' chains and thus creating even more persistent particles (Rujni´c-Sokele and Pilipovi´c, 2017; Ryan, 2018); biodegradation is the complete mineralization of materials in compounds such as carbon dioxide (CO_2), water (H_2O), ammonium (NH_4^+), nitrogen (N_2), hydrogen (H_2) and biomass through the biological action of microorganisms such as bacteria, algae and fungi (Urbanek, Rymowicz, Strzelecki, Kociuba, Franczak and Miron´czuk, 2017; Bilo et al., 2018). Therefore, biodegradation is not detrimental to the natural environment since the compounds produced are naturally occurring in the ecosystem (Urbanek, et al., 2017). The residues from the bioplastics' biodegradation are not generally toxic and can be consumed by other living organisms (Gómez and Michel, 2013). However, some issues have been mentioned in recent literature (Wierckx et al, 2018; Accinelli, Abbas, Shier, Vicari, Little, Aloise, and Giacomini, 2019) linked to the presence of fossil resins in the bioplastic items composition. Another factor affecting the rate of biodegradation is the thickness of the biodegradable material: The thicker the product, the longer its biodegradability (Rujni´c-Sokele and Pilipovi, 2017). Additional way to quantify the biodegradability of a product is the decrease of TC (total carbon) (Adhikari et al., 2016).There are also specific test methods that use the physical changes (e.g., discoloration, fragmentation) that the polymers undergo as indicators of biodegradability (Haig, Morrish, Morton and Wilkinson, 2018). The process of

biodegradation is influenced, apart from the material's physico-chemical structure, by the polymer's chain configuration. The longer the polymer's chain is, the more difficult it is to degrade. However, the polymer's crystallinity is also an important parameter of biodegradation, as the amorphous parts of the polymer are easier to degrade compared to the crystalline parts (Degli-Innocenti, 2014). As stated in Massardier-Nageotte, Pestre, Cruard-Pradet, Bayard (2006) "the more complex the formula, the less degradable because several micro-organisms are required to attack the different functions of the polymer".

Additionally, there are other parameters affecting the biodegradability of a material as the conditions and properties of the test systems. These include volume and shape of the vessels, open or closed bottles, mixing or shaking modes, oxygen supply, and test duration. Water-soluble polymers are easier to degrade, as water is a key factor for the development of microorganisms (Massardier-Nageotte et al., 2006).

2.5.3. Degradation Process

The study of biodegradation process is useful for assessing the environmental impact of bioplastic waste and finding inappropriate measure for implementing waste legislation and policies (Nandakumar, Chuah, and Sudesh 2021; Ghasemlou, et al., 2022).

Biodegradation is the process by which materials can be decomposed by microorganisms and used as a food source. The final products of the biodegradation process are CO_2 and H_2O, as also biomass and methane. However, although the material is biodegradable, it may not be in all circumstances or conditions. Several factors influence the biodegradation process, which adds up to microbial density and environmental conditions (i.e., temperature, humidity). These factors are polymer composition, molecular weight, crystallinity, pH, chemical structure, morphology, hydrophilicity, and breakdown products, but the relative extent of their effects is unclear (Meereboer, Misra, and Mohanty, 2020). The first stage involves the enzymatic or chemical hydrolysis of the polymer chain and the consequent formation of degradation products whose size allows for microorganisms 'encapsulation. It follows the decomposition and bio-assimilation of the fragmented polymers up to their conversion into carbon dioxide, nitrogen oxide, methane, and water. For this reason, several international methods for assessing the biodegradability of

plastics are based on the quantification of carbon dioxide production or the biochemical oxygen demand during the decomposition process (Suzuki, Tachibana, and Kasuya, 2021).

The abiotic or chemical degradation of bioplastics involves various techniques: pyrolysis, hydrolysis, alcoholysis, and glycolysis. Pyrolysis is a process of thermal cracking, in which the polymer, by heating in an inert environment, is converted into organic vapors, carbons, and gases. In the second stage, these by-products are converted into oil through a condensation process. Alcoholisys occurs when there is a trans-esterification reaction in which the alcohol group cleaves the external bonds, and the polymer chain splits into its monomers or oligomers. The glycolysis of polyesters involves the insertion of glycol in the polymer chains, breaking the external bonds and replacing them with hydroxyl terminals (Lambert, Román-Ramírez, and Wood, 2020). The reason for the large commercial spread of biodegradable bioplastics lies in their easy degradation, which would allow, under natural conditions, in soil, water, and sediment, to rapidly reduce the amount of plastic waste eventually improperly disposed off. The factor limiting the degree of degradation of most bioplastics is the surface of the polymer both in the case of chemical or enzymatic degradation. As already mentioned, the polymer surface is a fundamental aspect in the study of the degradation processes. Biotic (enzymatic) degradation occurs onthe surface due to the enzymes' large size which prevents their permeation in the inner polymer structure. Enzymatic hydrolysis of biopolymers is a two-step process: degradation begins with the adsorption of enzymes on the polymer's surface-active sites. The second step is a hydrolytic cleavage of polymer chain bonds, which is induced by the binding site of the hydrophobic portion and the catalytic site respectively (Meereboer et al., 2020). Accelerated by enzymes, surface degradation is much faster, especially in soil, and over time causes an increase in surface and roughness of the biopolymer and consequently higher hydrophilicity (Meereboer et al., 2020; Muthukumar and Veerappapillai, 2015).

2.6. Biodegradation under Different Environments

Plastics enter the environment as large or small plastic pieces can cause various environmental problems. They can change the way ecosystems work and harm living things. Eventually, they can end up in the food chain, which can harm human health. At present, stopping plastics from entering the environment in various forms is impossible. Thus, reducing environmental pollution caused by plastics or microplastics becomes more important. Recently, biodegradable film

packaging is a good and attractive option for plastics that can degrade in the environment. Biodegradable plastics will decompose completely over time, whereas non-biodegradable plastics remain in the environment for hundreds of years (Khalid and Arif, 2022).

A plastic is biodegradable if all its organic parts break down into carbon dioxide, water, mineral salts, and biomass under anaerobic conditions or carbon dioxide, methane, mineral salts, and biomass under aerobic conditions. During the biodegradation of plastics, some of the carbon is released into the atmosphere as CO_2/CH_4, whereas the rest is used to grow biomass such as microorganisms and fungi. The chemical structure of the polymer and surrounding environmental conditions greatly influence the biodegradation process (Khalid and Arif, 2022).

The biodegradation rate is affected by temperature, amount of water, nutrient availability, pH, amount of oxygen, concentration and activity of microorganisms, etc. Under the same environmental conditions, the decomposition rate of different products or materials may also vary. Under the same environmental conditions, the decomposition rate of different products or materials may also vary. As a result, it is necessary to take into account the biodegradation characteristics of biopolymer-based films in different environments. The biodegradation process and mechanism of biopolymer-based composite films in different Environments: (a) Soil environment; (b) Compost environment; (c) Water environment.

2.6.1. Soil Burial

Compared with petroleum-based plastic packaging, polymer packaging materials can biodegrade and decompose under the influence of microorganisms found in the environment. Soil stockpiling can be used to determine important details of the biodegradation process and illustrate the actual state of the biodegraded material. Soil conditions vary widely. Some soils are wetter and have more microorganisms than others. Differences in temperature and pH can also slow down the biodegradation rate. In soil and compost, scientists have found >90 types of microorganisms that can recycle biodegradable plastics. Generally, the film degradation process in soil occurs in two stages, starting with water diffusing into the film causing the film to swell accompanied by the growth of microorganisms which is followed by secretory degradation induced by enzymes and other substances, resulting in weight loss and film destruction (Khalid and Arif, 2022).

The degradation performance of composite films or mixtures is influenced by the film constituent materials and soil properties. Importantly, different locations, seasons, and rainfall have led to different soil qualities, and these elements directly influence how quickly the film can degrade in soil. Moreover, the exchange of gases and liquids in the soil and environment is affected bythe particle size of the soil. When the particle size is <2 mm, the soil is thick and has little exchange space with the environment and vice versa (Kliem, Kreutzbruck and Bonten, 2022).

The soil environment contains various microorganisms, such as bacteria and fungi, which can use biopolymers as their energy source and convert them into carbon dioxide, water, and new biomass, which in turn can contribute indirectly to the synthesis of various biopolymers (Wróblewska-Krepsztul, Rydzkowski, TBorowski, Szczypin´, Klepka, and Thakur, 2018). As an advantage, the use of soil burial to test the biodegradation of composite films or blends provides the most accurate picture of the environment and film deterioration process, and the testing cost is relatively low. Soil burial also has the following disadvantages: (1) it takes a long time, and most experiments take months;(2) biodegradability determined by weight loss sometimes cannot accurately reflect the actual results because removing soil, debris, and attached microorganisms from the material is difficult; and (3) degradation characteristics generally cannot be determined through repeated testing because of regional dependence (Kliem, Kreutzbruck, and Bonten, 2022).

2.6.2 Compost Environment

Composting (also known as organic recycling) is a biodegradation process that occurs under certain circumstances, depending on time, temperature, and the presence of microorganisms. Composting indicates that the material not only decomposes but also contributes nutrients to the soil in addition to being a usable component of the compost (Nilsen-Nygaard et al., 2021). In the composting process, the relative humidity is generally controlled at 40–55%, and the pH value is 6.5–7.5 (Di Piazza et al., 2020).

Several researchers have conducted degradation tests of packaging films under degradation conditions on compost. Recently, Mohammed, Gaduan, Chaitram, Pooran, Lee and Ward (2023) reported that alginate composite films extracted directly from Sargassum natans seaweed

degraded after 14 days under simulated conditions. Within the first week, an increase in deformation and opacity was observed, indicating the start of the hydrolytic breakdown process. This caused the alginate composite film matrix to crystallize and crack. With a different polymer material, Media-Jaramillo, Ochoa-Yepes, Bernal, and Famá, (2017) used films from cassava starch added with green tea and basil extracts. The film demonstrated significant degradation after 12 days in composting. Also, composite/mixed films can degrade quickly in days because the rich microflora of the composting soil likely contributes to the acceleration of film degradation (Abdillah and Charles, 2021).The characteristics of the composite film/mixture can also influence the degradation rate of films during composting. For example, the addition of biodegradable components (starch, protein, etc.) in the films can increase the film's hydrophilicity so that it degrades quickly. Factors such as temperature and humidity in the compost environment also significantly influence the speed of film degradation (Ruggiero, Carretti, Gori, Lotti and Lubello, 2020).

2.6.3. Water Environment

Studies on film biodegradation in aquatic environments mainly focus on seawater, freshwater, and river water. Seawater has highly variable temperatures ranging from 30 to −1°C, is highly saline (34–37 ppt), and has a lower concentration of microorganisms than freshwater. Freshwater can be stagnant (lakes) and moving (rivers) water, and significant difference from seawater is that the salt content is lower than 1 ppt. Freshwater has a pH range of 6–9, and biodegradation is generally caused by bacteria and fungi (Kliem et al., 2020). Abdillah and Charles (2021), comprehensively studied the biodegradation rate of arrowroot starch (AS)/carrageenan (IC)-based films in seawater and a compostable environment. Their results revealed that the AS 4% + IC 0% and AS 3.5% + IC 0.5% blend films were completely degraded after 42 days in seawater compared with only 7 days in a compost environment, which is relatively fast, because plastic materials degrade slower in the sea than in the soil environment due to less exposure to thermal oxidation. In addition, films biodegrade faster in the soil environment than in water because the water environment has a relatively low temperature and insufficient microbial abundance.The water environment is also strongly influenced by climate, light, and other factors that affect film.

2.7. Bioplastic-Degrading Microorganisms

Even though microorganisms are a key factor for bioplastics disintegration, their role during the biodegradation process is still poorly understood (Karamanlioglu et al., 2017). Biodegradation, through the microorganisms' action, enables carbon to be mineralized at the end of the biodegradable polymers' useful lives following their disposal, without releasing harmful compounds into the environment (Volova, Gladyshev, Trusova, and Zhila, 2010).

Typically, biodegradable polymers are decomposed by microbial attack in a single step (Karamanlioglu et al., 2017).The depolymerization releases monomers that are assimilated by the surrounding microorganisms (Degli-Innocenti et al., 2014). Depolymerization occurs due to the functional process of intracellular and extracellular enzymes, with the latter consisting of endo- and exo-enzymes. Endo-enzymes are responsible for the random breaking of the internal bonds of the main polymer chain, whereas exo-enzymes break the polymer chain sequentially (Jain and Tiwari, 2015).

Another basic mechanism in the biodegradation of biodegradable polymers via the action of microorganisms is the hydrolysis by enzymes, which improves the hydrophilicity of the material, resulting in lower molecular weight polymers, which facilitates microbial assimilation (Thakur, Chaudhary, Sharma, Verma and Tamulevicius, 2018). Differences in the biodegradation rates amongst biodegradable polymers are due to the structural and physicochemical properties of their surfaces, allowing stronger or weaker attack of microorganisms on the surface (Bátori et al., 2018).The process of biodegradation involves prokaryotic (bacteria, archaea) and eukaryotic (fungi and protozoa) microorganisms, which have the ability to degrade the polymeric matrix and/or utilize the energy-storing materials (Rujni´c-Sokele and Pilipovi´c, 2017). More than 90 types of microorganisms are responsible for biodegradation in different environments (Thakur et al., 2018). Bacteria and fungi are more commonly involved in bio-polymers' biodegradation (Li, Witt, Xie, Warren, Halley, and Gilbert, 2015). There are several bioplastics that present different biodegradation behaviors under aerobic and anaerobic conditions. One major reason, highlighting the importance of the microbial communities in biodegradation, is the influence of fungi. Fungi are only active in aerobic environment as well as in compost and soil. In other words, some polymers are mainly (or even only) degraded by fungi and not by bacteria, and will therefore biodegrade to a higher extent under aerobic conditions (Rujni´c-Sokele and Pilipovi,

2017). According to some studies, rates of biodegradation are slower under anaerobic conditions due to the lack of oxygen and due to the limited microbial diversity enzyme availability (Thakur et al., 2018).

It is important to note that, not every type of bioplastic can be degraded by every type of microorganism and vice versa. On the contrary, it is essential to take into consideration the fact that each material biodegrades better under specific conditions. For example, it has been reported that PCL can be degraded by bacteria isolates that exist in deep sea sediments, but these isolates are incapable of degrading other types of bioplastics, such as PLA, PHB, and PBS; however, there exist composting bacteria capable of degrading the latter (Emadian et al., 2017). An interesting detail is the fact that PCL-degrading microbes that have been located in deep seawater have not been found in coastal environments (Suzuki et al., 2017). Another example would be that of PBS that under aerobic conditions (composting) presents an extent biodegradation of approximately 31% in 80 days, while PBS under anaerobic conditions (landfill) biodegrades only by 2% in 100 days (Cho et al., 2011). Polylactic acid (PLA) is well degraded by *actinobacteria* that belong to the family *Pseudonocardiaceae*, and by other taxa that include members of the family *Micromonosporaceae*, *Streptomycetaceae*, *Streptosporangiaceae*, and *Thermomonosporaceae* (Butbunchu *et al*., 2015). Polyhydroxyaliphatic acids (PHAs) and its copolymers can be degraded by several bacteria and fungi through production of intracellular and extracellular depolymerases (Roohi *et al*, 2019). In particular, *Enterobacter sp*. (four strains), *Bacillus sp*., and *Gracilibacillus sp*. were found to be the PHA-degrading strains in a tropical marine environment (Volova et al., 2010).

2.8. Waste Management Options for Bioplastics

Bioplastics is a large family of polymers that include many different materials. Each should be treated by a different waste management option according to its characteristics (European Bioplastics, 2020). Fossil-based polymers have been only minimally replaced by bioplastics and the impact of these new materials on waste accumulation is still not completely evaluated (Vu, Åkesson, Taherzadeh, and Ferreira, 2020). Although petroleum- and bio-based plastics may have similar mechanical properties, they are produced from different raw materials, thus the introduction of bioplastics in treatment/recycling systems used for traditional plastics is not feasible. In this sense, the redesign of current recycling systems is needed to further limit the

potential environmental impact of bioplastics (Vu et al., 2020) and to avoid an excessive resource use. On the other hand, products like PLA can be processed by several waste recovery methods, such as mechanical recycling and chemical recycling (Kawashima, Yagi, and Kojima 2019).

The options for the management of bioplastics at the end of their life are mainly dependent on the physico-chemical conditions of the treatment, and include: Biological waste treatment, recycling, incineration, and landfilling (Song et al., 2008; Hottle, Bilec, and Landis, 2017; Rujni´c-Sokele and Pilipovi´c, 2017).The process of biodegradation occurs during the biological waste treatment (e.g., composting, anaerobic digestion) and it can take place in a variety of environments (Volova et al., 2015). For instance, bioplastics can be degraded in aerobic environments such as in soil, composting, and some aquatic environments, but also in anaerobic environments such as anaerobic digestion plants, landfills, and a few aquatic environments (Rujni´c-Sokele and Pilipovi´c, 2017).

2.8.1 Mechanical and Chemical Recycling

According to EPA, only 9% of the annually manufactured plastic enters the waste stream for recycling (Kawashima et al., 2019). However, bioplastics can be mechanically recycled. Mechanical recycling of biopolymers was introduced in the 1970s and consists of the mechanical processing of plastics' waste to obtain secondary raw material for the production of new objects with similar properties. Mechanical recycling consists of the following phases (Ignatyev et al., 2014; Singh et al., 2017; Vu et al., 2020).

• Removal of contaminants, such as food waste (Kawashima et al., 2019);

• Grinding/shredding/crushing or milling, to obtain a material as much homogeneous as possible.

• Further processing, such as extrusion, injection molding, or drawing.

The main disadvantage of this scenario is the fact that normally each polymer should be separately treated to obtain a good secondary raw material and that every time a biopolymer is reprocessed there is loss in the physical and mechanical properties of the material (Scott, 2000). For instance, even though very few studies on pure polyhydroxyalkanoates (PHA) recycling are available PHA can be recycled but it exhibits a loss in mechanical properties (Fábio Rivas et al.,

2017). Similar findings are observed in terms of reduction of tensile strength for polyhydroxybutryates (PHB) after multiple processing cycles, even though its chemical structure and thermal stability remains unchanged (Fábio Rivas et al., 2017). Other biocomposites, such as polyhydroxybutryate-co-hydroxy valerate (PHBV) (a copolymer of PHB and PHV), have the ability to be recycled up to five times without experiencing any physico-mechanical losses (Soroudi and Jakubowicz, 2013).

Apart from that, when a biopolymer enters into the recycling stream with the rest of the conventional plastics, it might cause contamination to the waste as not all of the bioplastics are compatible for recycling, resulting in the potential downgrade in the quality and physical integrity of the producing mixed-plastic products (Soroudi and Jakubowicz, 2013; Ashok and Rejeesh, 2019).

2.8.2. Biological Treatment

Biodegradability evaluation is crucial when bioplastics are biologically treated (Mohee et al., 2006). As demonstrated by Scott (2009) "polymers must remain stable during manufacture and use but should break down rapidly after disposal with conversion to biomass and/or mineralization in an acceptable time". This summarizes the main idea that pushed to develop the bioplastics. Among the other treatment methods for used bioplastics, microbial degrading activity is a useful way for increasing environmental safety and economic value (Butbunchu et al., 2019).

Biological treatment applies strictly on biodegradable bioplastics, due to the fact that this disposal option utilizes the feature of biodegradability that these polymers provide. The benefit resulting from these treatments, apart from reducing the amount of waste that would otherwise be dumped in the landfills, is energy recovery via anaerobic digestion and/or production of a soil amendment through composting (Song et al., 2009).

2.8.1.3. Incineration

Incineration, which is a frequent waste management option, is the thermochemical decomposition of a substance by heating, where the organic materials are burnt for energy recovery. Its main benefit is that it can be applied to all types of polymers and the produced energy from polymers' incineration can be profitable if sold (Garrison et al., 2016). Another

noteworthy ability, apart from the same calorific value, that bio-based bioplastics have over the conventional plastics is that they are CO_2-neutral. On the contrary, the fossil-derived biodegradable bioplastics, contribute to the rise of emissions as their feedstock is of fossil origin. The differentiation between biogenic and fossil carbon lies in the fact that the latter was fixed by primary producers from the atmosphere millions of years ago, and then sequestered, becoming unavailable to global bio-geo-chemical cycles. However, its sudden and massive recent release into the atmosphere following combustion processes, in the form of fossil-derived CO_2 is believed to cause global warming (Garrison et al., 2016). Moreover, although recycling and materials recovery should be preferred over other waste disposal options, incineration represents a viable alternative to landfill disposal for many countries (Kawashima et al., 2019) as it is a means to recover energy.

2.8.1.4. Landfilling

It is estimated that almost 40% of the annually produced plastics is discarded into sanitary landfills (Jafari-Sales, 2017; Rahman and Syamsu, 2018). Landfilling is still a popular waste management scenario, due to its low cost and simplicity of operation, as previous sorting of waste, or any other pretreatment, is not required. Nonetheless, disposal of bioplastic waste to a sanitary landfill remains the least preferable option (but preferred over the uncontrolled dumping). That is mainly because under anaerobic conditions, such as in landfill or dumps, anaerobic decomposition results in fugitive methane, which is a greenhouse gas when escaping the recovery system (Hottle, Bilec, and Landis, 2017). In fact, these generated greenhouse gases include methane (CH_4), which is a gas with a warming potential 25–36 times that of CO_2 (Hottle et al., 2017). However, the landfill gas can be at least partially recovered and combusted for energy production (Hottle et al., 2017).

2.8.5. Qualitative studies

Qualitative analysis of building blocks in bioplastics is possible by spectroscopic methods such as fluorescence, nuclear magnetic resonance (NMR), Fourier-transform infrared spectroscopy (FTIR), or UV-VIS spectrophotometry techniques (Kumar, Singhal, Verma, and Thakur, 2017; Akdoğan and Çelik, 2018). Microparticles can be determined analytically either by analyzing them as they are or by analyzing them after dissolution in solvents. FT-IR micro imaging, gas chromatography with mass spectrometry detector, and thermal analysis represent the most used

analytical techniques for the analysis of the solid debris of microparticles, while liquid chromatography, always with mass spectrometry detection, is currently the most widely used technique in the analysis of the dissolved microplastics or of their fractions (Sikorska, 2020; Ye, 2022; Yusuf, 2022). Microparticles can be determined in their native status, or after dissolution in solvents. FT-IR micro imaging, gas chromatography coupled with mass spectrometry (GC-MS), and thermal analysis represent the most used analytical techniques for the investigation of solid microparticles. Liquid chromatography, coupled with mass spectrometry (LC-MS) detection, is currently widely used for dissolved microplastic fractions, (Sikorska, Zi, Musioł, Kowalczuk, Janeczek, and Chaber, 2020; Ye and Zhao 2022; Yusuf, Sodiq, Giwa, Eke, Pikuda and Eniola, 2022).

2.8.5.1 Fourier transform infrared spectroscopy

Fourier transform infrared spectroscopy (FT-IR) analysis has the advantage of being a non-destructive and rapid technique, with minimal sample preparation. The poly-hydroxy-alkenoates (PHAs) analysis is based on the study of the variation of the intensity of the stretching band of the carbonyl of PHAs as a function of concentration, in the range of 1728–1740 cm−1 of wave numbers (Godbole 2016; Isak 2016). However, in the last years, to investigate particles up to 10 μm, the more helpful instrument is the FT-IR micro imaging system, (μFT-IR Imaging). μFT-IR is an instrument with a state-of-the-art infrared detector that simultaneously generates a high number of spatially resolved spectra and analyses large sets of microplastic data showing a "visible" image of the sample. Through appropriate software, it compares the spectra of the microparticles present in environmental matrices with a data base of the reference spectra, allowing a very good identification of the microplastic by size, volume, and mass (Karami, Golieskardi, Choo, Larat, Karbalaei, and Salamatinia, 2018; Liebezeit, 2014).

2.8.5.2. Ultraviolet-vis spectrophotometry

This classical and easy technique finds an interesting use in the determination of PHB by exploiting the degradation of the molecule of P (3HB) to crotonic acid by heating in concentrated sulphuric acid and determining its content by studying the absorbance mass of the crotonic acid band at 235 nm (Duvigneau, Kettner, Carius, Griehl, Findeisen, and Kienle, 2021). This technique nowadays, for its easiness, is still used, though do not allow us to determine PHB copolymers.

2.8.5.3 Fluorescence Spectroscopy

The application of this technique in PHAs analysis is based on the fluorescence of Nile-Red, a lipid fluorochrome that easily penetrates the suspended cells making fluorescent the polymer portion contained in them. The concentration of polymer in the cells can be determined from the analysis of the fluorescence intensity. The intensity of the fluorescence emission of red stained cells with Nile-Red increases with the biopolymer concentration.This method for the determination of the biopolymer concentration has several advantages: it is fast and reproducible, measurements can be made immediately after sampling, sample preparation time is shorter than traditional methods and sample volumes for analytical determination are very small (Godbole, 2016; Rajankar, 2018). However, Nile Red can stain not only biopolymers (PHAs or PLA) but also other lipophilic compounds (Arikawa, Sato, Fujiki, and Matsumoto, 2017) and for this reason, fluorescence spectroscopy can be classified as qualitative rather than quantitative technique.

2.8.5.4. Qualitative Thermal Analysis

Among the different techniques available, thermal techniques are widely used in the design, preparation, and characterization of polymeric materials. Thermal analysis (Differential Scanning Calorimetry and Thermogravimetric Analysis) offers, in addition to high precision in measurement, smart execution, allowing to obtain with a very limited amount of material valuable information regarding the property-structure correlation (Blanco and Siracusa, 2021).

2.8.6. Quantitative Studies

2.8.6.1 Gas Chromatography

Gas chromatography has the advantage of providing very detailed, accurate, reproducible, and precise measurements, but conversely uses solvents harmful to the environment and requests long sample pre-treatment and large quantities of the sample (Khok, Suwa, Ito, Hazwan Hussin, Ishida, and Sudesh, 2020).

The most common method used to determine the polyhydroxyalkanoate (PHA) content in cells is gas chromatography with a flame ionization detector (GCFID). This method is quite laborious, but it has high accuracy and provides extensive information on the composition of the monomer of PHAs (Isak, 2016). Gas chromatography mass spectrometry (GC-MS) allows the

determination of polyhydroxybutryate(PHB) content in cell biomass after an initial stage of methanolysis (Khok et al., 2020), poly(3 hydroxybutyrate) P(3HB) content after acidic or basic digestion, and PHA content from freeze-dried bacteria or natural sample extracts after acidic digestion (Godbole, 2016).

The GC-MS drawback is the use of not eco-friendly solvents. To overcome this side effect, it is better to use the pyrolysis technique directly coupled with gas chromatography (Py-GC), which is a valid method in the direct analysis of the content of PHB and its copolymers in prokaryotes (Baidurah, Murugan, Joyyi, Fukuda, Yamada and Sudesh, 2016; Khang, Kim, Yoo, Sohn, Jeon, and Park, 2021).

2.8.1.6.1.2 Quantitative Thermal Analyses

Thermogravimetric analysis associated with mass spectrometry (TGA-MS) is a direct method for the quantitative determination of polyhydroxybutyrate (PHB) and polylactic acid (PLA) in soil. This technique is based on the analysis of the masses of the products developed during gaseous pyrolysis in an inert atmosphere. A portion of gas degradation products is transferred to a quadruple mass spectrometer via a heated capillary. The loss of mass at a specific temperature may be related to the mass signal of the gaseous pyrolysis products. These pyrolysis products' formation can be related to their specific degradation temperatures determined by the TGA. The advantages of the method are the absence of sample pre-treatment and the use of an internal standard but the sample to be measured is often of some micrograms giving problems of homogeneity and significance in sampling (Duvigneau et al., 2021).

2.8.6.3 High-performance liquid chromatography-HPLC

High-performance liquid chromatography (HPLC) is a valid method in the analysis of the soluble fraction of biopolymer degradation products. In the case of PHB analysis the degradation products such as crotonic acid and 2-pentenoic acid, are easily separable and can be quantified. The analytical results are comparable to those obtained with GC-MS, but with shorter analysis times (Duvigneau et al., 2021) and easier sample treatment.

2.9. Applications

Plastic is the primary environmental pollutant that is used on a daily basis (Pradhan et al., 2014). Therefore, instead of using petrochemical-based products, many research has turned to biodegradable plastic to reduce environmental pollution. Numerous environmental problems can be resolved in this way (Kalia et al., 2011). According to Shamsuddin et al., (2017), biodegradable plastics have special qualities that make them eco-friendly, compostable, biodegradable, and energy-efficient. The uses of biodegradable plastics in various industries are listed below;

2.9.1 Healthcare industry

Several kinds of thermoplastics have been used in the healthcare sector because of their superior application-specific qualities. Plastic medical equipment, such as IV tubes, surgical gloves, blood bags, syringes, and instrument packaging, are widely used and helps maintain hygienic conditions. These days, biodegradable bioplastics are used in medical applications like therapeutic device implantation and controlled drug delivery systems (Narancic et al., 2020). Any medical equipment must be sterilized (e.g., by high temperature steam sterilization, ethylene oxide (EtO), or gamma irradiation). It also has a high probability of coming into contact with bodily fluids or other chemicals, which frequently caused the rate of biodegradation to increase and the molecular weight of polymers to decrease (Pérez et al., 2021).

In other words, materials used in these kinds of applications need to be highly resistant to various chemicals and sterilization procedures while still preserving the instruments' functionality and safety (Sastri et al., 2010). Due to their ability to be biodegraded by bacteria and fungi found in human bodies, biodegradable bioplastics like poly (lactic-co-glycolic acid) (PLGA), PLA, and poly (ε-caprolactone) PCL are utilized in medical applications, including tissue engineering (Bano et al. 2018).

Bioplastics are applied to human bodies for use in biomedical applications. This frequently calls for the bioplastics to be non-toxic and biodegradable, meaning that using them does not need the applicants to undergo any additional procedures to remove them from their bodies or suffer any negative effects (Bano et al., 2018).

As stated by Advanced Drug Delivery Reviews (2016), applications of using reinforced PLA include gene delivery, tissue engineering, implants, shape memory, and controlled-release drug delivery (Liu et al. (2011; Saini et al., 2016). Reinforced PLA through blending with ethylene vinyl acetate copolymer (EVA) to produce paclitaxel-eluting stent coatings, which could modulate the drug release amount and rate through adjusting the PLA amount in the formulation. In a research conducted by Liu et al., (2011) and Saini et al.(2011).It was found that the use of PLA as one of the components in tissue engineering scaffolds gives better processing property as well as using reinforced PLA, i.e., poly(glycerol sebacate) (PGS)/PLA blend, incorporated the properties of faster degradation and better wettability (the tendency of one fluid to adhere to a solid surface in the presence of other immiscible fluids), which could further improve the biodegradability and compatibility with the tissue recovery period (Saini et al., 2016). Besides, reinforced bioplastics are widely applied in packaging for healthcare products, including medical and personal care (Saini et al., 2016).

Reinforced bioplastics increase the functionality of the packaging since bioplastics have been shown to be hygienic for the packaging of healthcare products. With better mechanical qualities, reinforced bioplastic packaging is more resilient to tearing or shattering from mechanical stress. These qualities may also help shield medical equipment packed in reinforced bioplastics from bacterial and viral contamination. Polysaccharides, chitosan, and chitin give the packaging antibacterial qualities while extending its shelf life (Pellicer et al., 2017).

According to research by Dan Kai et al. (2018), lignin-reinforced bioplastics have a high level of antioxidant activity and may be used as antioxidants to shield the skin from radiation and other pollutants, protect humans from oxidative stress, and encourage the regeneration of cartilage tissue (Kai et al., 2019). Similarly, bioplastics are used as a foundation for the manufacturing of sanitary products because the materials are waterproof and breathable, allowing water vapor to pass through (Kai et al., 2019).Soft bioplastic foils are already in use as disposable gloves, incontinence products, bed underlays, diaper foil, and women's sanitary products (Shah and Srush, 2017).

2.9.2 Electrical and electronic industry

These days, bioplastic is widely used in the electrical and electronic industry (E and E industry). Bioplastics transmitters, also known as strong polymeric electrolytes (SPEs), are used to improve electrochromic devices, batteries, diodes and power modules (Gökçe et al., 2020; Sidek et al., 2019). In addition, bioplastics are widely used in the gather part for requesting purchaser items, such as packaging for PC components and cell phones, speakers, PC mice, and vacuum cleaners (Harnkarnsujarit et al., 2021). Bioplastics can be used as layers for electroacoustic devices, support for electronic paper, and for water treatment (Sidek, et al., 2019).

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Both cathode nanotubes and cellulose nanofiber-reinforced bioplastics are used in a variety of industrial applications, including sensors, roll-to-roll fabrication processes, flexible photovoltaic cells (solar cells), and advanced electronics (Bhat et al., 2016).

When added to bioplastics, graphene, which has exceptional thermal, mechanical, and electrical properties, significantly improves the mechanical qualities of the material while retaining its high flexibility and adding electrical conductivity (Catald et al., 2018). It was stated that the PLA reinforced with graphene might find use in orthopaedic and scaffold applications (Bustillos et al., 2017). Additionally, PLA is enhanced with carbon fiber, which has superior mechanical, electrical, and thermal properties. This gives the bioplastics exceptional electrical conductivity and shields them from electromagnetic interference (EMI) (Solafide et al., 2019).

Figure 2.3: Bioplastic Electronics
Source: www.greencompostables.com/Bioplasticnews.com

2.9.1.3 Architecture and Construction Industry

Plastics have been used in building and architecture for a few decades now. These industries frequently use plastic for cables, floor coverings, pipes, insulation, and other purposes. Bioplastic materials are used in the architecture and construction sectors for wall cladding, pipes, geotextiles, and façade elements. High strength is needed for traditional plastic applications in the industry, such as textile fleece, cables, and floor coverings, in order to withstand various extreme workload scenarios. Bioplastic materials offer a wide range of applications in the industry, but their cost is high because higher-quality bioplastics typically require additional processing costs, and conventional bioplastics performance in the market is insufficient and unsuitable for widespread use (Gökçe, 2018).

According to Ivanov and Stabnikov (2017), the use of biodegradable bioplastics can benefit the construction industry in ways like environmental and bioeconomic sustainability, lower disposal costs for construction waste, and lower costs associated with temporary excavation for construction (Ivanov et al., 2017). Reinforced bioplastics can increase strength, durability, recyclability, and water resistance. Bioplastics are commercially available to be used as stabilisers for earthen construction materials, which previously used cement (Oberti, et al., 2022).A study on the use of gel-type bioplastics reinforced with xanthan gum and natural fibers revealed that these materials have lower shrinkage values and better mechanical properties (Oberti, et al., 2022). Moreover, because of their hydrophobic quality and resistance to bio-deterioration, lignocellulosic fiber-reinforced bioplastics are frequently utilized in construction products, such as window frames and doors (Verma et al., 2021).

Furthermore, due to their hydrophobicity and resistance to bio-deterioration, lignocellulosic fiber-reinforced bioplastics are widely utilized in construction products, such as door and window frames (Verma et al., 2021). Lignocellulosic fiber-reinforced bioplastics also offer strong mechanical properties, elasticity, and biodegradability. In addition, bioplastics reinforced with clay or carbon nanotubes (CNT) such as PLA, have been found to exhibit improved mechanical properties, including tensile strength, scratch resistance, and break elongation (Ivanov et al., 2017).

Figure 2.4: Bioplastic Façade
Source:urbannext.net

2.9.4 Agricultural Industry

Due to their strength, resistance to water, light weight, and protective qualities, plastics are used extensively in the industry and can increase crop yields (FAO, 2021). In the agricultural sector, bioplastics are also used in mulching, seedling trays, pots, and polymer-coated fertilizer (Harmaen et al., 2016; Acquavia et al., 2021; FAO, 2021). After seeds sprout and grow, seedling trays usually disintegrate in the soil; during this process, non-toxic chemicals are released into the soil or taken up by plants (Paul et al., 2021). In addition, polyhydroxyalkanoates (PHAs) has been used as a carrier for fertilizers, seed encapsulation, crop protection films, and insecticides (George et al., 2021).

Furthermore, lignocellulosic fiber-reinforced bioplastics speed up the rate at which the polymeric matrix degrades in soil, therefore they are better suited for use as plant nursery bags or pots in the agricultural sector (Cinelli et al., 2019). Reinforced bioplastics have also been used as agricultural product packaging because their porous nature allows for better air flow, which preserves freshness, and because their increased strength and flexibility prolong the packaging shelf life (Iriani et al., 2019).

2.9.5 Packaging Industry

In the packaging sector, bioplastic is frequently used to make bags, films, and wraps. Non-biodegradable bioplastics are used in the beverage packaging market. For example, PepsiCo produced 100% Bio-PET for their product packaging using switchgrass, corn husks, and pine bark, while Coca-Cola created PlantBottle® synthesis from bio-based ethylene glycol (which contains 30% Bio-PET) (Lorite et al., 2017).

Fresh fruit packaging uses reinforced bioplastics made from agricultural waste materials like rice straw. Research indicates that these reinforced bioplastics may extend the shelf life of fresh fruit by enhancing the permeability of the bioplastics, which in turn facilitates better air flow for fresh fruit transpiration (Iriani et al., 2019). Food packaging is one of the industrial uses for bioplastics. (Hong et al., 2021).

Packaging industry also uses bioplastics in the form of takeout bags, dishes, films, bottles, and containers for dairy products (Hong et al., 2021). In addition, it has been reported that active food packaging uses fiber-reinforced PHBV, which can extend the shelf life and give the packaging film antimicrobial properties, increasing the likelihood of preventing food spoilage

(Torres-Giner et al., 2018). Furthermore, in packaging film developments, cellulose-based films reinforced with clay also exhibit antimicrobial properties with improved properties in gas permeability and thermal stability (Ibrahim et al., 2021). Furthermore, it was demonstrated that reinforced bioplastics have antimicrobial properties when poly(butylene adipate-co-terephthalate) (PBAT) reinforced with thermoplastic starch (TPS) bioplastic films could lower the number of mold and yeast while preventing food from darkening which further proved that reinforced bioplastics possess antimicrobial properties (Jariyasakoolroj et al., 2020).

In a report by Gadhave et al. (2018), the addition of acetylated starch in corn starch-based films was found to show higher thermal stability with reinforced resistance against sealing, making it a an excellent material for heat sealing packaging Gadhave et al. (2018). Furthermore, it was stated that reinforced bioplastics could be the material for optoelectronic packaging in the future (Kalia et al., 2011). Optoelectronic packaging (OEP) is a type of electrical packaging that involves providing mechanical support, as well as electrical and optical connection, to electronic devices for the device continuous functionality, and the most commonly used material for this application is epoxy (Boudreau, et al., 1994; Kalia et al., 2011).The eco-friendly property of bioplastic also make it a suitable material for airline cosmetics (Kuciel, et al., 2019).

Figure 2.5: Bioplastic Packaging Material

Source:www.shimadzu.com/europlas.com.vn

2.10. Benefits and Constraints of Bioplastics

One of the targets for the development of bioplastics is to achieve the sustainability goal in the plastic industry. However, due to the weakness of bioplastics, the applications of bioplastics are limited in various industries. Some reinforcing materials could improve the biodegradability of the bioplastics, but using non-biodegradable or synthetic materials in reinforcing bioplastics could also reduce the biodegradability (Hubbe et al., 2020). Furthermore, problems might occur with the use of biodegradable plastics such as high cost, brittleness and thermal instability (Nafisa et al., 2015; Shivam et al., 2016).

Nonetheless, as the industry nowadays tends to develop 'green' technologies, using sustainable and biodegradable materials are preferable in terms of reducing carbon footprints, preserving non-renewable resources (fossil-fuel resources), having less toxicity, and reducing plastic wastes, which lead to pollution that is harmful to the environment and human beings (Shamsuddin et al., 2017; Sidek et al., 2019), energy efficiency, partly based on natural feedstock and eco-safety (Reddy et al., 2013; Arikan., et al., 2015; Shamsuddin et al., 2017).

Bioplastics (i.e., PLA, bio-PE, bio-PP, and bio-PBS) and reinforcing materials (i.e., cellulosic fibre, cellulose and starch) could be obtained from various feedstocks and biomass, which further reduce and utilize the wastage of various functional materials (Shamsuddin et al., 2017).

CHAPTER THREE

MATERIALS AND METHODS

3.1 MATERIALS

3.1.1 Chemicals/Materials

All the materials used in the study were produced from natural source; this include ripe plantain peel powder, cassava starch, glycerol, acetic acid (vinegar) and eggshell powder.

3.1.2 Apparatus and Equipment.

Digital weighing balance-Mettler PT 320 (Mettler-Wagen, Switzerland), Digital Microscope (BXAW-AX-BC, China), Fourier transform infrared spectroscopy (FTIR-4600, Jasco Corp., Japan), Gas chromatography with flame ionization detector GC-FID (GC 7890, Japan), GC-MS ((Agilent 7890B) equipped with a mass spectrometry detection system (Agilent 5977A- MSD) were used . Scanning electron microscope (SEM; JSM-6360LA; JEOL Ltd., Japan)TGA analyzer (Mettler Toledo TGA/DSC1 simultaneous analyzer).Universal tester (HAIDA, International equipment CO. LTD, CHINA) were also used in the study.

3.2 METHODS

3.2.1 Collection of plantain peels, palm oil, cassava roots, pineapples and eggshells

The Ripe plantain peels were obtained from a Cafeteria (Buka 9) in Federal University of Technology, Owerri (FUTO) market (Longitude $6^0$59'E and Latitude $5^0$23'N). The palm oil used in the study was purchased from Ihiagwa market (Longitude $7^0$01'E and Latitude $5^0$24'N). The cassava root used in the study were harvested from the farm in Federal University of Technology, Owerri (FUTO) market (Longitude $6^0$59'E and Latitude $5^0$22'N). The Fresh egg shells were obtained from a Restaurant (Lala Meshai Spot) in Federal University of Technology, Owerri (Longitude $6^0$59'E and Latitude $5^0$23'N). The pineapple peels were collected from road side fruit sellers in Ihiagwa market.The picture of the ripe plantain peel is shown in plate 1 (Appendix**,** page 139)

3.2.2 Experimental Design

I. Collected plantain peels, sundried for one month, grounded into powder and characterized using FTIR, GC-MS and GC-FID.

II. Collected cassava root, peeled, grated, filtered of the slurry and sundried the cassava starch and characterized using FTIR and GC-MS

III. Collected fresh eggshells, sundried and grounded into powder and characterized using FTIR and GC-MS.

IV. Collected palm oil, bleached to remove colour, reacted with NaOH to form soap, filtered to obtain glycerol and characterized using FTIR and GC-MS.

V. Collected pineapple peels, washed, fermented with yeast and sugar for 14 days to obtain alcohol, further fermented to acetic acid (vinegar) and characterized using FTIR and GC-MS.

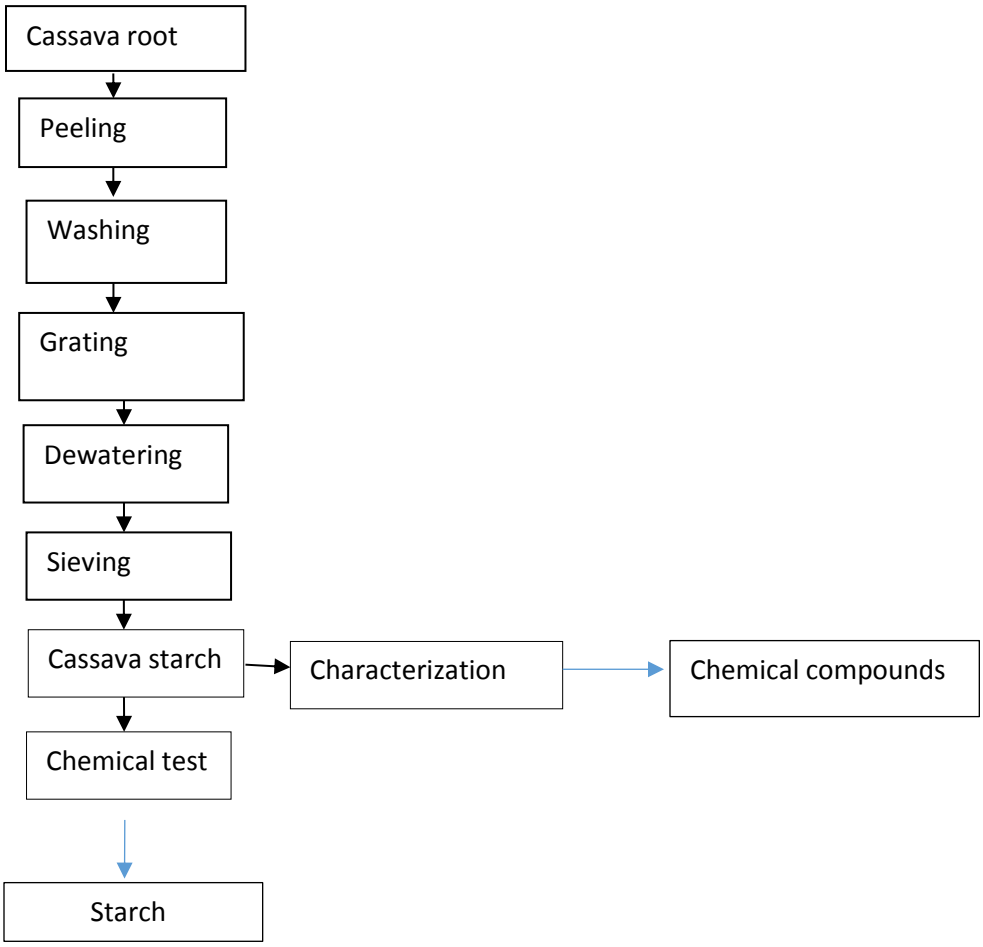


Figure 3.1: Flow chart for production of cassava starch

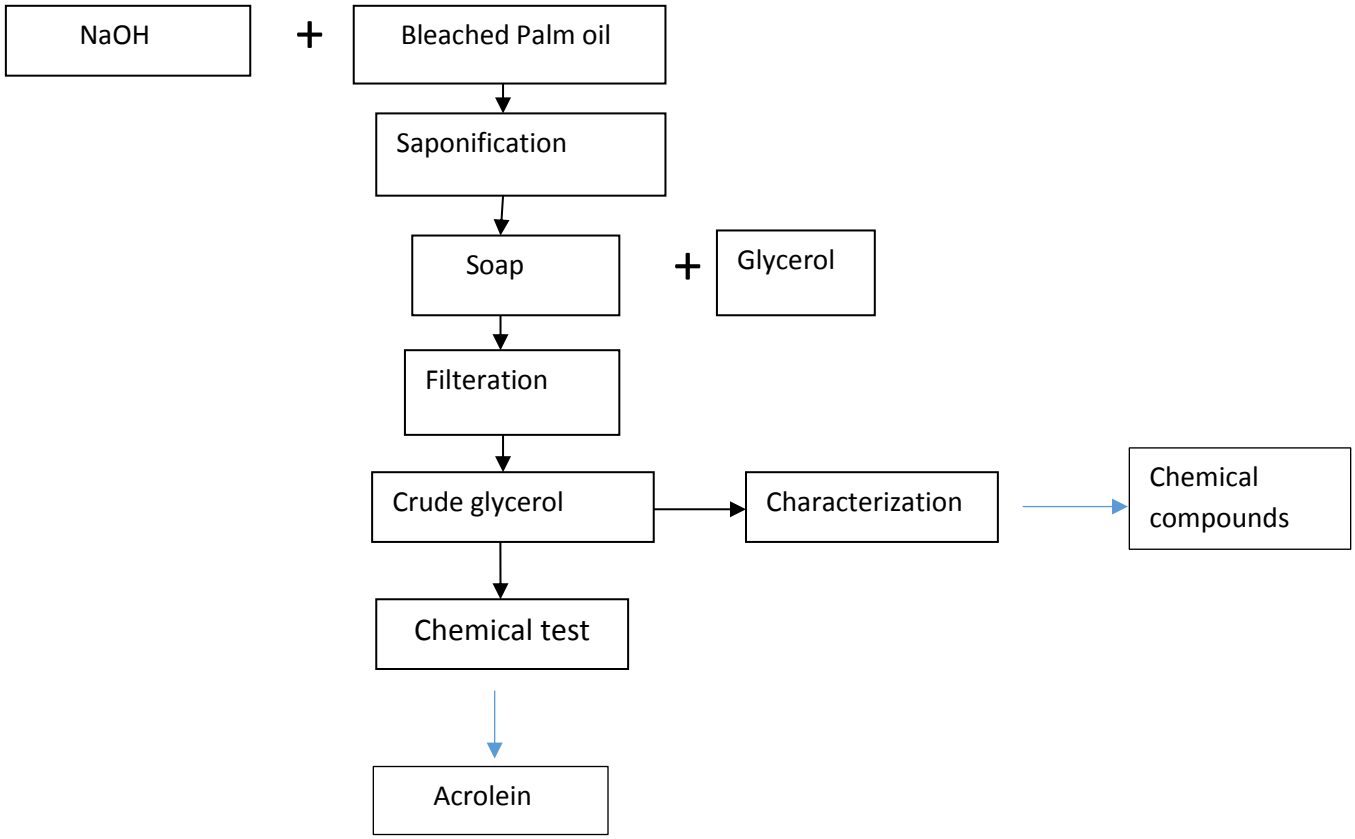


Figure 3.2: Flow chart for production of crude glycerol

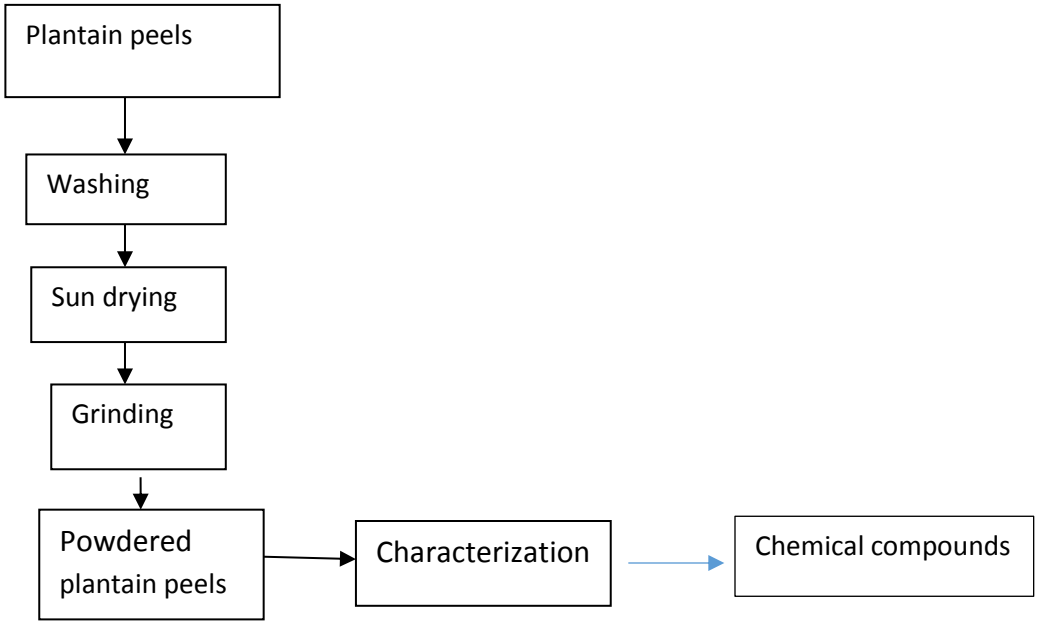
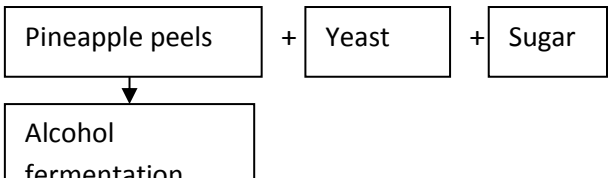


Figure 3.3: Flowchart for production of powdered plantain peels



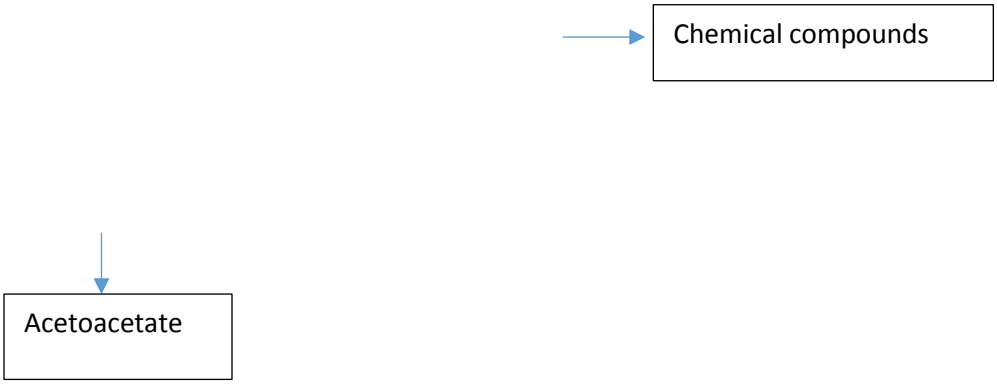
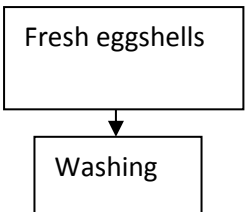


Figure 3.4: Flowchart for production of vinegar



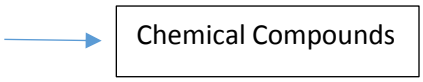


Figure 3.5: Flow chart for production of powdered eggshell

3.2.3 Treatment of plantain peels

The plantain peels were treated based on the modified method of Jachayandra and Vinay (2016). The ripe plantain peels were washed and weighed (2kg) and cut into pieces. They were sun dried for one month (to prolong the shelf life of the biofilm) and further grounded in a commercial blender to obtain a fine powder (30g). The picture of the grounded plantain peels is shown in plate 2 (Appendix**,** page 139).

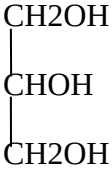
3.2.4 Preparation of crude glycerol

Crude glycerol was produced according to the method described by (Unilever, 1964). Palm oil (300 ml) were bleached in a hot air (the palm oil was heated for 5hr using firewood until the colour changed to light yellow when dropped on a white paper). One hundred millilitres (100ml) of the bleached Palm oil was taken into 200 ml beaker, and heated for 5min. 170ml of 60% NaOH was added and stirred while heating. The soap formed was prepared in the ratio of (1:1). It was cooled after 5min. Then, 25 g NaCl was added and stirred to precipitate the soap in solid form. The soap was filtered with filter paper to obtain the glycerol (58.82%) v/v. The picture of the bleached palm oil and the crude glycerol is shown in plate 3 and plate 4 (Appendix, page 139).

$$\% \text{ Glycerol} = \frac{\text{weight of glycerol + NaOH}}{\text{Weight of glycerol}} \times 100 \qquad (3.1)$$

3.2.4.1Test for glycerol

Potassium hydrogen bisuphate (1g) was added to a sample of 1ml glycerol. Heat was applied to the sample slowly on a magnetic stirrer. A yellowish brown coloration and a pungent smell was observed indicating the formation of acrylic aldehyde (acrolein) due to the presence of glycerol (Unilever, 1964).



Structure of glycerol

3.2.5 Preparation of Cassava Starch

The cassava root (200g) were washed, peeled and grated using hand grater. Then, one hundred and fifty milliliter (150ml) of water was added to the mash and stirred. The mash was filtered to obtain a slurry. The starch was prepared in the ratio of 1:1.The slurry was left to stand for 24h. It was further decanted to obtain the starch. The starch was sun dried for 24 h to obtain a fine powder (35 g) (Kamaljit, Preeti, and Hira 2016). The picture of the cassava root and cassava starch is shown in plate 5 and plate 6 (Appendix, page139).

3.2.5.1 Test on Cassava starch

The cassava starch was weighed (0.27g) into a test tube.Then 5ml of distilled water was added and stirred. To it, 3 drops of 5% potassium iodide solution was added, the result was a blue black colour indicating the presence of starch (Kamaljit et al., 2016).

3.2.6 Preparation of Acetic Acid (Vinegar).

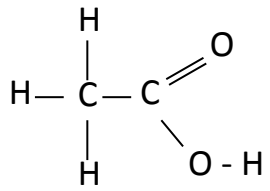
The vinegar was produced following the modified method of (De Ory, Romero, and Cantero, 1999). The Pineapple peels were washed and cut into pieces. It was weighed (500g) and taken into 1L can. Then, 800ml of water was added, sugar (42g) was added and stirred. *Saccharomyces cerevisae* (40g)(industrial yeast) was added and stirred. The can was covered with a breathable cloth and left to undergo alcoholic fermentation for 14 days. The fermented pineapple was sieved on the 15th day. Then it was exposed to air (natural aeration) for 6 weeks to allow *Acetobacter aceti* in the environment to cause acetic acid fermentation to take place. The vinegar was prepared in the ratio of 1:1:17:18. The vinegar was consequently purified by simple distillation to obtain a pure acetic acid. (62%) w/v. The picture of the pineapple peels and acetic acid (vinegar) is shown in plate 7 and plate 8 (Appendix, page 140).

Cal: wt of solute / wt of solution ×100

3.2.6.1Test on acetic acid (vinegar)

The pH of the acetic acid was determined (3.5). The acetic acid (2mls) was taken into a test tube. Then, 2mls of ethyl alcohol was added and 3drops of concentrated sulphuric acid were added and warmed in a hot water bath for 5min. The solution was poured into a test tube half filled with

water. The result was a sweet smell indicating the formation of ethyl acetate due to the presence of carboxylic acid (De Ory et al., 1999).



Structure of Acetic Acid

3.2.7 Treatment of eggshells

Eggshells were washed, sun dried and 80g was ground in a commercial blender to a fine powder (Syarifah et al., 2018). The picture of the eggshells and the eggshell powder is shown in Plate 9 and plate 10 (Appendix, page 140).

3.2.8 Characterization of synthesized raw materials.

The synthesized raw materials were characterized using Fourier transform infrared spectroscopy and Gas Chromatography Mass Spectrometry.

3.2.8.1 Fourier transform infrared spectroscopy analysis of the raw materials

The sample (eggshells, cassava starch, vinegar, glycerol and plantain peels) (0.5g) was mixed with 0.5g of kbr (potassium bromide) after which 1ml of nujol (a solvent for preparation of sample by Buck M530 IR-spectrophotometer) was introduced into the sample with aid of a syringe to form a paste before introducing it into the instrument sample mould and allowed to scan at a wavelength of 600-4000nm to obtain its spectra wavelength (jumare, Magashi, Rabah, Sokoto, and Hisbullahi 2019).

3.2.8.2 Gas Chromatography Mass Spectrometry analysis of the raw materials

To identify the constituents of the raw materials, their individual mass spectral peak values were compared with the database of National Institute of Science and Technology, 2014 (NIST, 2014). Then the phytochemicals were identified after comparing the unknown peak value and chromatogram from the GC-MS against the known chromatogram, peak value from the NIST library database. Subsequently, the details about the retention time and percentage content were also obtained (Mie-ling et al., 2003).

3.2.8.3 Toxicity test of synthesized raw materials

The analysis of PAHS in the raw materials was performed on a Gas chromatography equipped with a flame ionization detector. A RESTEK 15 meter MXT-1 column (15m x 250um x 0.15um) was used. The injector temperature was $280^{o}C$ with splitless injection of 2 ul of sample and a linear velocity of $30cms^{-1}$, Helium 5.0 pa.s was the carrier gas with a flow rate of 40ml/min. The oven operated initially at $200^{0}C$, it was heated to $330^{0}C$ at a rate of $3^{0}c\ min^{-1}$ and was kept at this temperature for 5min. the detector operated at a temperature of $320^{0}C$. Polycyclic aromatic hydrocarbons (PAHS) were determined by the ratio between the area and mass of internal standard and the area of the identified compounds. The concentration of the different PAHS expressed in ug/ml and percentage. It was conducted according to the method described by Okparamma and Muazen, (2013).

3.2.9 Production of biofilm

Two biodegradable biofilm were synthesized. Plantain peel based biodegradable biofilm (P-BF) and another biodegradable film without plantain peel (NP-BF).

3.2.9.1 Synthesis of plantain-based biofilm (P-BF).

The plantain-based biofilm was produced according to a modified method of (Divyar and Rachael, 2021). Powdered plantain peel (8g) was weighed into a 200ml beaker. Cassava starch (10g) was added along with 4g powdered eggshell. The crude glycerol (3ml) was added along with 3ml acetic acid (vinegar) and 10mls of water and the mixture was stirred vigorously. The materials were added in the ratio of 1:1:2:3:3. It was warmed gently on heat ($50^{0}C$) while stirring for 5min until it formed a gel. The mixture was poured on a tray and sun-dried for 3 days. The biodegradable biofilm was further dried at room temperature for 7 days before testing.

The biofilm produced weighed 38.05g. The picture of the plantain peel based biofilm (P-BF) is shown inplate11 (Appendix, page 141).

3.2.9.2 Synthesis of non- plantain peel biodegradable plastics (NP-BF)

The non-plantain biofilm was produced according to the modified method of Divyar and Rachael, (2021). Cassava starch (10g) was taken into a 25ml beaker, 5ml of water was added to the starch and stirred, along with 4ml of acetic acid (vinegar) and 2 ml of glycerol. The materials were added in the ratio of 1:1:2:4. The mixture was stirred vigorously while on heat (50^0C) for 4mins. It was poured into a plate and left to dry for 10 days at room temperature before testing. The biofilm produced weighed 15.11g. The picture of the non-plantain peel based biofilm (NP-BF) is shown in plate 12 (Appendix, page 141).

3.2.10. Characterization of biodegradable films

The synthesized biodegradable films were characterized using Fourier transform infrared spectroscopy.

3.2.10.1. Morphology study

The morphology study of the prepared samples was analyzed using a Digital Microscope and a Scanning Electron Microscope (Vishitta, Lenita, Sadaf, Harshitha and Sanjay, 2023).

3.2.10.2 Biodegradability test

The degradability test of the plastic was conducted following the method of Jayachandra et al., (2016).The biodegradability behaviour test was carried out to determine the biodegradability of bioplastics using compost soil. P-BF (2.98g) and NP-BF (1.01g) sample were weighed and buried in two glass jar containing soil sample. Water was added to the soil to enhance microbial activity. The degradation was checked by weighing the samples after every 6 days interval for 12 days.The following equation was used to calculate weight loss;

W_t loss (%) $=(W_o-W_t)/Wo$ x100 (3.2)

Where W_o is the weight after being buried in the soil.

W_t is the weight before being buried in the soil.

3.10.3 Water absorption test

The water absorption test was conducted following the method of Jayachandra et al., (2016**).** Water absorption evaluates the durability and suitability of bioplastics for moisture or water-related applications. A sample of the synthesized biodegradable plastics (P-BF and N-BF) was weighed. The samples were immersed in a 60ml distilled water for 24h, and their weight measured afterwards to determine the amount of water absorbed. This test provides insights into the material's resistance to water infiltration and its dimensional changes or expansion when exposed to moisture.The test was carried out in triplicate. The water absorption was calculated thus;

$$\text{Water Absorption (\%)} = [W_t - W_o) / W_o] \times 100 \quad (3.3)$$

W_t is the weight before being immersed in water.

W_o is the weight after being immersed in water.

3.10.4 Swelling test

The swelling test was conducted following the method of Jayachandra, Yaradoddi and Vinay (2016**).** Each sample (2.01g and 2.02g) of the synthesized biofilms (P-BF and NP-BF) was weighed. The samples were dipped in a 200ml beaker containing different solvent; 60ml methanol and chloroform and left for 24 h. The test was carried out in duplicate and the result was calculated thus;

$$\text{Swelling (\%)} = (W_o - W_t)/W_o \times 100 \quad (3.4)$$

Where W_o is the weight after being immersed in water.

W_t is the weight before being immersed in water.

3.10.5. Solubility test

The solubility test was conducted based on the method of **(**Jayachandra et al., 2016**).** Solubility is the main properties to check whether the synthesized bioplastic material is sustainable or not. (May, Myo and Zin, 2019). A sample (2.01g and 2.02g) of the synthesized biodegradable biofilm (P-BF and N-BF) was weighed. The samples were dipped in different solvent (ethanol, sulfuric acid and acetone) and left for 24h. The test was carried out in duplicate and the result was calculated thus;

$$\text{Solubility (\%)} = (W_o - W_t)/W_o \times 100 \quad (3.5)$$

Where W_o is the weight after being immersed in water.

W_t is the weight before being immersed in solvent.

3.10.6. Mechanical test

To measure the mechanical properties, the formed film sample was sliced into size of about 5 × 5 mm. The mechanical tests were carried out by performing ultimate tensile, flexural, % elongation and hardness tests to the sliced samples (Budiman Triawan F., Adziman and Nurprasetio 2017; Sukrawan, Hamdani and Mardani, 2019**).**

3.2.10.6.1. Ultimate tensile test

This is a measure of the maximum stress a material can withstand without breaking or falling under tension. It's a fundamental property used to predict how a material or a component will behave under load. The tensile properties were tested out on a Control Universal Testing Machine at a crosshead speed of 2 kg/min with the load cell of 50 kN. It was conducted according to Budiman et al., (2017).

Ultimate Tensile strength;

$$\sigma_{max} = \frac{P_{max}}{A_0} \quad (3.6)$$

Where P_{max} = maximum load,

A_0= original cross sectional area (mm^2).

3.10.6.2 Hardness test

The hardness Shore D tests were carried out by Mitutoyo Shore D hardness tester (Sukrawan et al., 2019).

3.2.10.6.3 % Elongation

Elongation at break is an extension of a material when tested tensile until finally fracture. It was conducted as described by Budiman et al., (2017). It was calculated thus;

$$\% \text{ elongation} = \frac{L - L_0}{L_0} \times 100 \qquad (3.7)$$

Where L_0 is the final elongation of the material (mm), and *L* is the initial length of the material (mm).

3.2.10.6.4. Flexural strength/ bending test

Flexural strength refers to how much a material will take before it tears, ruptures, breaks or permanently bends, i.e. yields. This method is used to determine the strength and dimensional changes in the properties of plastics when subjected to tensile, compressive and shear stresses. This was determined according to Budiman et al., (2017). It was calculated thus;

Flexural strength:

$$\sigma = \frac{3FL}{2bd^2} \qquad (3.8)$$

Where F is the maximum load at the fracture point (N), L is the length of the support span

b is width

d is thickness

3.2.10.6.5 TGA analysis

Thermogravimetric analysis is a method used to study the reaction of thermal decomposition (ASTM, 2024) between weight change and temperature which are lost due to the effect of temperature on the material (zhaosheng et al., 2008; Ahmad, Anuar, and Yusof, 2011). The result of the thermal analysis is in the form of a curve called a thermogram. Thermal decomposition is a process of changing the form of a sample into a simpler form (Sukarni et al., 2015).The thermal decomposition of biofilm was analyzed using a thermogravimetry (Mettler Toledo TGA/DSC1 simultaneous analyzer). The 10 mg sample of (P-BF) and (N-BF) were heated from 29.92^0C (room temperature) to 500^0c and 22.17^0C (room temperature) to 500^0c respectively with a heating rate of 10^oC/min in the presence air with a flow rate of 50 mL/min. The thermogravimetric (TG), derivative thermogravimetric (DTG) can identify the thermal decomposition that occurs in biofilm through the loss of weight. It was conducted following the method described by Nanang, Eddy, Heru, and Sukarni, (2017).

3.2.11. Toxicity Test

3.2.11.1. Toxicity Test of soil containing biodegraded biofilm

The toxicity test was conducted by determining the presence of PAHS(Polycyclic aromatic hydrocarbons) in the soil samples containing synthesized biodegraded films using GC-FID(Gas chromatography flame atomization detector, BUCK M910) Buck 530 gas chromatograph equipped with an on – column, automatic injector, Flame Ionization detector, HP 88 capillary column (100m x 0.25µm film thickness). It was conducted as described by Hu et al., (2014)

3.2.12. Statistical Analysis

The obtained data were analysed statistically using student T- test of significance and Analysis of variance (Anova). Values were considered significant at $p< 0.05$.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 RESULTS

4.1.1 Characterization of synthesized raw materials

4.1.1.1 Fourier transform infrared spectroscopy glycerol

Table 4.1 shows the FTIR results for the crude glycerol**,** peak values around 1102.833cm^{-1} and 1298.699cm^{-1} was assigned to CO stretching vibration of ether compound. The absorbance around 1393.905cm^{-1} was assigned to C=C anti-symmetric vibration of alkene compound. Medium band around 1620.021cm^{-1} was assigned to N-H stretching vibration of 1^0 amine compound. Spectra height around 1837.330cm^{-1} corresponds to CO stretching vibration of cyclic ester compound. Wavelength around 2045.241cm^{-1} and 2277.092cm^{-1} were assigned to COO stretching vibration of carboxylic acid compounds respectively. The peak around 2451.037cm^{-1} and 2594.904cm^{-1} were assigned to CN stretching vibration of nitrile compound respectively. The weak bands around 2682.750cm^{-1} and 2919.177cm^{-1} were both assigned to C-H stretching vibration of methylene compound respectively. Strong bands around 3024.229$cm^{-1,}$ 3165.812cm^{-1} , 3275.510cm^{-1} , 3558.926cm^{-1} , 3704.989cm^{-1} and 3704.989cm^{-1} were assigned to OH stretching vibration of 1^0 & 3^0 alcohols respectively

Table 4.1: Result of Fourier transform infrared spectroscopy (FTIR) analysis of glycerol

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	1102.833	R-O-R	Ether CO symmetric stretch
2	1298.699	R-O-R	Ether CO symmetric stretch
3	1393.905	$H_2C=CH$	Ethene CH anti-symmetric stretch
4	1620.021	RNH_3	1^0 amine NH stretch
5	1837.330	R-O-R	Cyclic ester CO stretch
6	2045.241	RCOOH	Carboxylic acid COO stretch
7	2277.092	RCOOH	Carboxylic acid COO stretch
8	2451.037	R-C≡N	Nitriles CN antisymmetric stretch
9	2594.904	R-C≡N	Nitriles CN antisymmetric stretch
10	2682.750	CH_2	Methylene CH symmetric stretch
11	2919.177	CH_2	Methylene CH symmetric stretch
12	3024.229	RCHOH	1^0 alcohol OH symmetric stretch
13	3165.812	RCHOH	1^0 alcohol OH symmetric stretch
14	3275.510	RCHOH	1^0 alcohol OH symmetric stretch
15	3558.926	R_3CHOH	3^0 alcohol OH symmetric stretch
16	3704.989	R_3CHOH	3^0 alcohol OH symmetric stretch
17	3809.275	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.1.2: Result of Fourier transform infrared spectroscopy (FTIR) analysis of vinegar

Table 4.2 shows FTIR results for Vinegar**,** the absorbance around 11021.292cm^{-1} was assigned to CO stretching vibration of ether compound. The peak values around 1322.171cm^{-1} and 1437.724 cm^{-1} were assigned to C=C stretching vibration of ethene compound respectively. The medium band around 1626.265cm^{-1} were assigned to N-H stretching vibration of 1^0 amine compound. The peak value around 1889.889cm^{-1} was assigned to CO stretching vibration of cyclic ester compound. Absorbance around 2026.817cm^{-1} and 2113.309cm^{-1} were assigned to COO stretching vibration of carboxylic acid respectively whereas the peak located at 2201.730cm^{-1} was assigned to CO stretching vibration of carbonyl compound. The peak around 2449.360cm^{-1} was assignd to CN stretching vibration of nitrile compound. The weak peak values located around 2619.551cm^{-1}, 2742.866cm^{-1} and 2911.437cm^{-1} were all assigned to C-H stretching vibration of methylene compound respectively. Strong bands wavelength located around 3184.373cm^{-1} , 3374.118cm^{-1} , 3575.131cm^{-1} and 3803.590cm^{-1} were assigned to OH stretching vibration of 1^0. 2^0 & 3^0, respectively.

Table 4.2: Result of Fourier transform infrared spectroscopy (FTIR) analysis vinegar

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	11021.292	$H_2C=CH$	Ethene CH anti-symmetric stretch
2	1322.171	$H_2C=CH$	Ethene CH anti-symmetric stretch
3	1437.724	$H_2C=CH$	Ethene CH anti-symmetric stretch
4	1626.265	RNH_3	1^0 amine NH stretch
5	1889.889	R-O-R	Cyclic ester CO stretch
6	2026.817	RCOOH	Carboxylic acid COO stretch
7	2113.309	RCOOH	Carboxylic acid COO stretch
8	2201.730	$R_2C=O$	Carbonyl compound CO stretch
9	2449.360	R-C≡N	Nitriles CN anti-symmetric stretch
10	2619.551	CH_2	Methylene CH symmetric stretch
11	2742.866	CH_2	Methylene CH symmetric stretch
12	2911.437	CH_2	Methylene CH symmetric stretch
13	3184.373	RCHOH	1^0 alcohol OH symmetric stretch
14	3374.118	R_2CHOH	2^0 alcohol OH symmetric stretch
15	3575.131	R_3CHOH	3^0 alcohol OH symmetric stretch
16	3803.590	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.1.3: Result of Fourier transform infrared spectroscopy (FTIR) analysis cassava starch

Table 4.3 shows FTIR results for cassava starch, the peak value around 1058.841cm^{-1} was assigned to CO stretching vibration of ether compound while the peak value around 1360.767cm^{-1} was assigned to C=C stretching vibration of ethene compound. The medium band around 1624.580cm^{-1} and 3405.882cm^{-1} were assigned to N-H stretching vibration of 1^0& 2^0 amine compound respectively. The peak value around 1885.184$cm^{-1}$was assigned to CO stretching vibration of cyclic ester compound. Absorbance around 2054.885cm^{-1} and 2163.940cm^{-1} were assigned to COO stretching vibration of carboxylic acid respectively whereas the peaks located around 2624.921cm^{-1} and 2929.717cm^{-1} were both assigned to C-H stretching vibration of methylene compounds. The peak values located around 3054.420cm^{-1}, 3175.895cm^{-1} and 3516.919cm^{-1} were all assigned to OH stretching vibration of 1^0, 2^0& 3^0 alcohols respectively.

Table 4.3: Result of Fourier transform infrared spectroscopy (FTIR) analysis of cassava starch

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	1058.841	R-O-R	Ether CO symmetric stretch
2	1360.767	$H_2C=CH$	Ethene C=C anti-symmetric stretch
3	1624.580	RNH_3	1^0 amine NH stretch
4	1885.184	R-O-R	Cyclic ester CO stretch
5	2054.885	RCOOH	Carboxylic acid COO stretch
6	2163.940	RCOOH	Carboxylic acid COO stretch
7	2439.478	R-C≡N	Nitriles CN anti-symmetric stretch
8	2540.328	R-C≡N	Nitriles CN anti-symmetric stretch
9	2624.921	CH_2	MethyleneCH symmetric stretch
10	2790.356	CH_2	Methylene CH symmetric stretch
11	2929.717	CH_2	Methylene CH symmetric stretch
12	3054.420	RCHOH	1^0 alcohol OH symmetric stretch
13	3175.895	RCHOH	1^0 alcohol OH symmetric stretch
14	3405.882	R_2NH	2^0 amine NH symmetric stretch
15	3516.919	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.1.4: Result of Fourier transform infrared spectroscopy (FTIR) analysis of eggshell

Table 4.4 shows the results of FTIR analysis for eggshell, the peak values around 1383.668cm^{-1} and 1467.005cm^{-1}were assigned to C=C stretching vibration of ethene compound. The medium band around 1623.465cm^{-1} and 3457.769cm^{-1} were assigned to N-H stretching vibration of 1^0amine compound. The peak value haround 1830.269cm^{-1} and 1988.089cm^{-1} were assigned to CO stretching vibration of cyclic ester compound respectively. Absorbance around 2120.197cm^{-1} was assigned to COO stretching vibration of carboxylic acid respectively whereas the peaks located around 2746.925cm^{-1} and 2904.522cm^{-1} were both assigned to C-H stretching vibration of methylene compounds. The peak values located around 3008.660cm^{-1}, 3242.667cm^{-1}, 3345.205cm^{-1}, 3700.921cm^{-1}, and 3839.897cm^{-1} were all assigned to OH stretching vibration of 1^0, 2^0& 3^0 alcohols respectively.

Table 4.4: Result of Fourier transform infrared spectroscopy (FTIR) analysis of eggshell

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	1383.668	R-O-R	Ether CO symmetric stretch
2	1467.005	$H_2C=CH$	Ethene C=C anti-symmetric stretch
3	1623.465	RNH_3	1^0 amine NH stretch
4	1863.428	R-O-R	Cyclic ester CO stretch
5	2000.617	RCOOH	Carboxylic acid COO stretch
6	2179.558	RCOOH	Carboxylic acid COO stretch
7	2272.479	RCOOH	Carboxylic acid COO stretch
8	2469.356	R-C≡N	Nitriles CN anti-symmetric stretch
9	2746.925	CH_2	Methylene CH symmetric stretch
10	2904.522	CH_2	Methylene CH symmetric stretch
11	3008.660	RCHOH	1^0 alcohol OH symmetric stretch
12	3242.667	RCHOH	1^0 alcohol OH symmetric stretch
13	3345.205	R_2CHOH	2^0 alcohol OH symmetric stretch
14	3457.769	R_2NH	2^0 amine NH stretch
15	3700.921	R_3CHOH	3^0 alcohol OH symmetric stretch
16	3839.897	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.1.5: Result of Fourier transform infrared spectroscopy (FTIR) analysis of plantain peels

Table 4.5 shows the result of FTIR analysis forplantain peels,the peak values around 1328.304cm^{-1} and 1433.162cm^{-1} were assigned to C=C stretching vibration of ethene compound. The medium bands located around 1620.894cm^{-1} and 1667.273cm^{-1} were assigned to N-H stretching vibration of 1^0 amine compound respectively. The peaks around 2024.931cm^{-1}, 2086.796cm^{-1} and 2214.360cm^{-1} were all assigned to COO stretching vibration of carboxylic acid respectively whereas the peaks located around 2513.176cm^{-1} was assigned to CN stretching vibration of nitrile compound. The weak bands located around 2627.409cm^{-1}, 2750.631cm^{-1} and 2849.984cm^{-1} were all assigned to C-H stretching vibration of methylene compounds. The strong bands located around 3083.774cm^{-1}, 3190.627cm^{-1}, 3350.071cm^{-1} and 3661.209cm^{-1} were all assigned to 0H stretching vibration of 1^0, 2^0& 3^0 alcohols respectively.

Table 4.5: Result of Fourier transform infrared spectroscopy (FTIR) analysis of plantain peels

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	1328.304	$H_2C=CH$	Ethene C=C anti-symmetric stretch
2	1433.162	$H_2C=CH$	Ethene C=C anti-symmetric stretch
3	1620.894	RNH_3	1^0 amine NH stretch
4	1667.273	RNH_3	1^0 amine NH stretch
5	2024.931	RCOOH	Carboxylic acid COO stretch
6	2086.796	RCOOH	Carboxylic acid COO stretch
7	2214.360	RCOOH	Carboxylic acid COO stretch
8	2513.176	R-C≡N	Nitriles CN anti-symmetric stretch
9	2627.409	CH_2	Methylene CH symmetric stretch
10	2750.631	CH_2	Methylene CH symmetric stretch
11	2849.984	CH_2	Methylene CH symmetric stretch
12	3083.774	RCHOH	1^0 alcohol OH symmetric stretch
13	3190.627	RCHOH	1^0 alcohol OH symmetric stretch
14	3350.071	R_2CHOH	2^0 alcohol OH symmetric stretch
15	3661.209	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.2 Result of Gas chromatography-mass spectrometry (GCMS) analysis of cassava starch

4.1.2.1: Chemical compounds in cassava starch

The GC-MS analysis of cassava starch showed 15 peaks. The peaks indicate the presence of 15 compounds. The compounds identified are; Anthiaergostan-5,7,9,22-tetraen-3-ol, 7-Dehydrodiosgenin. (5.89%) with a retention time of 3.113RT, Benzoic acid, 4-(5,5-dimeth 4-(5,5-dimethyl-1,3- dioxan-2-yl)-, methyl ester (4.12%) with retention time of 3.799., 1H-1,2,4-Triazole, 3-methyl-5-(methylthio)-Dihexyl monoselenide(4.216%) with the retention time of 9.98, Dihexyl monoselenide, 1-Dodecanol(7.731%) with a retention time of 4.17., Dihexyl monoselenide (11.54%) with a retention time of 9.023RT. Dihexyl monoselenide, 3-Trifluoroacetoxypentadecane. 2,1,3-benzoselenadiazole, 5,6-dichloroThiophene, 2-(methylselenyl)-5-(propylthio)-2,1,3- (11.34%), with a retention time of 10.177, Benzoselenadiazole, 4,6-dichloro- (6.85%) with a retention time of 10.920, Mercury, chloromethyl-Pyrimidine, 5-bromo-2,4-bis(methyl thio)-N-(3-Nitro-thiobenzoyl)-morpholine(4.12%) with retention time of 11.189.,N-[3-Aminophenyl]-1-piperidinecarbothioamide, Silane, 9-anthracenyltrimethyl-Benzothiophene-3-carboxamide, 4,5,6,7-tetrahydro-2-amino-6-tert-butyl- (4.30%) 12. 384 Methaqualone(8.71%) with a retention time of 13.018RT, Mercury, chloromethyl-2,1,3-Benzoselenadiazole, 4,6-dichloro-2-Pentene, 1-(pentyloxy)-, (E)- (6.43%) with a retention time of 13.892RT, 3-(2-Hydroxy-6-methylphenyl)-4(3H)-quinazolinone 5-Methyl-6-nitro-2-phenyl-Spiro[(5-bromoacenaphthen-1-one)-2 , 2'-(5',5'-dimethyl-1',3'-dioxane (7.19%) with a retention time of 14.390, Mercury, chloromethyl-Pyrimidine, 5-bromo-2,4-bis(methyl thio)-Phenol, 2,4-dibromo- (4.67%) with a retention time of 14.858, Mercury, chloromethyl-2,1,3-benzoselenadiazole, 5,6-dichloro, N-Nitroso-2,4,4-trimethyloxazolidine (5.37%) with a retention time of 18.08RT, Methaqualone , N-[3-Aminophenyl]-1-piperidinecarbothioamide (5.34%) with a retention time of 19.225. Among all the compounds identified, the maximum retention time was shown by Methaqualone (19.255 RT), and the lowest retention time was shown by anthiergostan (3.11 RT). Also, the most aboundant compounds was identified as Dihexyl monoselenide, 2-Amino-oxazole,Trichloroacetic acid, decyl ester(11.54%),Dihexyl monoselenide 3-TrifluoroacetoxypentadecanePentadecane, 8-methylene- (11.34%),1 H-1,2,4-Triazole, 3-methyl-

5-(methylthio)-Dihexylmonoselenide, 2-t-Butyl-5-methyl-10H-acridin-9-one(9.98%) and Methaqualone (8.71%).

Table 4.6: Chemical compounds in cassava starch

Peak	Compound	Retention time	Concentration %
1	Anthiaergostan-5,7,9,22-tetraen-3-ol, 7-Dehydrodiosgenin,	3.113	5.89
2	Benzoic acid, 4-(5,5-dimethyl-1,3dioxan-2-yl)-, methyl ester .	3.799	4.12
3	1 H-1,2,4-Triazole, 3-methyl-5-(methylthio)-Dihexylmonoselenide	4.216	9.98
4	Dihexyl monoselenide,1-Dodecanol	7.731	4.17
5	Dihexyl monoselenide	9.023	11.54
6	Dihexyl monoselenide 3-Trifluoroacetoxypentadecane	10.177	11.34
7	2,1,3-benzoselenadiazole, 5,6-dich loroThiophene, 2-(methylselenyl)-	10.920	6.85
8	Mercury, chloromethyl-Pyrimidine, 5-bromo-2,4-bis(methyl thio)-N-(3-Nitro-thiobenzoyl)-morpholine	11.189	4.12
9	N-[3-Aminophenyl]-1-piperidinecarbothioamide Silane	12.384	4.30
10	Methaqualone	13.018	8.71
11	Mercury, chloromethyl-2,1,3-Benzoselenadiazole	13.892	6.43
12	3-(2-Hydroxy-6-methylphenyl)-4(3H)-quinazolinone.	14.390	7.19
13	Mercury, chloromethyl-Pyrimidine.	14.858	4.67

14	Mercury, chloromethyl-2,1,3-benzoselenadiazole.	18.08	5.37
15	Methaqualone, N-[3-Aminophenyl]-1-piperidinecarbothioamide Pyrimidine,	19.225	5.33

4.1.2.2: Result of Gas Chromatography-Mass Spectrometry (GCMS) analysis of eggshells

The analysis of egg shells by GC-MS (table 4.7) showed 15 bioactive compounds which include; 5-Methyl-6-nitro-2-phenyl-1H-indol(8.68%) with a retention time of (4.210 RT). N-[3-Aminophenyl]-1-piperidinecarbothioamide(8.37%) with a retention time of 6.697RT, 3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde(7.26%) with a retention time of 7.69RT, Dihexyl monoselenide (5.91%) with a retention time of 9.103RT, N-[3-Aminophenyl]-1-piperidinecarbothioamide(4.73%) with a retention time of 9.829 RT, Dihexyl monoselenide Dodecane (5.62 %) with a retention time of 10.663 RT. 2-Methyl-1-tetradecene(4.90%) with a retention time of 11.246RT, 1-Hexene, 3,4-dimethyl-(Z)-Hex-2-ene (9.26%) with a retention time of 12.075 RT, 4-Piperidinemethanamine (6.95%) with a retention time of 12.635 RT. 1,2-Benzenedicarboxylic acid, monobutyl ester,(11.62%) with a retention time of 13.018 RT.Dihexyl monoselenide(4.68%) with a retention time of 14.321 RT, Cyclobutanone, 2-methyl- 2-Methyl-(6.89%) with a retention time of 14.544 RT, 1-Hexene with a retention time of 17.327RT, Chloroacetic acid, hexyl ester, (5.06 %) with a retention time of 18.236RT, 1-Pentene, 2-methyl-Cyclobutanone, 2-methylCyclobutanone (5.03%) with a retention time of 19.293 RT. However, 1,2-Benzenedicarboxylic acid, monobutyl ester, monodecyl ester was identified to be the most prevailing bioactive compounds (11.62%), followed by 1-Hexene(9.26%),N-[3-Aminophenyl]-1-piperidinecarbothioamide, 5-Methyl-6-nitro-2-phenyl-1H-indol,3-(2-Hydroxy-6-methylphenyl)-4(3H)-quinazolinone(8.37%).Other compounds was detected in minimal concentrations.

Table 4.7: Chemical compounds in eggshells

Peak	Compound	Retention time	Concentration %
1	5-Methyl-6-nitro-2-phenyl-1H-indole	4.210	8.68
2	N-[3-Aminophenyl]-1-piperidinecarbothioamide	6.697	8.37
3	3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde	7.691	7.26
4	Dihexyl monoselenide	9.103	5.91
5	N-[3-Aminophenyl]-1-piperidinecarbothioamide.	9.829	4.73
6	Dihexyl monoselenide Dodecane.	10.663	5.62
7	2-Methyl-1-tetradecene, 1-Hexene, 3,4-dimethyl-1-Pentene, 2-methyl-	11.246	4.90
8	1-Hexene, 3,4-dimethyl-(Z)-Hex-2-ene,	12.075	9.26
9	4-Piperidinemethanamine	12.635	6.95
10	1,2-Benzenedicarboxylic acid, monobutyl ester	13.018	11.62
11	.Dihexyl monoselenide	14.321	4.68
12	Cyclobutanone, 2-methyl	14.544	6.89
13	1-Hexene	17.327	5.05
14	Chloroacetic acid, hexyl ester	18.236	5.06
15	1-Pentene, 2-methyl-Cyclobutanone	19.239	5.03

4.1.2.3: Result of Gas Chromatography-Mass Spectrometry (GCMS) analysis of acetic Acid (Vinegar).

The compounds in the synthesized acetic acid (vinegar) was identified by GC-MS analysis as shown in (table 4.9). 15 bioactive compounds was identified which include; Methylene chloride (1.36 %) with a retention time of 3.365RT, Methylene chloride, (1.24%) with a retention time 4.022RT.Methylene chloride (1.24 %) with a retention time of 4.296 RT, Benzoic acid (17.04) with a retention time of 5.102 RT, Ethanol, 2-[2-(2-methoxyethoxy)ethoxy](3.11 %) with a retention time of 5.445RT. Carbonic acid, pentadecyl prop-1-en-2-yl ester(2.11%) with a retention time of 5.737RT,N-Methyl-3-piperidinecarboxamide, (1.52 %) with a retention time of 5.920RT. Acetic acid, dichloro-Methylene chloride(1.90 %) with a retention time of 6.405RT. Valeric acid, 3,5-dihydroxy-2,4-dimethyl-, .delta.-lactone(2.78%) with a retention time of 6.674RT, Tetradecane (3.49%) with a retention time of 6.971RT, N-Isopropyl-N-methyl aminoethyl-2- chloride (0.93%) with a retention time of 7.308RT. 2,6,10-Trimethyltridecane, (2.40%) with a retention time of 7.623RT, Ethanol, 2-[2-(2-butoxyethoxy)ethoxy]-(5.02%) with a retention time of 7.846RT. Pentadecane, Pentadecane (3.58%) with a retention time of 8.051RT. Quinoline, 2-phenyl(3.51%) with a retention time of 8.223RT. 2-Butenedioic acid, dibutyl ester, (7.51 %) with a retention time of 8.503RT., 11-Dodecen-1-ol difluoroacetate, (1.28 %) with a retention time of 9.246RT. Methoxyacetic acid, 2-tridecyl ester(0.95%) with a retention time of 9.492 RT. 2H-Inden-2-one, octahydro-3a-methyl-, trans-Naphthalene(1.35 %) with a retention time of 20 9.783 RT, Pentadecane, 2,6,10-trimethyl-Hexadecane, (2.00 %) with a retention time of 10.017 RT.Undec-10-ynoic acid, dodecyl ester (0.97 %) with a retention time of 10.212 RT., 1,1,1,5,7,7,7-Heptamethyl-3,3-bis(trimethylsiloxy)tetrasiloxane,(1.74 %) with a retention time 10.572RT. Octadecane, (1.13%) with a retention time of 10.926 RT. 1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester, (19.70%) with a retention time of 11.561 RT.Tetratriacontyl pentafluoropropionate(0.90%) with a retention time of 12.538RT.trans-13-Octadecenoic acid, methyl ester(2.38%) with a retention time of 13.292 RT(2.17 %) with a retention time of 15.407RT.1H-Naphtho[2,1-b]pyran-1-one,3-acetyl-7,8-dimethoxy-2-methyl-trans-4-Ethoxy-2',4' dimethoxychalcone,-(1.86 %) with a retention time of 17.379 RT, Decanedioic acid, bis(2-ethylhexyl) ester (2.52%) with a retention time of 18.196RT. Bis(2-methylpropyl) ester, 1,2 -Benzenedicarboxylic acid, Phthalic acid, 8-bromoctyl isobutyl ester, 1,2-Benzenedicarboxylic acid, butyl 2-methylpropyl ester was found to have the maximum

concentration (19.70). With N-Isopropyl-N-methyl aminoethyl-2- , Ethyl oxachloride having the lowest concentration (0.93%).

Table 4.8: Chemical compounds in acetic acid (vinegar)

Peak	Compound	Retention time	Concentration %
1	Methylene chloride	3.365	1.36
2	Methylene chloride	4.022	1.24
3	Methylene chloride	4.296	1.24
4	Benzoic acid	5.102	17.04
5	Ethanol, 2-[2-(2-methoxyethoxy)ethoxy	5.445	3.11
6	Carbonic acid, pentadecyl prop-1-en-2-yl ester	5.737	2.11
7	N-Methyl-3-piperidinecarboxamide	5.920	1.52
8	Acetic acid, dichloro-Methylene chloride	6.405	1.90
9	Valeric acid, 3,5-dihydroxy-2,4-dimethyl-, .delta.-lactone	6.674	2.87
10	Tetradecane	6.971	3.49
11	N-Isopropyl-N-methyl aminoethyl-2-chloride	7.30	0.93
12	2,6,10-Trimethyltridecane	7.623	2.40
13	Ethanol, 2-[2-(2-butoxyethoxy)ethoxy]-	7.846	5.02
14	Pentadecane	8.051	3.58
15	Quinoline, 2-phenyl-	8.22	3.51
16	2- Butenedioic acid,	8.503	7.51
17	Hexadecane	9.05	2.32
18	11-Dodecen-1-ol difluoroacetate	9.246	1.28

19	Methoxyacetic acid, 2-tridecyl ester	9.492	0.95
20	2-H-Inden-2-one, octahydro-3a-methyl-	9.783	1.35
21	Pentadecane, 2,6,10-trimethyl-Hexadecane	10.017	2.00
22	Undec-10-ynoic acid, dodecyl ester	10.212	0.97
23	1,1,1,5,7,7,7-Heptamethyl-3,3-bis(trimethylsiloxy)tetrasiloxane	10.572	1.74
24	Octadecane	10.926	1.13
25	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester	11.561	19.70
26	Tetratriacontyl pentafluoropropionate, .	12.538	0.90
27	trans-13-Octadecenoic acid, methyl ester	13.292	2.38
28	Decanedioic acid, bis(2-ethylhexyl) ester,	15.40	2.17
29	1H-Naphtho[2,1-b]pyran-1-one, 3-acetyl-7,8-dimethoxy-2-methyl-trans-4-Ethoxy-2',4'-dimethoxychalcone, 4H-1-Benzopyran-4-one,	17.379	1.86
30	Decanedioic acid, bis(2-ethylhexyl) ester,	18.196	2.52

4.1.2.4: Result of Gas Chromatography-Mass Spectrometry (GCMS) analysis of plantain peel powder

Fifteen compounds were identified in the Gc-ms analysis of ripe plantain peel powder as shown in (table 4.10) .The chemical compounds include: Dihexyl monoselenide, (5.87%) with a retention time of 4.210 RT. Dihexyl monoselenide 9.58%) with a retention time of 5.091 RT., Indan-1,3-dione, 2-(1,3-dimethyl-1H-pyrazol-4-ylmethylene)(6.94%) with a retention time of 5.651RT., N-(3-Nitro-thiobenzoyl)-morpholine (9.39%) with a retention time of 6.685RT. 3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde, (7.10%) with a retention time of 7.120RT. 3-(2-Hydroxy-6-methylphenyl)-2-methyl-4(3H)-quinazolinone (5.55%) with a retention time of 8.040RT, N-[3-Aminophenyl]-1-piperidinecarbothioamide(5.38 %) with a retention time of 8.680RT, N-(3-Nitro-thiobenzoyl)-morpholine -(2-Hydroxy-6-methylphenyl)-4(3H)-quinazolinone 3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde, (11.13%) with a retention time of 10.183RT, 3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde, (6.78%) with a retention time of 10.543RT, 1-(2-Bromo-2-chloroethenyl)-4-chlorobenzene(4.79%) with a retention time of 12.640RT, Silane, 9-anthracenyltrimethyl-Methaqualone (3.88%) with a retention time of 14.321RT. N4-Phenethylmorpholine-4-carbothioamide, (0.65%) with a retention time of 9.646RT.Mercury, chloromethyl-Pyrimidine (4.18%) with a retention time of 15.692RT, Mercury, chloromethyl-Pyrimidine, (3.98%) with a retention time 16.161RT, 2,1,3-Benzoselenadiazole, 4,6-dichloro-Silane, (5.30%) with a retention time of 16.910RT. Among the compounds identified the most prevailing compounds were N4-Phenethylmorpholine-4-carbothioamide (10.13%).

Table 4.9: Chemical compounds in plantain peels

Peak	Compound	Retention time	Concentration %
1	Dihexyl monoselenide	4.210	5.87
2	Dihexyl monoselenide	5.091	9.58
3	Indan-1,3-dione, 2-(1,3-dimethyl-1H-pyrazol-4-ylmethylene)	5.651	6.94
4	N-(3-Nitro-thiobenzoyl)-morpholine .	6.685	9.39
5	3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde.	7.120	7.10
6	3-(2-Hydroxy-6-methylphenyl)-2 methyl -4(3H)-quinazolinone,	8.040	5.55
7	N-[3-Aminophenyl]-1-piperidinecarbothioamide	8.680	5.38
8	N-(3-Nitro-thiobenzoyl)-morpholine.	10.183	11.3
9	3,5-Di-t-butyl-4-methoxy-1,4-dihydrobenzaldehyde,	10.543	6.78
10	1-(2-Bromo-2-chloroethenyl)-4-chlorobenzene	13.640	4.79
11	Silane, 9-anthracenyltrimethyl-Methaqualone.	14.321	3.88
12	N4-Phenethylmorpholine-4-carbothioamide.	15.115	10.13
13	Mercury, chloromethyl-Pyrimidine	15.692	4.18
14	Mercury, chloromethyl-Pyrimidine,	16.16	3.98
15	2,1,3-Benzoselenadiazole, 4,6-dichloro-Silane.	16.910	5.30

4.1.2.5: Result of Gas Chromatography-Mass Spectrometry (GCMS) analysis of Glycerol

Thirty compounds were identified by GC-MS analysis of glycerol as shown in (table 4.11) These compounds include; Ethanol, 2-[2-(2-methoxyethoxy)ethoxy]-Ethanol, (10.30%) with a retention time of 3.479RT., 5-Hydroxymethylfurfural, (18.23%) with a retention time of 3.69RT, Ethanol, 2-[2-(2-ethoxyethoxy)ethoxy]-Ethanol(3.13%) with a retention time of 4.073RT., Butane, 1,3-dimethoxy-Methyl 2,5,8,11-tetraoxatridecanoate, (7.88%) with a retention time of 4.325RT., Methylene chloride (0.68%) with a retention time of 5.108RT., Ethanol, 2-[2-(2-butoxyethoxy)ethoxy]-3,6,9,12-Tetraoxahexadecan-1-ol (12.14 %) with a retention time of 5.862 RT., Tetraethyleneglycol monomethylethe (4.91 %) with a retention time of 6.182RT., 2-Butenedioic acid, dibutyl ester,(2.86%) with a retention time of 6.519RT. 2,2-Dimethyl-1-oxa-spiro[2.4]heptane (2.00 %) with a retention time of 7.085 RT. Methylene chloride(0.40%) with a retention time of 7.588RT. 2- Bromopropionic acid, hexyl ester (0.88%) with a retention time of 7.89RT. 3,6,9,12-Tetraoxahexadecan-1-ol (9.72%) with a retention time of 8.326RT, 1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester, Phthalic acid, butyl isohexyl ester, Phthalic acid, isobutyl nonyl ester (9.55%) with a retention time of 9.269 RT.3-(4-Pyridyl)acrylic acid, 3,5-Diethyl-2-n-propylpyridine, Benzeneacetonitrile, 3,4-diethoxy-(0.65%) with a retention time of 9.646RT. n-Hexadecanoic acid,n-Hexadecanoic acid, n-Hexadecanoic acid (2.44 %) with a retention time of 10.023RT.3,6,9,12,15-Pentaoxanonadecan-1-ol, 3,6,9,12-Tetraoxahexadecan-1-ol 15-Crown-5 (1.62%) with a retention time of 10.486RT., Disparlure 4,4-Dimethyl-cyclohex-2-en-1-ol Benzene, 1,3-(0.58%) with a retention time of 11.046RT. Anthranilic acid, N-pyruvoyl- Benzene (1.03) with a retention time of 11.555RT. Benzene, 1,3-dichloro- 2, Benzene (0.45) with a retention time of 12.509RT. Dodecyl isobutyl ether, Carbonic acid, octadecylvinyl ester (0.61) with a retention time of 13.224RT. Dodecane, 2,6,10-trimethyl- (0.61) with a retention time of 13.224RT. Anthranilic acid, N-pyruvoyl-Benzene(0.39%) with a retention time of 13.441RT., 2-Methyltetracosane, Octadecane, 1-chloro- Carbonic acid, octadecyl prop-1-en-2-yl ester(1.07) with a retention time of 13.932RT.Diisooctyl phthalate (0.42%) with a retention time of 14.218RT. Benzene, 1,4-dichloro-Benzene(0.83%) with a retention time of 15.172RT. 2-Propanone, 1-[(6- chloro-3-pyridazinyl)thio]-Anthranilic acid, (0.75%) with a retention time of 15.384RT.3-

pyridinesulfonamide, 5,6-dichloro-L-Methionine (0.34%) with a retention time of 15.853RT.Decanedioic acid, bis(2-ethylhexyl) ester, (2.30%) with a retention time of 16.327RT. Squalene (3.12%) with a retention time of 16.476RT. 2(1H)-Quinazolinone, 4-Aminopyrido[3,2-c]pyridazine, Benzene, 1,2-dichloro-(0.33%) with a retention time of 17.104RT. 2(1H)-Quinazolinone, (0.40%) with a retention time of 17.447RT. Among the compounds identified, the most prevailing compounds identified include: 5-Hydroxymethylfurfural (18.23%) was identified to have the maximum concentration.

Table 4.10: Chemical compounds in glycerol

Peak	Compound	Retention time	Concentration %
1	Ethanol, 2-[2-(2methoxyethoxy)ethoxy]	3.479	10.30
2	5-Hydroxymethylfurfural	3.69	18.23
3	Ethanol, 2-[2-(2-ethoxyethoxy)ethoxy]-	4.073	3.13
4	Butane, 1,3-dimethoxy-Methyl 2,5,8,11-tetraoxatridecanoate.	4.325	7.88
5	Methylene chloride	5.10	0.68
6	Ethanol, 2-[2-(2-butoxyethoxy)ethoxy]-	5.862	12.14
7	Tetraethyleneglycol monomethyle the	6.182	4.91
8	2-Butenedioic acid, dibutyl ester	6.51	2.86
9	2,2-Dimethyl-1-oxa-spiro[2.4]heptane.	7.085	2.00
10	Methylene chloride	7.588	0.4%
11	.2- Bromopropionic acid, hexyl ester.	7.897	0.88%
12	3, 6,9,12,15-Pentaoxanonadecan-1-ol.	8.32	9.72
13	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester..	9.269	9.55
14	3-(4-Pyridyl)acrylic acid	9.64	0.65
15	n-Hexadecanoic acid	10.023	2.44
16	3,6,9,12,15-Pentaoxanonadecan-1-ol	10.486	1.62
17	Disparlure 4,4-Dimethyl-cyclohex-2-en-1-ol Benzene.	11.046	0.58

18	Anthranilic acid, N-pyruvoyl-Benzene.	11.555	1.03
19	Benzene, 1,3-dichloro- 2, Benzene	12.509	0.45
20	Dodecyl isobutyl ether	13.224	0.61
21	Anthranilic acid, N-pyruvoyl-Benzene	13.441	0.39
22	2-Methyltetracosane	13.392	1.07
23	Diisooctyl phthalate	14.21	0.42
24	Benzene, 1,4-dichloro-Benzene.	15.17	0.83
25	2-Propanone, 1-[(6-chloro-3-pyridazinyl)thio]-Anthranilic acid.	15.384	0.75
26	3- pyridinesulfonamide, 5,6-dichloro-L-Methionine.	15.853	0.34
27	Decanedioic acid.	16.327	2.30
28	Squalene	16.47	3.12
29	2(1H)-Quinazolinone	17.10	0.33
30	2(1H)-Quinazolinone	17.447	0.40

4.1.3: Characterization of synthesized biodegradable biofilm

4.1.3.1. Result of Fourier transform infrared spectroscopy analysis of P-BF

Table 4.11shows the FTIR results for sample P-BF**.** The absorbance around 1353.483cm^{-1} and1655.624cm^{-1} were assigned to ethene compound C=C stretching vibration. Medium band around 1617.923cm^{-1} and 3497.720cm^{-1} were assigned to primary and secondary amine compounds N-H stretching vibration of 1^0& 2^0. The peaks around 1840.529cm^{-1} was assigned to cyclic ester compound CO stretching vibration. Wavelength around 2031.918cm^{-1} and 2144.031cm^{-1} were assigned to stretching vibration of carboxylic acid COO. The peak around 2466.928cm^{-1} and 2512.480cm^{-1} were due to stretching vibration of nitrile compound CN respectively. The weak bands around 2622.968cm^{-1}, 2849.760cm^{-1} and 2925.917cm^{-1} were both assigned to stretching vibration of methylene compound C-H respectively. Strong bands around 3218.984$cm^{-1,}$ 3686.776cm^{-1} and 3810.973cm^{-1} were assigned to stretching vibration of primary and secondary alcohols (1^0& 3^0 alcohols) OH .

Table 4.11: Result of Fourier transform infrared spectroscopy analysis of P-BF

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	1353.483	$H_2C=CH$	Ethene CH anti-symmetric stretch
2	1445.958	$H_2C=CH$	Ethene CH anti-symmetric stretch
3	1655.624	RNH_3	1^0 amine NH stretch
4	1840.529	R-O-R	Cyclic ester CO stretch
5	2031.918	RCOOH	Carboxylic acid COO stretch
6	2144.031	RCOOH	Carboxylic acid COO stretch
7	2466.928	R-C≡N	Nitriles CN antisymmetric stretch
8	2512.480	R-C≡N	Nitriles CN antisymmetric stretch
9	2622.968	CH_2	Methylene CH symmetric stretch
10	2849.760	CH_2	Methylene CH symmetric stretch
11	2925.917	CH_2	Methylene CH symmetric stretch
12	3218.984	RCHOH	1^0 alcohol OH symmetric stretch
13	3497.720	R_2NH	2^0 amine NH stretch
14	3686.776	RCHOH	1^0 alcohol OH symmetric stretch
15	3810.973	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.3.2: Result of Fourier transform infrared spectroscopy analysis of NP-BF

Table 4.12 shows the FTIR analysis results for NP-BF**,** the peak values around 1432.4124cm^{-1} were assigned to C=C stretching vibration of ethene compound. The medium band around 1615.605cm^{-1} was assigned to N-H stretching vibration of 1^0 amine compound. The peak value around 1830.269cm^{-1} and 1988.089cm^{-1} were assigned to CO stretching vibration of cyclic ester compound respectively. Absorbance around 2120.197cm^{-1} was assigned to COO stretching vibration of carboxylic acid respectively whereas the peaks located around 2727.653cm^{-1} and 2866.758cm^{-1} were both assigned to C-H stretching vibration of methylene compounds. The peak values located around 3000.204cm^{-1}, 3144.288cm^{-1}, 3328.285cm^{-1}, 3511.090cm^{-1}, 3660.359cm^{-1} and 3817.882cm^{-1} were all assigned to OH stretching vibration of (primary, secondary and tertiary alcohols) 1^0, 2^0& 3^0 alcohols respectively.

Table 4.12: Result of Fourier transform infrared spectroscopy analysis of NP-BF

S/N	Wavelength (cm^{-1})	Functional group	Compounds
1	1432.4124	$H_2C=CH$	Ethene C=C anti-symmetric stretch
2	1615.605	RNH_3	1^0 amine NH stretch
3	1830.269	R-O-R	Cyclic ester CO stretch
4	1988.089	R-O-R	Cyclic ester CO stretch
5	2120.197	RCOOH	Carboxylic acid COO stretch
6	2469.075	R-C≡N	Nitriles CN anti-symmetric stretch
7	2727.653	CH_2	Methylene CH symmetric stretch
8	2866.758	CH_2	Methylene CH symmetric stretch
9	3000.204	RCHOH	1^0 alcohol OH symmetric stretch
10	3144.288	RCHOH	1^0 alcohol OH symmetric stretch
11	3328.285	R_2CHOH	2^0 alcohol OH symmetric stretch
12	3511.090	R_3CHOH	3^0 alcohol OH symmetric stretch
13	3660.359	R_3CHOH	3^0 alcohol OH symmetric stretch
14	3817.882	R_3CHOH	3^0 alcohol OH symmetric stretch

4.1.4. Morphology study

4.1.4.1. Result of Morphology study of P-BF

The micrograph of the morphology study of P-BF is shown in plate 11. The morphology study of P-BF showed white patches on the composite, the white patches were identified to be the egg shell powder used as a re-enforcement material. There were also black patches and a ring like substance contributed by plantain peel.

4.1.4.2. Result of Morphology study of NP-BF

Plate12 shows the micrograph morphology study of N-BF. The morphology study of N-BF shows clusters of constituent in the composites. This could be due to the fibres in the plantain peels.

Plate 1: Micrograph of P-BF with a magnification of ×175.

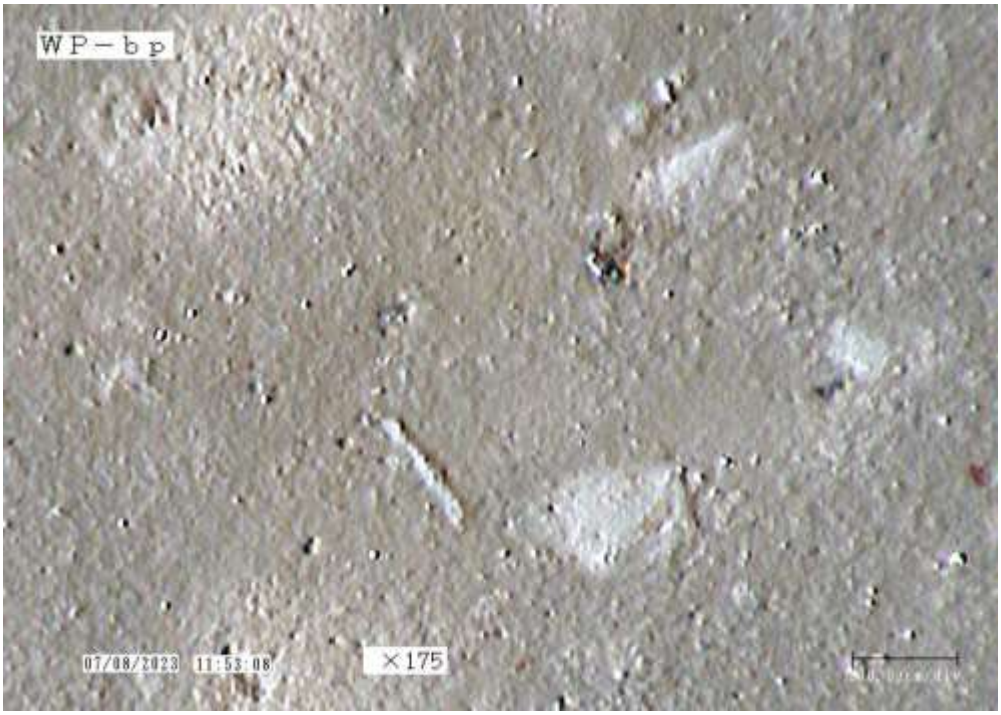


Plate 2: Micrograph of NP-BF with a magnification of ×175.

4.1.5. Result of Thermogravimetric analysis

4.1.5.1. Result of Thermogravimetric analysis of P-BF

Figure 4.1 shows TGA (thermogravimetric analysis). First step decomposition occurred at the temperature of 29.92^0C - 137^0C for 12 min. About 14.75% of the material was decomposed leaving 85.32% of the composite. This reduction in the weight of the biofilm is caused by the release of moisture or water. In this stage, the very light volatile compounds was also lost and the initial stage of the thermal decomposition process occurs due to evaporation of the water (Yeum, Park and Choi, 2006; Sukarni et al., 2015). About 14.75% of the material was decomposed leaving 85.32% of the composite. Consequently, 4.12% of the composite was decomposed at the temperature of 137^0C- 203.02^0C for 25 min. whereas 81.10% of the composite was left. Stage 2 is the process of releasing volatile matter (Li et al., 2001; Raabe et al., 2015). Third step decomposition occurred at a temperature of 203.02^0C- 331.04^0C for 16 min. About 27.38% of the composite was decomposed remaining 53.75%. . Stage 3 is the stage after the release of volatile matter in the samples occurred. The fixed carbon content of the biofilm was relatively low. In this stage, the charcoal is flammable as it is surrounded by volatile matter and oxygen diffused on the surface of the charcoal, which burn the charcoal and volatile matter simultaneously. This stage occurs after the release of volatile matter which leaves or forms carbon (Azevedo et al., 2015).

Stage 4 is the last stage of the thermal decomposition process in biofilm that occurred at 331.04^0C -500°C for 23 min. This process does not produce ash, since the composite was almost completely decomposed remaining 44.19% residue. This thermal behavior agrees with other starch bio-based polymeric films reported by other authors in literature such as cassava starch films (Marques et al., 2006; Rodríguez et al., 2015). Sugar palm starch films, and corn starch films (Shi et al., 2007; Sanyang, Sapuan, Jawaid, Ishak, and Sahari, 2015). In summary, the thermo-gravimetric analysis determined that the degradation of P-BF starts around 137 °C; therefore, it should be processed at temperatures below that.

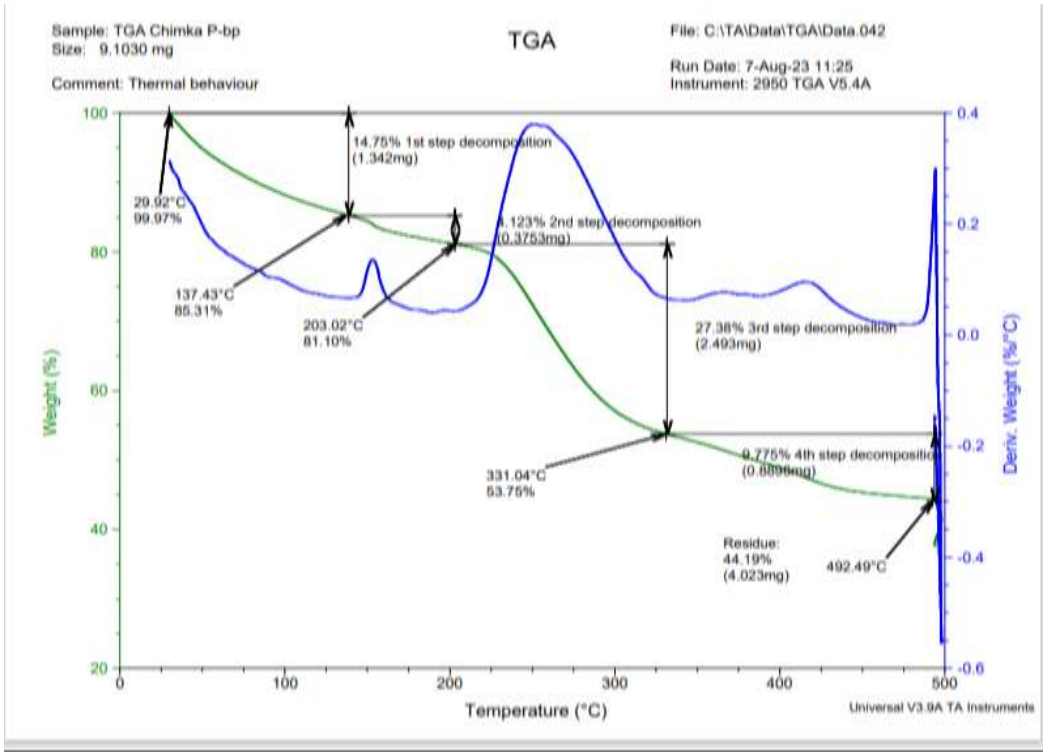


Figure 4.1: Thermogravimetric Analysis of P-BF

4.1.5.2. Result of Thermogravimetric analysis of NP-BF

Figure 4.2 shows TGA (thermogravimetric analysis) of NP-BF. First step decomposition occurred at the temperature of 22.17^0C - 139.95^0C for 12 min. About 11.9% of the material was decomposed leaving 88% of the composite. This is the reduction weight caused by the release of moisture or water. In this stage, the very light volatile matter compounds was also lost and the initial stage of the thermal decomposition process occurs due to evaporation of the water (Yeum, Park and Choi 2006; Sukarni et al., 2015)

Consequently, second step decomposition took place in which 8.1% of the composite was decomposed at the temperature of 139.95^0C - 213^0C for 25 min. whereas 79% of the composite was left. Stage 2 is the process of releasing volatile matter (Li and Yeh, 2001; Raabe et al., 2015). Thereafter, third step decomposition occurred at a temperature of 213^0C-366^0C for 16 min. About 48% of the composite was decomposed remaining 32% of the composite. Stage 3 is the stage after the release of volatile matter in the samples occurred. The fixed carbon content of the biofilm was relatively low. In this stage, the charcoal is flammable as it is surrounded by volatile matter and oxygen diffused on the surface of the charcoal, which burn the charcoal and volatile matter simultaneously. This stage occurs after the release of volatile matter which leaves or forms carbon (Azevedo et al., 2015). Stage 4 is the last stage of the thermal decomposition process in the biofilm that occurred at 366°C-500°C for 23 min. This process does not produce ash, since the composite was almost completely decomposed remaining 11.9% residue.

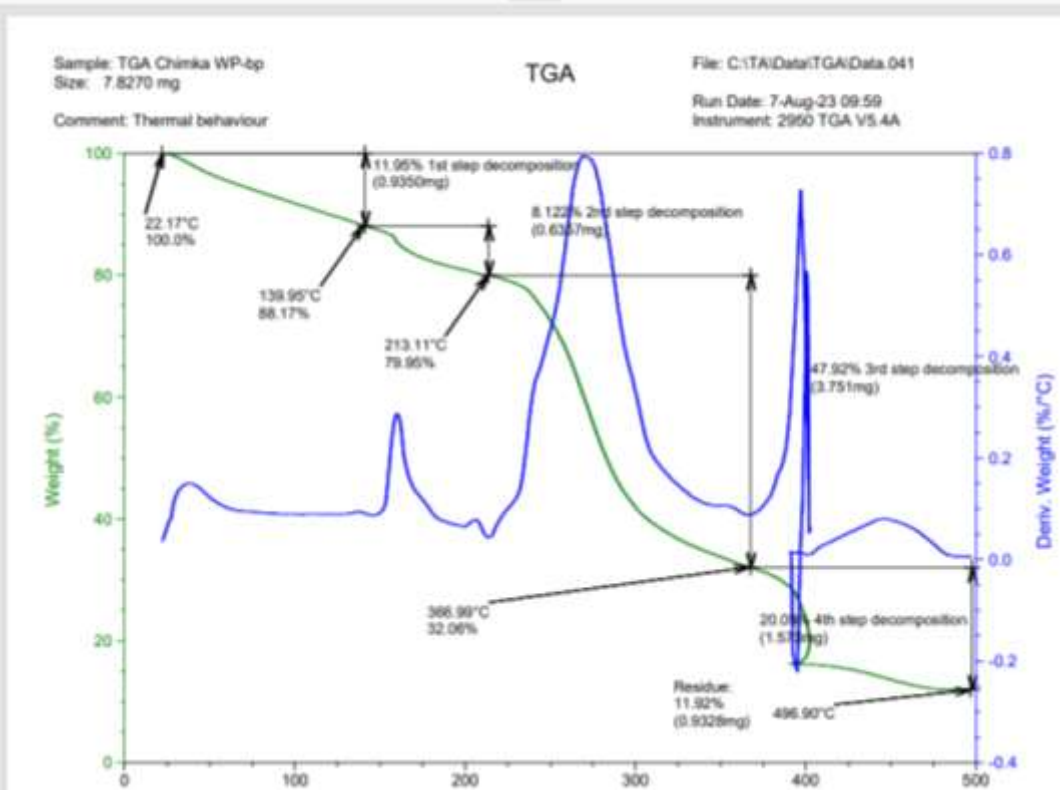


Figure 4.2: Thermogravimetric analysis of NP-BF

4.1.5.3. Result of TGA (thermogravimetric) analysis of P-BF and N-BF

Figure 4.3 shows the result of TGA (thermogravimetric analysis) of P-BF and NP-BF. The result showed that the P-BF degraded thermally less than the NP-BF with more residual value than the NP-BF. This implies that the plantain peel based biofilm can withstand high temperatures upto 500^0C (Nurul Aina et al., 2016).

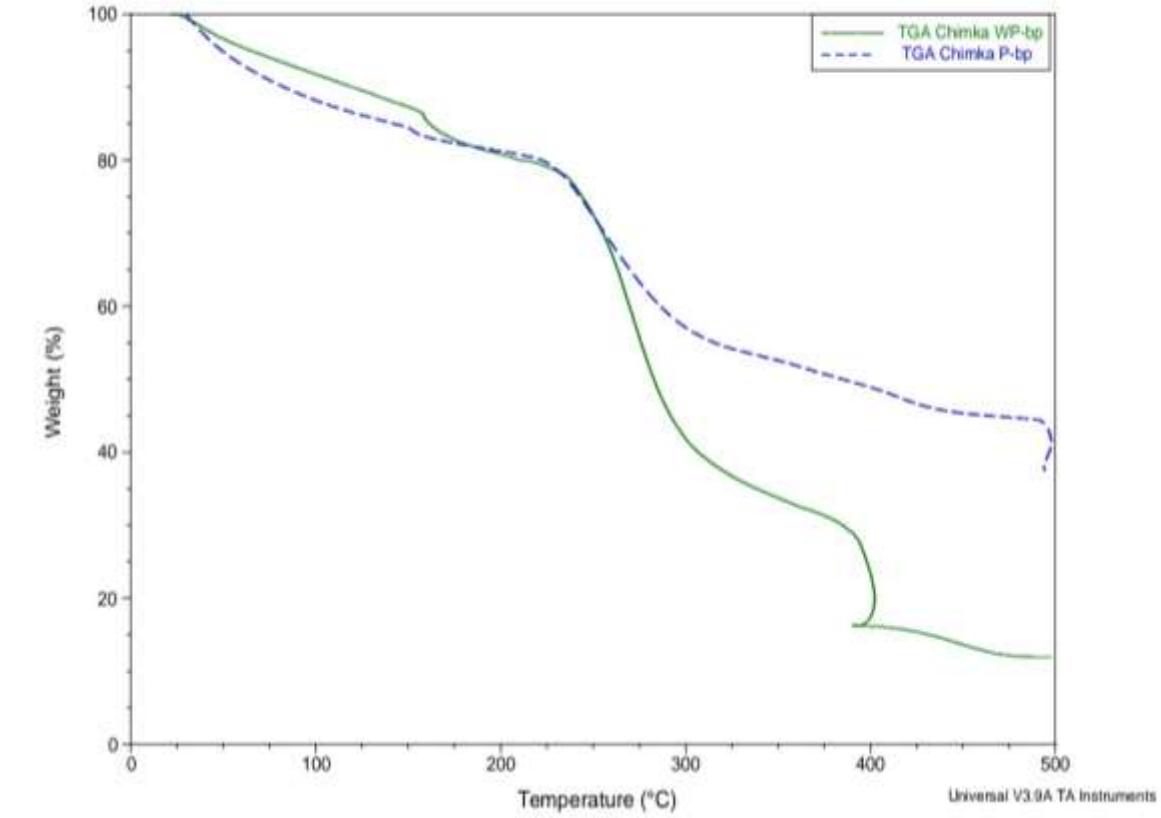


Figure 4.3: Thermogravimetric analysis of P-BF and NP-BF

4.1.6. Result of water absorption analysis

Table 4.13 shows the result for water absorption analysis of the synthesized biodegradable films. The results for water absorption test showed a significantly ($p < 0.05$) lower water absorption by the P-BF compared to the NP-BF.

Table 4.13: Result of water absorption analysis of synthesized biofilm

Sample	**Initial wt (g)**	**Final wt (g)**	**Difference** (g)	**% Water Absorb**	**MEAN±SD**
P-BF	1.09	1.73	0.28	37	$0.31\pm.04^{a}$
	1.64	2.10	0.28	28	
	1.44	1.85	0.28	28	
Average				31	
N-PF	0.46	0.79	0.72	72	0.46 ± 0.19^{b}
	0.79	1.04	0.32	32	
	0.51	0.69	0.35	35	
Average				46	

Results are presented as mean±SD (n=6) of triplicate determinations. Mean values with different superscript letters are statistically significantly ($p<0.05$) different. P-BF= plantain peel biofilm, N-BF= non- plantain peel biofilm. SNI (Indonesian National Standard) for water absorption of bioplastics is 21.5% at (25^{0} C) room temperature and 69.09% at 100^{0}C

4.1.7. Result of swelling test

Table 4.14 showed the result of swelling test of the synthesized biofilm. The results of the study showed that there was not much change when P-BF was soaked in chloroform and methanol, but N-BF showed slight increase in weight when kept in chloroform and methanol.

Table 4.14: Result of swelling test of the synthesized biofilm.

Sample	Initial wt (g)	Final wt (g)	Weight gain (g)	Initial wt (g)	Final wt (g)	Weight gain (g)	Mean±SD
Methanol							
P-BF	2.01	2.10	0.10	2.02	2.12	0.10	0.10 ± 0.00^{b}
N-BF	2.02	2.12	0.10	2.01	2.13	0.12	0.10± 0.01
Chloroform							
P-BF	2.00	2.05	0.05	2.00	2.04	0.04	0.04 ± 0.00^{a}
N-BF	2.00	2.02	0.02	2.00	2.02	0.02	0.02±0.00

Results are presented as mean±SD (n=6). Mean values with different superscript letters are considered statistically significanty ($p<0.05$) different. P-BF: plantain peel based biofilm; N-BF: non- plantain peel based biofilm.

4.1.8. Result of solubility test

Table 4.15 shows the solubility test for synthesized composites using different solvents.ammonia, acetic acid, acetone, sulphuric acid and ethyl alcohol.The solvents were chosen in such a way that the activity of material with parameters like high acidic solvent, polar solvent, non-polar solvent and weak acid were determined.The P-BF sample were not completely soluble in different medium used in comparison to the N-BF which were completely soluble in acetone but partially soluble in other solvent. These are certainly desired results for preparation of bioplastics. This shows that the plantain peel biofilm are stable (Yaradoddi et al., 2016).

Table 4.15: Result of solubility test of synthesized biofilms using different solvents.

Sample	Solvent	insoluble	Partially soluble	Completely soluble
P-BF	Acetone	-	+	-
NP-BF	Acetone	-	-	+
P-BF	Sulphuric acid	-	+	-
N-BF	Sulphuric acid	-	+	-
P-BF	Ethyl alcohol	-	+	-
NP-BF	Ethyl alcohol	+	-	-

Results are presented as P-Bp=: plantain peel based biofilm; N-BF: non plantain peel based biofilm. **Positive: +** **Negative: -**

4.1.9. Result of biodegradability test

Table 4.16 shows the results of biodegradation test of the synthesized biofilms. The results showed that N-BF had a significantly ($p<0.05$) higher biodegradation activity compared to P-BF.

Table 4.16: Result of the biodegradability test of synthesized biofilm

Sample	Day 0 wt (g)	Day 6 wt (g)	Day 12 wt (g)	Difference(g)	Percent of Degrade %	Mean± STD
P-Bf	2.98	2.59	0	0.13	100	$0.13\pm.017^{b}$
NP-Bf	1.01	0.81	0	0.19	100	$0.19\pm.02^{a}$

Results are presented as mean±SD (n=6). Mean values with different superscript letters are statistically significantly ($p<0.05$) different. P-BF=: plantain peel based biofilm; NP-BF: non-plantain peel based biofilm.

4.1.10. Result of Mechanical Properties

Table 4.17 shows the results of mechanical indices as exhibited by the synthesized biodegradable biofilm. The NP-BF was shown to have a significantly ($p<0.05$) higher tensile strength, Hardness ShoreD, % elongation and Flexural strength than the P-BF.

Table 4.17: Result of the Mechanical indices of synthesized biofilm.

	P-BF	**N-BF**	**Standard value**
(Nmm^2)			
Tensile strength	2.87 ± 0.02^{b}	5.45 ± 0.02^{a}	1-10 (Nmm^2)
Hardness ShoreD	22.00 ± 1.78^{b}	49.00 ± 1.78^{a}	
%Elongation	6.29 ± 0.01^{b}	13.85 ± 0.03^{a}	10-20%.
Flexural strength	0.41 ± 0.01^{b}	1.56 ± 0.02^{a}	1-8 (Nmm^2)

Results are presented as mean±SD (n=6). Mean values with different superscript letters are statistically significantly ($p<0.05$) different.

4.1.11. Result of toxicity test

GC-FID was used to determine concentration of toxic compounds (PAHs) (polycyclic aromatic hydrocarbons) in the synthesized raw materials.

4.1.11.1 Result of GC-FID analysis of eggshell powder

Table 4.18 shows PAH analysis results of eggshell powder. Nine compounds were identified in the egg shell powder by GC-FID, of which dibenzy (a,h)pyrene was identified to have the highest concentration (20.27%) followed by benzo(k)fluoranthene(15.01%) and phenanthrene (14.74%).

Table 4.18: Result of GC-FID analysis of eggshell powder

Component	Concentration (µg /g)	% Concentration	Permissible limit (µg/g)EUNo835/2011
Pyrenees	1.07	7.54	0.03
Benz(k)fluoranthene	2.13	15.01	0.2
Acenaphthylene	1.52	10.75	0.6
Phenanthrene	2.09	14.74	0.9
Dibenzy(a,h)pyrene	2.87	20.26	0.3
1_2Benzanthracene	0.96	6.81	0.06
Benzo(a)pyrene	0.64	4.52	0.02
Benzo(g_h_i)perylene	1.80	12.72	0.01
Benzo (c) chrysene	1.07	7.54	0.01
Total	14.2034		

4.1.11.2. Result of GC-FID analysis of glycerol

Table 4.19 shows PAH analysis results of glycerol. Eight compounds were identified, the most prevailing polycyclic aromatic compounds are 1_2benzanthracene (44.24%), Fluoranthene **(**24.54%) , Benzo [a] pyrene **(**13.60%) and Pyrene(10.25%). The rest of the compounds occurred in minimal quantities. This include; Benzo[e]pyrene (0.10%), Phenanthrene (0.70%) and Perylene (0.04%).

4.19: Result of GC-FID analysis of crude glycerol

Component	**Concentration (mg /ml)**	**% Concentration**	**Permissible limit (µg/g)EURegNo835/2011**
Benzo [a] pyrene	0.42	13.60	0.05
Perylene	0.001	0.04	0.05
Fluoranthene	0.76	24.54	0.05
1_2 Benzanthracene	1.38	44.23	0.06
Phenanthrene	0.02	0.70	0.9
Dibenzyl[a_h]anthracene	0.20	6.50	0.06
Benzo[e]pyrene	0.003	0.10	0.01
Pyrene	0.32	10.25	0.03
Total	3.12		

4.1.11.3. Result of GC-FID analysis of Vinegar

Table 4.20 shows PAH analysis results of vinegar. Seven compounds were identified by GC-FID.The most aboundant compounds were identified include; Benzo[e]pyrene (44.86%), 1_2 Benzanthracene (13.92%), Pyrene(13.49%), Phenanthrene(10.46%), Benzo[g_h_i]perylene (9.29%),Other compounds were identified in trace amounts such as; Anthracene(3.13%) and Fluoranthene (4.83%).

Table 4.20: Result of GC-FID analysis of vinegar.

Component	Concentration (mg/ml)	% Concentration	Permissible limit (mg/kg)EURegNo835/2011
Fluoranthene	0.14	4.83	0.05
1_2 Benzanthracene	0.42	13.92	0.06
Phenanthrene	0.32	10.46	0.9
Anthracene	0.09	3.13	0.06
Benzo[e]pyrene	1.38	44.86	0.01
Pyrene	0.41	13.49	0.03
Benzo[g_h_i]perylene	0.28	9.29	0.01
Total	3.08		

4.1.11.3. Result of GC-FID analysis of cassava starch

Table 4.21 shows PAH analysis results of cassava starch. Four compounds were identified in the cassava starch by GC-FID. The most prevailing compound was identified as 1_2 benzanthracene (43.29%), other compounds occurred in minimal amounts, This include; Benzo[b] Fluoranthene (21.46%), Pyrene (21.00%) and Benzo [g_h_i]perylene**(**14.20%**).**

Table 4.21: Result of GC-FID analysis of cassava starch

Component	Concentration (mg/ml)	% Concentration	Toxic limit (mg/kg) EU Reg No 835/2011
Pyrene	0.48	21.00	0.03
1_2 Benzanthracene	0.99	43.29	0.06
Benzo[g_h_i]perylene	0.32	14.20	0.04
Benzo[b]Fluoranthene	0.49	21.46	0.01
Total	**2.28**		

4.1.11.4. Result of toxicity test of soil containing biodegraded biofilm

Table 4.22 shows toxicity test of soil containing biodegraded biofilm.

Three soil samples (loamy soil) were used coded C (control), W (containing biodegraded NP-BF) and P (containing biodegraded P-BF). Fifteen PAHs were identified in different soil samples. Acenapthylene was the most abundant in soil P, but was found to be low in soil W. However, it was much lower compared to the control. Furthermore, Acenaphthene was the most abundant in soil W but was found to be low in soil P. Nevertheless, it was much lower compared to the control.

Table 4.22: Result of toxicity test of soil containing biodegraded biofilm.

PAHs	soil C (µg /g)	soil P (µg /g)	soil W(µg /g)
Napthalene	0.21 ± 0.00^{b}	0.11 ± 0.00^{a}	0.11 ± 0.01^{a}
Acenapthylene	1.55 ± 0.23^{b}	0.43 ± 0.15^{a}	0.22 ± 0.00^{a}
Acenaphthene	0.51 ± 0.03^{a}	0.15 ± 0.00^{b}	0.60 ± 0.48^{a}
Fluorene	0.43 ± 0.01^{a}	0.37 ± 0.07^{a}	0.37 ± 0.07^{a}
Phenanthracene	0.67 ± 0.24^{a}	0.27 ± 0.07^{a}	0.27 ± 0.21^{a}
Anthracene	0.66 ± 0.12^{b}	0.22 ± 0.00^{a}	0.11 ± 0.00^{a}
Fluoranthene	0.61 ± 0.05^{a}	0.28 ± 0.03^{b}	0.12 ± 0.02^{c}
Pyrene	0.02 ± 0.00^{a}	0.27 ± 0.00^{b}	0.09 ± 0.0^{c}
Benzo(a) anthracene	0.11 ± 0.00^{a}	0.22 ± 0.00^{b}	0.10 ± 0.00^{c}
Chrysene	0.40 ± 0.27^{b}	0.03 ± 0.00^{a}	0.21 ± 0.01^{a}
Benzo(k)fluoranthene	0.51 ± 0.03^{a}	0.22 ± 0.00^{b}	0.09 ± 0.02^{c}
Benzo(a)pyrene	0.43 ± 0.12^{a}	0.11 ± 0.00^{b}	0.09 ± 0.02^{c}
Indeno(1_2_3)pyrene	0.36 ± 0.00^{b}	0.18 ± 0.08^{a}	0.11 ± 0.00^{a}
Benzo(a_h)anthracene	0.61 ± 0.03^{b}	0.21 ± 0.08^{a}	0.09 ± 0.00^{a}
Benzo(a_h)perylene	0.52 ± 0.04^{b}	0.27 ± 0.08^{a}	0.17 ± 0.03^{a}

Results are presented as mean±SD (n=2) of triplicate determinations. Mean values with the different superscript$^{a,b \text{ or } a,b,c}$ are considered statistically significant ($p<0.05$). Soil c; control, soil P: soil with P-BF, soil W: soil with N-BF.

Recommended limit for PAHS in the soil: 1mg/kg (DPR, 2002) and 1mg/kg, 1500 µg/kg, and 5 mg/kg (ANZECC 2000; MHSPEN 2000; DEPA 2002).

4.2. DISCUSSION

Biodegradable plastics are made from natural polymeric materials such as starch, vegetable oil, cellulose, lignin, and also materials derived from animals such as proteins and lipids. Bioplastics are easily degraded by microbes, and the degradation process does not take a long time. (Ezeoha and Ezenwanne, 2013; Siangian and Tarigan, 2016).

The results of the FTIR analysis as depicted in Table 4.1, 4.2, 4.3, 4.4. 4.5, 4.6, 4.11 and 4.12 revealed 17 peaks for glycerol, 16 peaks for vinegar, 15 peaks for cassava starch, 16 peaks for eggshell, 15 peaks for plantain peel and likewise 15 peaks for P-BF and 14 peaks for N-BF biofilm.

These peaks ranged from 1102.833cm^{-1} to 3809.275cm^{-1} of wavelength for glycerol and 11021.292 cm^{-1} to 3803.590cm^{-1} of wavelength for vinegar., 1058.841cm^{-1} to 3516.919cm^{-1} of wavelength for cassava starch and 1383.668cm^{-1} to 3839.897 cm^{-1} for eggshell. Likewise 1328.304cm^{-1} to 3661.209 cm^{-1} wavelength for plantain peels, 1353.483 cm^{-1} to 3810.973cm^{-1} of wavelength for P-BF and 1432.4124cm^{-}1 to 3817.882cm^{-1} of wavelength for NP-BF. These peaks represent the following functional groups, ether, ethene, amine, carboxylic acid, nitriles, methylene cyclic ester, primary, secondary and tertiary alcohols common to hydrocarbons respectively. These functional groups indicate that bioplastics are safe for human use and the environment. The presence of carboxylic acid is important for enhancing the mechanical and chemical properties of bioplastics, making them a viable alternative to petroleum-based plastics (Vishishtta *et al*., 2023). Amines contribute to the flexibility, strength, and thermal degradation resistance of bioplastics, thus aiding in their biodegradability (Vishishtta et al., 2023).

The chemical compounds in the synthesized raw materials (glycerol, cassava starch, plantain peel, eggshell, acetic acid) were analysed using GC-MS. The result of the GC-MS analysis of cassava starch showed 15 peaks representing 15 chemical compounds. The most aboundant compound was dihexyl monoselinide with a concentration of 11.54 % and the least aboundant was benzoic acid with a concentration of 4.12 %. Dihexyl monoselinide is a chemical compound that plays a role in enhancing the chemical properties of biofilms. Benzoic acid helps in the durability of biofilms.

The result of the GC-MS analysis of eggshells showed 15 peaks representing 15 chemical compounds. The most aboundant compound was 1,2 benzene dicarboxylic acid with a concentration of 11.62 % and the least aboundant was dihexyl monoselinide with a concentration of 4.68 %. 1, 2 benzene dicarboxylic acid plays a role as a plasticiser enhancing the strength and flexibility of biofilm. Dihexyl monoselinide is a chemical compound that plays a role in enhancing the chemical properties of biofilms.

The result of the GC-MS analysis of acetic acid showed thirty peaks representing thirty chemical compounds. The most aboundant compound was benzoic acid with a concentration of 17.04 % and the least aboundant was Tetratriacontyl pentafluoropropionate with a concentration of 0.90 %. Benzoic acid helps to improve the durability of biofilms.

Furthermore, the result of the GC-MS analysis of plantain peel powder showed fifteen peaks representing fifteen chemical compounds. The most aboundant compound was N-(3-Nitro-thiobenzoyl)-morpholine with a concentration of 11.03 % and the least aboundant was Silane, 9-anthracenyltrimethyl-Methaqualone with a concentration of 3.88 %. N-(3-Nitro-thiobenzoyl)-morpholine helps to improve the strength, flexibility and longevity of biofilms.

Additionally, the result of the GC-MS analysis of crude glycerol showed thirty peaks representing thirty chemical compounds. The most aboundant compound was Ethanol, 2-[2-(2-butoxyethoxy) ethoxy]- with a concentration of 12.14 % and the least aboundant was 3-pyridinesulfonamide, 5,6-dichloro-L-Methionine with a concentration of 0.34 %. Ethanol, 2-[2-(2-butoxyethoxy) ethoxy]- enhances the mechanical properties of biofilms.

Among the chemical compounds detected, few of them possessed certain biological activities. These included; 2-Butenedioic acid (Z) and dibutyl ester, was reported to have antioxidant and anti-inflammatory activity. Okechukwu, (2020) reported the antiin- flammatory, analgesic and antipyretic properties of pentadecane. Bis (2-methylpropyl) ester and Benzoic acid possess antioxidant and antimicrobial activity. Also, methaqualone a substituted quinazolines has been reported to be a sedative hypnotics and possess antispasmodic property (Gass, 2008). Likewise, dibut-3-enyl phthalate, 1, 2-Benzenedicarboxylic acid and 1-Dodecanol was reported by Naoko *et al*., (2007); Roy et al., (2006) to exhibit antiinflammatory activity. This implies that the

synthesized biofilm when degraded by microbial activity will cause no harm to the environment and as well be harmless to man.

Likewise, the biological activities of 3-(2-Hydroxy-6-methylphenyl)-4(3H)-quinazolinone and diosgenin include; anticancer, cardio protective, antiasthmatic, anti diabetic, (Kalailingam et al., 2014; Kim and Koh, 2016; Sethi, Shanmugam and Warrier, 2018).These properties of the synthesized biofilm indicate that the biofilm is eco-friendly. Furthermore, 1,2 Benzenedicarboxylic also helps to increase the flexibility of the biofilm. 1-Hexene enhances the mechanical strength of the biofilm.

However, the compounds identified here were completely different from previous reports by other authors (Mie-ling et al., 2003). The difference in composition of these extracts could be due to the treatment of the peel before extraction. In the previous reports, the banana peels were dried for two weeks at atmospheric conditions, powdered, and extracted with ethanol or methanol. However, in this study, the plantain peels and egg shell were sun dried for one month and then powdered and analysed. The cassava starch was sun dried after extraction and then analysed. Organic solvent was not used in the study. The difference may also have been attributed to the species of the raw materials (Mie-ling et al., 2003).

The morphology study of P-BF as depicted in plate 1, shows that there were agglumeration of constituent materials on the composite shown in the form of white patches. This was identified to be the egg shell powder used as a re-enforcement material. There was also black patches and a ring like substance contributed by the plantain peel. This showed that the constituent materials were not uniformly distributed in the composite. The morphology study of N-BF as depicted in plate 2, showed the constituents' materials in the N-BF composite was more evenly distributed compared to that of the P-BF composite which reflect in their mechanical properties (Nurul Aina, Syuhada and Yahyac, 2016).

The TGA analysis of the synthesized composite (P-BF and NP-BF) was shown in figure 4.1 and 4.2. Figure 4.1 showed TGA (thermogravimetric) analysis of P-BF. The composite was heated at 29.92^0C (room temperature) to 500^0C. First step decomposition occurred at the temperature of 137^0C. The high volatility first weight loss step is due to the loss of plasticizers from the polymer (Nurul Aina et al., 2016). About 14.75% of the material was decomposed leaving 85.315% of the

composite. Consequently, in the 2nd step decomposition, 4.123% of the composite was decomposed at the temperature of 203.02^{0}C whereas 81.10% of the composite was left. The low volatility second step weight loss is due to degradation of the residual polymer (Nurul Aina et al., 2016).

3rd step decomposition occurred at a temperature of 331.04^{0}C in which 27.38% of the composite was decomposed remaining 53.75%. Weight loss in the third step occured due to combustion of compounds such as residual carbon from polymer breakdown or burn off of carbon black filler. Furthermore, at 492.49^{0}C, 9.75% of the composite was decomposed remaining 44.19% residue. The residue value is the remaining ash due to inorganic filler materials that were present in the material. Additionally, the TGA anlalysis of P-BF showed the first decomposition occurred at 137.43^{0}C while the maximum loss occurred at 331.04^{0}C.

Figure 4.2 showed TGA (thermogravimetric analysis) of N-BF.The composite was heated at temperature of 22.17^{0}C (room temperature) to 500^{0}C. First step decomposition occurred at the temperature of 139.95^{0}C. About 11.9% of the material was decomposed leaving 88% of the composite. The high volatility first weight loss step was due to the loss of plasticizers from the polymer. Consequently, second step decomposition took place in which 8.1% of the composite was decomposed at the temperature of 213^{0}C whereas 79% of the composite was left. The low volatility second step weight loss was due to degradation of the residual polymer.Thereafter, 3rd step decomposition occurred at a temperature of 366^{0}C in which 48% of the composite was decomposed remaining 32% of the composite. Weight loss in the third step occurred due to combustion of compounds such as residual carbon from polymer breakdown or burn off of carbon black filler. Furthermore, at 496^{0}C, the composite was almost completely decomposed remaining 11.9% residue. The residue value was the remaining ash due to inorganic filler materials that were present in the material. In addition, the TGA anlalysis of P-BF showed the first decomposition occurred at 139.95^{0}C while the maximum loss occurred at 366.99^{0}C.

However, the result of the TGA analysis of P-BF and NP-BF showed that P-BF composite had more material left (44.19%) while NP-BF composite had 11.9% material remaining after decomposing at the same temperature of 500^{0}C. This implies that the P-BF composite was thermally stable at a high temperature compared to the NP-BF.

Figure 4.3 showed the thermograph of P-BF and NP-BF. As the temperature increased to 100^0, the biofilm decomposed causing the weight of the bioplastics to decrease until it got to a steady value at 200^0C where both graphs merged, at this point, there was loss of moisture content in the material. Further increase in temperature upto 500^0c, the NP-BF decomposed slightly causing a slight drop in the graph compared to the control (NP-BF) which decomposed drastically with a heavy drop in the graph. This could be due to the plantain peels in the P-bp which is the only material absent in the NP-BF. P-BF decomposed slightly compared to NP-BF having 85% of material left while 88% of NP-BFwas left after decomposition at the same temperature. This therefore implies that the plantain peel based biofilm can withstand high temperatures upto 500^0C.

Liquid impermeability is one of the conventional properties of plastic.The water absorption test evaluates the durability and suitability of bioplastics for moisture or water-related applications.
Water resistance is an important characteristic in determining a suitable source for biofilm. This test provides insights into the material's resistance to water infiltration and its dimensional changes or expansion when exposed to moisture (Vishishtta et al., 2023).The water absorption of the synthesized biofilm was carried out at room temperature for 24 hours to obtain the maximum water uptake data. The result of the water absorption test showed that P-BF absorbed less water compared to NP-BF. However, P-bf biofilm water engorgement were above the standard limit as set by SNI (Indonesian National Standard) for water absorption for bioplastics; 21.5% at (25^0 C) room temperature and 69.09% at 100^0C (Chika, Muhamad, and Rinda, 2024).
The P-BF films had a moisture engorgement above the standard value of 21.5% because biopolymers are hydrophilic in nature (May et al., 2019). The cellulosic fibres in the plantain peels are hydrophilic due to the several hydrogen bonds between macromolecules of cell wall in the fibres. When these fibres come in contact with water, the hydrogen bonds breaks and hydroxyl groups (-OH) forms a new hydrogen bonds with water molecules (May et al., 2019). Besides, the water molecules interact with hydroxyl group in starch structure, the plasticization of biopolymer with glycerol was also an important factor in this study. As glycerol is a hydrophilic low molecular carbohydrates, it has the tendency to absorb water depending on the number of hydroxyl group present and molecular weight of it structure. Glycerol has three carbons attached to their backbone with one hydroxyl group attached to each carbon which causes the molecules to bind to the highest amount of water corresponding to the weight portion

(May et al., 2019). Increasing sizes of hydroxyl groups concentration centre in bio composite matrix increases the water absorption of the film (May et al., 2019).

The results of swelling test showed slight swelling by the P-BF when chloroform was used. However, no change was observed between P-BF and NP-BF when methanol was used. It can be deduced that there was no much change in the integrity of biofilm material when the biofilm material was soaked in chloroform and methanol solvents. This may be due to the hydrophilic nature of cellulosic fibres in the plantain peels which has alternating hydroxyl side chain (Jamshidi et al., 1988).This was in line with research conducted by Vishishtta et al., (2023). In the findings, Banana based biofilm showed slight swelling in chloroform but no change with methanol.

Solubility are the main property to check whether the synthesized bioplastic material is sustainable or not. The result of the present study showed that P-BF was partially soluble in acetone and sulphuric acid while NP-BF was completely soluble in acetone but partially soluble in sulphuric acid. Similarly, P-BF was partially soluble in ethyl alcohol while NP-BF was insoluble in ethyl alcohol. Furthermore, the result showed that P-BF sample were not completely soluble in different medium used in comparison to the NP-BF and also had less engorgement compared to NP-BF. These are undoubtedly desired results for preparation of bioplastics. This showed that the plantain peel biofilms are stable (Yaradoddi et al., 2016). According to Yaradoddi et al., (2016), If the biofilm material possessed the property of less or zero engorgement property that could be considered as excellent material with stability as characteristic features (Yaradoddi et al., 2016).

Biodegradability tests evaluated how easily bioplastics break down in the environment. These tests assessed the speed of degradation and ecological integration to determine the environmental impact and sustainability of bioplastics. They also provide insights into the mechanisms of deterioration and help determine their suitability for specific applications. The degradation studies (soil burial test) conducted was helpful in preparation of environmental friendly product which are derived from natural polymers, they can be reused in bio compost preparation. This versatility of bioplastic plays key role in green applications (May et al., 2019).The result of the degradation showed that the synthesized composite (P-BF) degraded on the 6^{th} day from 2.98g to

2.59g with a percentage degradation of 100% whereas NP-BF composite degraded on the 6th day from 1.01g to 0.81g with a percentage degradation of 100%. However, both composites degraded completely on the 12th day. This implied that P-BFcomposite was biodegradable but less degradable compared to NP-BF composite which degraded faster than the P-BF composite. The biofilm P-BF therefore may have a longer shelf life compared to the control (NP-BF). This is in agreement with the work done by Chika et al., (2024). The level of biodegradability of bioplastics buried in the soil decreases in mass with increasing time, this is because starch and glycerol have OH groups that play a role in initiating hydrolysis reactions so that they can absorb water from the soil which causes the polymer from starch to be decomposed into small pieces until the bioplastic fully decomposed in the soil (Chika et al., (2024).

The tensile strength was conducted to determine the maximum load that a material can withstand before it breaks (Widyowijatnoko et al., 2018).

The obtained results showed that the biofilms (P-BF) had less tensile strength compared to the control (N-BF) composite. Smaller tensile strength indicates that the material can easily deform in plastic behavior (Jaramillo et al., 2016). This could be due to the (N-BF) had a higher degree of homogeneity (compared to P-BF), causing the distribution of bioplastics constituent molecules to be consistently distributed. On the contrary, the sample (P-BF) had a smaller degree of homogeneity, making an uneven distribution of bioplastic constituent molecules (Arini, Ulum, and Kasman, 2017) due to the fibres in the plantain peels which greatly influenced the tensile strength.

Furthermore, the added plasticizer content (glycerol) strongly influenced the tensile strength of biofilms. The tensile strength of biofilms will be lower when the concentration is higher. This is due to the decrease in hydrogen bonds that occurs in the film, thereby increasing flexibility (Arini et al., 2017). Therefore, the tensile strength of the film gets smaller. Besides that, the resulting film was softer and more flexible. However, the synthesized biofilm met the required standard value for tensile strength which is between 1-10 Nmm$^{\wedge 2}$ (Arini et al., 2017).

The elongation of break test is carried out to determine the magnitude of the increase in the length of a polymer before finally breaking up. The value of elongation shows the ability of the film to elongate. This property depends on the type of film formation material which will affect

the cohesion properties of the film's bioplastic structure. The effect of a measurement of elongation of the break was carried out together with the analysis of tensile strength.

Higher elongation value means that the biofilm sample was more deformable. The sample P-BF had lower elongation value than (control) NP-BF, indicating the NP-BF sample can hold its shape better than the P-BF under the same elongation. This unique deformation behavior is likely due to the uneven distribution of the constituent molecules inside the sample body which is due to the fibres in the plantain peel. Indeed, this reduces the strong intermolecular interactions between starch molecules (due to the longer hydrogen bonding) (Warsiki and Bawardi, 2018).The percentage of elongation of an edible film is good, if the value is more than 50 % and bad if the value is less than 10 % (Chodijah, Husaini, Zaman, and Hilwatulisan, 2019). Additionally, NP-BF composites met the required standard value of percentage elongation which is between 10-20 %. Whereas the sample P-BF showed lower percentage elongation below the standard value probably due to the constituents molecules in the composite were not uniformly distributed (Budiman et al., 2017).

Flexural/ bending test was used to evaluate the response of biodegradable films when used under compressive load. The result of the flexural test showed that the P-BF had lower flexural strength than the N-BF. This could be due to the constituent's materials in the P-BF biofilm were not uniformly distributed compared to the NP-BF biofilm.

The hardness tests on the produced biofilms was conducted to evaluate its response when it is used under compressive load. This indicated that the thickness did not affect the hardness of the material.

The results from the mechanical test of the synthesized composites showed that the sample P-Bf had a lower tensile, percentage elongation as well as hardness compared to N-BF (control). The mechanical performance in P-BF was likely due to the constituents molecules in the composite were not uniformly distributed which is due to the presence of the plantain peels (Budiman et al., 2017).

The Gas chromatography flame ionization detector (GC-FID) was used to determine concentration of polycyclic aromatic hydrocarbons (PAHs) in the synthesized raw materials.

Table 4.20 showed PAH analysis of eggshell powder. Nine compounds were identified in the egg shell powder by GC-FID. Of which dibenzy(a,h)pyrene was identified to have the highest concentration followed by Benzo(k)fluoranthene and Phenanthrene .Other compounds were detected in substantial amounts such as Pyrenees,Acenaphthylene,1_2Benzanthracene, Benzo(c)chrysene, (Benzo(a)pyreneandBenzo(g_h_i)perylene. According to EC regulations commission (2011), BaP and PAH4 (Benzo(a)pyrene, Chrysene, Benzo(b)Fluoranthene and Benzo(a)Anthracene) are used as markers for assessing toxicity level of PAHS in food. The BaP in the eggshell powder and BaA exceeded the maximum acceptable limit of PAHS in plant based powders which is 0.01mg/kg (EU, 2020). The International Agency for Cancer Research classifies Bap as a group 1 agent which are carcinogenic to humans and BaA as group 2 agent which is termed as probable carcinogen (IARC, 2010).

Similarly, Table 4.21 shows PAH analysis of glycerol. Eight compounds were identified with the most prevailing polycyclic aromatic compounds as1_2benzanthracene, Fluoranthene, Benzo [a] pyrene and Pyrene. The rest of the compounds occurred in minimal quantities. Benzo[e]pyrene (0.10%), Phenanthrene and Perylene and Dibenzyl [a_h] anthracene. The BaP and 1_2 Benzanthracene which are used as markers for assessing PAHS toxicity in food were identified in the crude glycerol. The BaP as well as 1_2 Benzanthracene were detected in amount which exceeded the maximum acceptable limit of PAHS in vegetable oils and fat which is 0.002mg/kg (Alexander et al., 2008; EU, 2011). The International Agency for Cancer Research classifies BaP as a group 1 agent which are carcinogenic to humans (IARC, 2010).

The result of PAH analysis of vinegar showed seven compounds. The most abundant compounds identified included; Benzo[e]pyrene, 1_2 Benzanthracene (Pyrene, Phenanthrene and Benzo [g_h_i] perylene. Other compounds were identified in trace amounts such as; Anthracene and Fluoranthene.The 1_2 Benzanthracene in the vinegar exceeded the maximum acceptable limit of PAHS in food which is 0.002mg/kg (Alexander et al., 2008, EU, 2011). The International agency for Cancer Research classifies BaA as a group 2 agent which are probably carcinogenic to humans (IARC, 2010).

The result of PAH analysis of cassava starch showed four compounds were identified in the cassava starch by GC-FID. The most prevailing compound was identified as 1_2 benzanthracene while other compounds occurred in minimal amounts, these included; Benzo[b] Fluoranthene,

Pyrene and Benzo [g_h_i] perylene. The 1_2 Benzanthracene in the Cassava Starch and Benzo(b) fluoranthine which are used as a marker for assessing PAHS in food exceed the maximum acceptable limit of PAHS in plant based powders which is 0.01mg/kg.(EU, 2020). The International agency for Cancer Research classifies BaP as a group 1 agent which are carcinogenic to humans and BaA as group 2 agent termed probable carcinogen (IARC, 2010).

The PAHs are among the pollutants that are considered ubiquitous in the environment (Tobiszewski and Namiesnik, 2012). They could be emitted from processes that occur naturally such as volcanic eruptions, biomass combustion, and diagenetic processes (Wang et al., 2011). The toxicity of the soil containing the synthesized biodegraded bioplastic was assessed using GC-FID. The presence of PAHs in the powdered plantain peels was worrisome as this suggested that the pulp may have been contaminated too.

The result showed 15 PAH compounds including; napthalene , acenapthylene, acenaphthene, fluorene, phenanthracene, anthracene, fluoranthene, pyrene, benzo(a) anthracene, chrysene, benzo(k)fluoranthene, benzo(a)pyrene, indeno(1_2_3)pyrene, benzo(a_h)anthracene and benzo(a_h)perylene. Napthalene mean concentration in soil P (0.11±0.00) and soil W (0.11±0.00)were found to be significantly lower compared to the control(0.21 ± 0.00).

Acenaphthylene in soil P and soil W werelower when compared to the control. Likewise, acenaphthene in soil P and soil w were found to be lower compared to the control. Furthermore, Anthracene (concentration in soil Pand W were found to be lower compared to the control. However, fluorene concentration in soil P and soil W showed no significant difference compared to the control. Furthermore, fluoranthene concentration in soil P and W werelower compared to the control. Likewise, phenanthracene concentration in soil P and soil W were lower compared to the control.On the other hand, Pyrene concentration in soil P and soil W were higher compared to the control.

Benzo (a) anthracene concentration in soil P were higher compared to the control. While Benzo (a) anthracene in soil W showed no significant difference compared to the control.
On the other hand, Chrysene concentration in soil P and soil W were significantly lower compared to the control. Benzo (a) pyrene concentration in soil P and soil W were lower compared to the control. Likewise, Benzo(a_h)anthracene in soil P were higher compared to the

control.Whereas benzo(a_h)anthracene in soil W did not differ with that of the control.Indeno(1_2_3)pyrene concentration in soil Pand soil W were lower compared to the control. The Benzo (a_h) perylene in soil P and soil W were lower compared to the control.

The result of the toxicity test also indicated that PAHs in soil P such as napthalene , acenapthylene, acenaphthene, fluorene, phenanthracene, anthracene, fluoranthene, benzo(a) anthracene, ,benzo(k) fluoranthene,benzo(a)pyrene, indeno(1_2_3)pyrene, benzo(a_h)anthracene and benzo(a_h)perylene were lower compared to the control with exception of chrysene and pyrene which were found to be higher compared to the control. Additionally, acenapthylene were found to be the highest while flourene showed no significant difference. Likewise the PAH compounds indicated in soil W such as napthalene , acenapthylene, phenanthracene, anthracene, fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, indeno(1_2_3)pyrene, benzo(a_h)anthracene and benzo(a_h)perylene were found to be lower compared to the control with exception of pyrene which were higher compared to the control. Furthermore, acenaphthene, fluorine, benzo(a) anthracene did not differ with that of the control. The results of the present study also showed that the control had higher mean concentration of PAH compounds compared to the soil samples containing degraded biofilm.

Atmospheric deposition could contribute to high PAH concentrations in soil by virtue of the economic and other anthropogenic activities in such areas (Marr et al., 2006; Tian et al., 2009; Opara et al. 2016; Njoku et al., 2016; Ibe et al., 2020).

In addition, the high permeability and porosity of the (control) soil may have contributed to the increased concentration of PAHs (Onyeagocha 1980; Ejiogu et al. 2019; Ibe et al. 2020). The above-stated factor may have influenced the PAH levels observed in the control.

However, the concentration of PAHs reported in the present study was lower than 1mg/kg recommended for soil cleanup by the Department of Petroleum Resources, Nigeria (DPR, 2002). Similarly, PAH values in the present study were within the allowed limits of 1mg/kg, 1500 µg/kg, and 5 mg/kg stipulated guidelines for soil cleanup by Denmark, Netherlands, and Australia, respectively (ANZECC 2000; MHSPEN 2000; DEPA 2002).

CHAPTER FIVE

CONCLUSION AND RECOMMENDATION

5.1 CONCLUSION

The present study showed that the synthesized biofilm contained important functional groups which makes the biofilm safe for human use as well the environment. The compounds identified by the GC-MS in the study were completely different from previous reports by other authors. The difference in composition of these extracts could be due to the pre-treatment conducted before extraction as well as difference in the specie of the raw materials.

The synthesized raw materials had PAHs above the required limit. However, the biofilm synthesized using the same raw materials were within the permissible limit for PAH toxicity in the soil. It can also be deduced from the present study that there was no much change in the integrity of biofilm material when soaked in organic solvents. Furthermore, the synthesized biofilm had less engorgement which are undoubtedly desired results for preparation of bioplastics. This showed that the plantain peel biofilm are stable. Also, the plantain peel based biofilm had good thermal stability as well as high biodegradation property.

With regards to thermal degradation, it has been determined that, for P-bF matrices, it starts at 130 °C, while N-PF fibers start degrading at 139 °C. Therefore, the processing temperature for ripe plantain peels is set to below 137 °C to avoid unwanted degradation of the material.

5.2 RECOMMENDATION

The cause of the uneven distribution of the constituent materials in the biofilm with eggshells and plantain peels should be investigated. Re-enforcement materials such as bamboo, hemp and flax should be considered in further study to enhance the mechanical properties.

5.3 CONTRIBUTION TO KNOWLEDGE

1. The study showed that an eco-friendly biodegradable biofilms can be synthesized from ripe plantain peels at 3% plantain peel, 1% acetic acid, 3% cassava starch, 1% eggshell and 3% glycerol.
2. The use of ripe plantain peels for production of biodegradable biofilm increases the availability of raw materilas due to the availability of the waste biomass.
3. Valorisation of food waste such as ripe plantain peel, eggshells and pineapple peels can help reduce environmental pollution as well as minimize the cost of solid waste management in Nigeria.
4. The synthesized biofilm showed 100% biodegradablility with complete degradation after six days and thus can serve as a substitute to the conventional plastics and also serve as a great potential to overcome environmental pollution.
5. Polycyclic aromatic hydrocarbons can be found in waste biomass.

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APPENDIX

Plate 3: Ripe plantain peels

plate 4: Grounded plantain peel.

Plate 5: Bleached palm oil

plate 6: Crude glycerol

Plate 7: Cassava root

plate 8: Cassava starch

Plate 9: pineapple peel

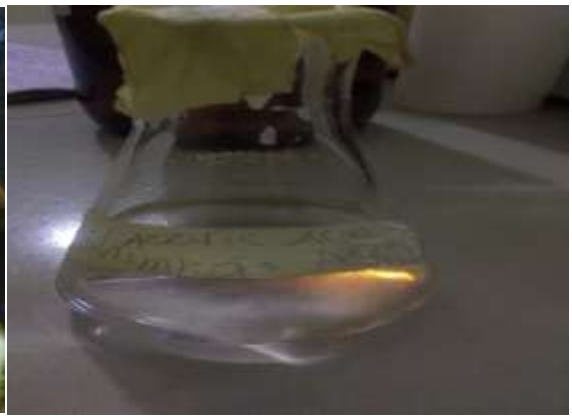


Plate 10: Acetic acid (vinegar)

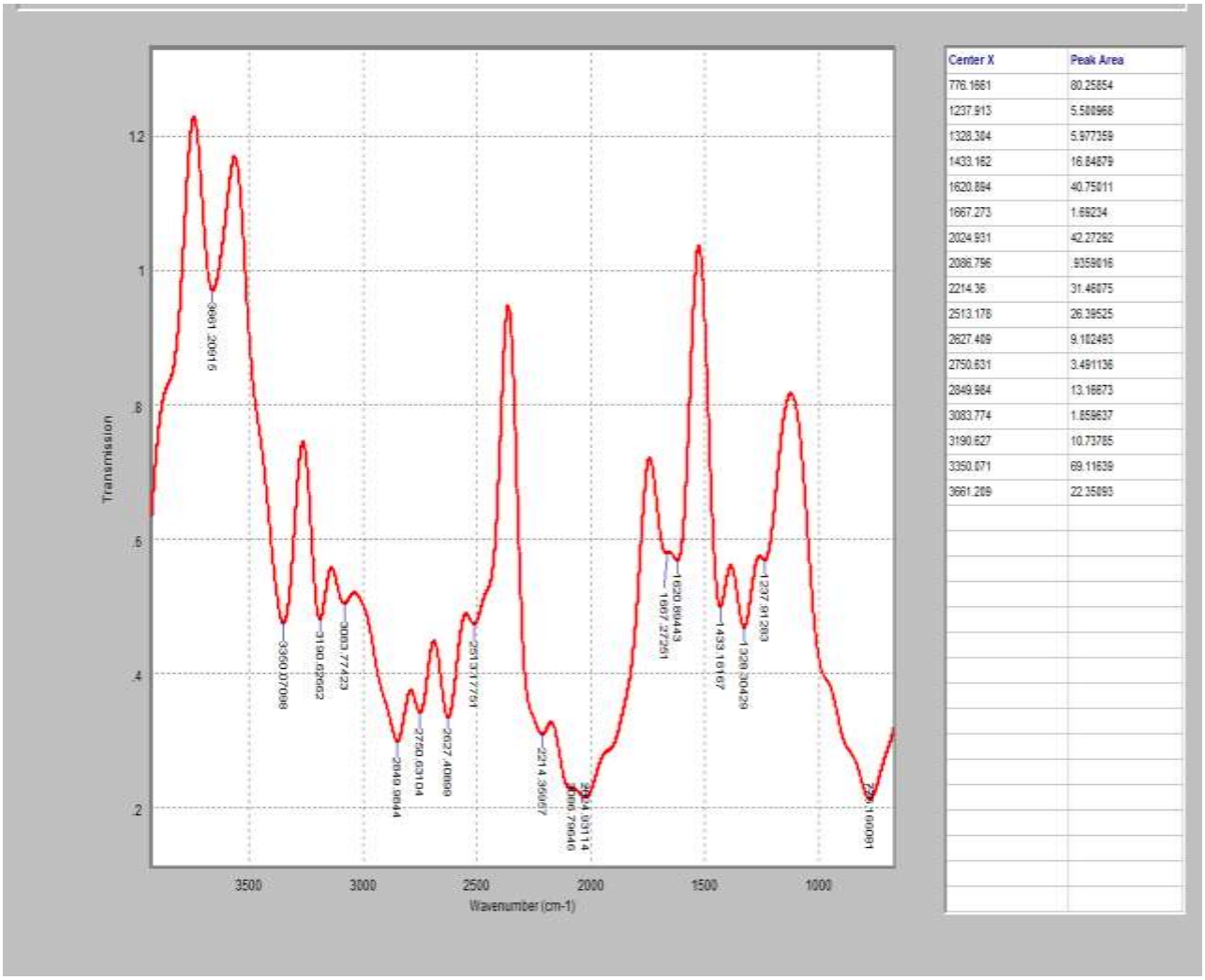
Plate 11: Egg shells

Plate 12: Egg shell powder

Plate 13: P-BF **Plate 14: NP-BF**

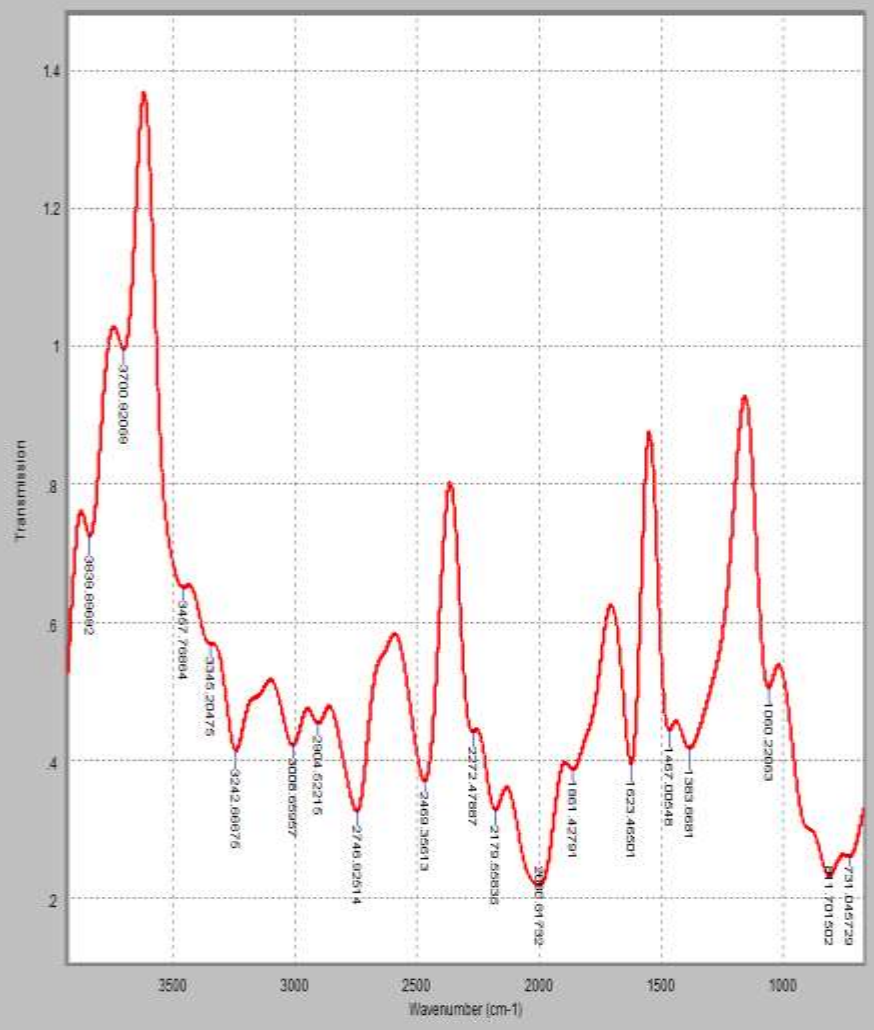
(plantain peel based biofilm) **(non-plantain peel based biofilm)**

Appendix 1: FTIR analysis of plantain peels



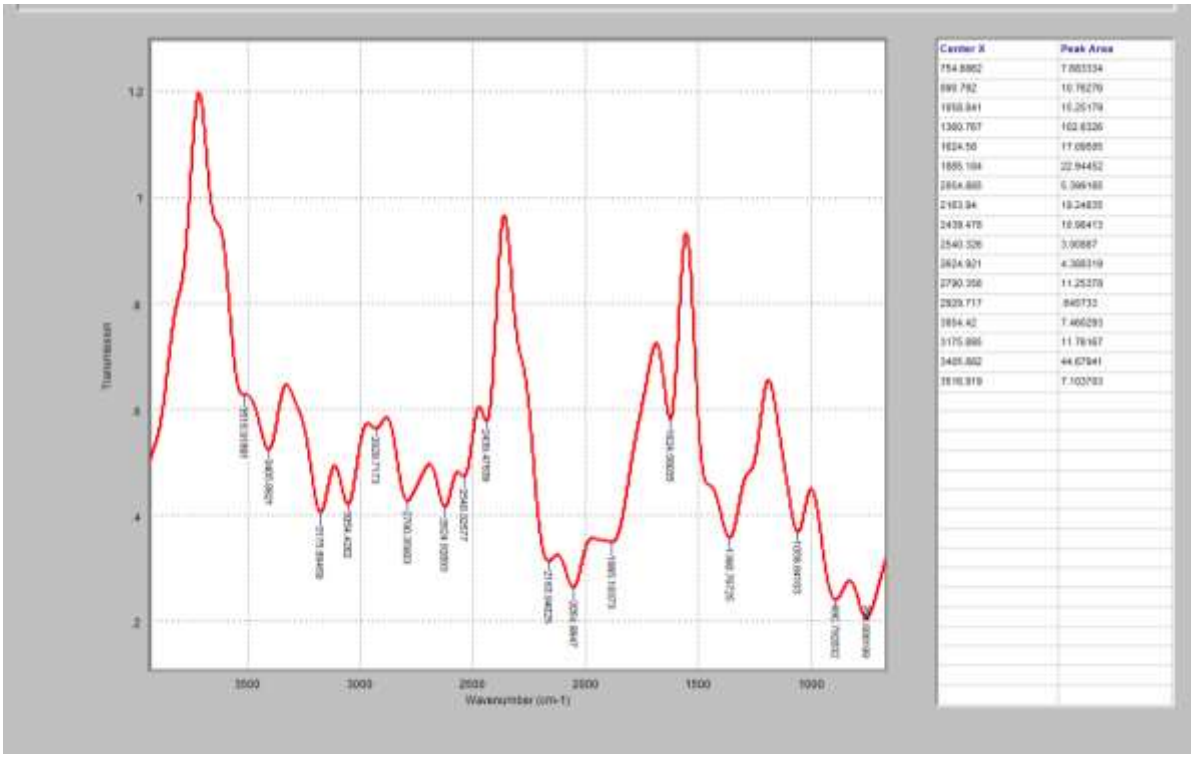
Center X	Peak Area
776.1661	80.25854
1237.913	5.580968
1328.304	5.977359
1433.162	16.84879
1620.894	40.75011
1667.273	1.69234
2024.931	42.27262
2086.796	.9359016
2214.36	31.46875
2513.178	26.39525
2627.409	9.102493
2750.631	3.491136
2849.984	13.16673
3083.774	1.859637
3190.627	10.73785
3350.071	69.11639
3661.209	22.35893

Appendix 2: FTIR analysis of eggshells

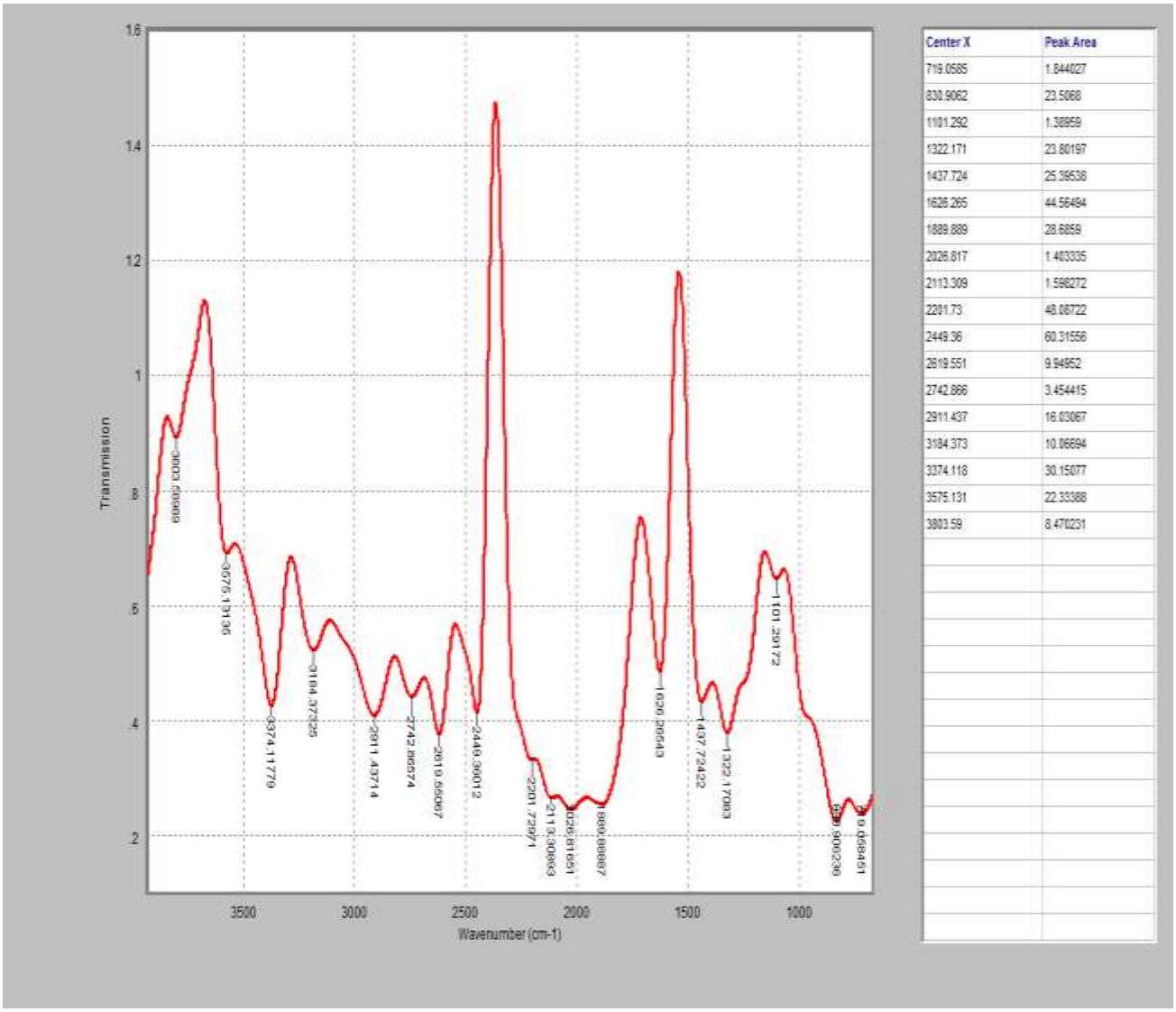


Center X	Peak Area
731.0457	1.379663
811.7015	18.45471
1060.221	15.3371
1383.668	126.3095
1467.005	8.65537
1623.465	29.22818
1861.428	9.488841
2000.617	22.01357
2179.558	4.247753
2272.479	4.789559
2469.356	41.60336
2746.925	21.39221
2904.522	1.028292
3008.66	5.227518
3242.667	13.20477
3345.205	1.413045
3457.769	26.74094
3700.921	13.14047
3839.697	8.323928

Appendix 3: FTIR analysis of cassava starch

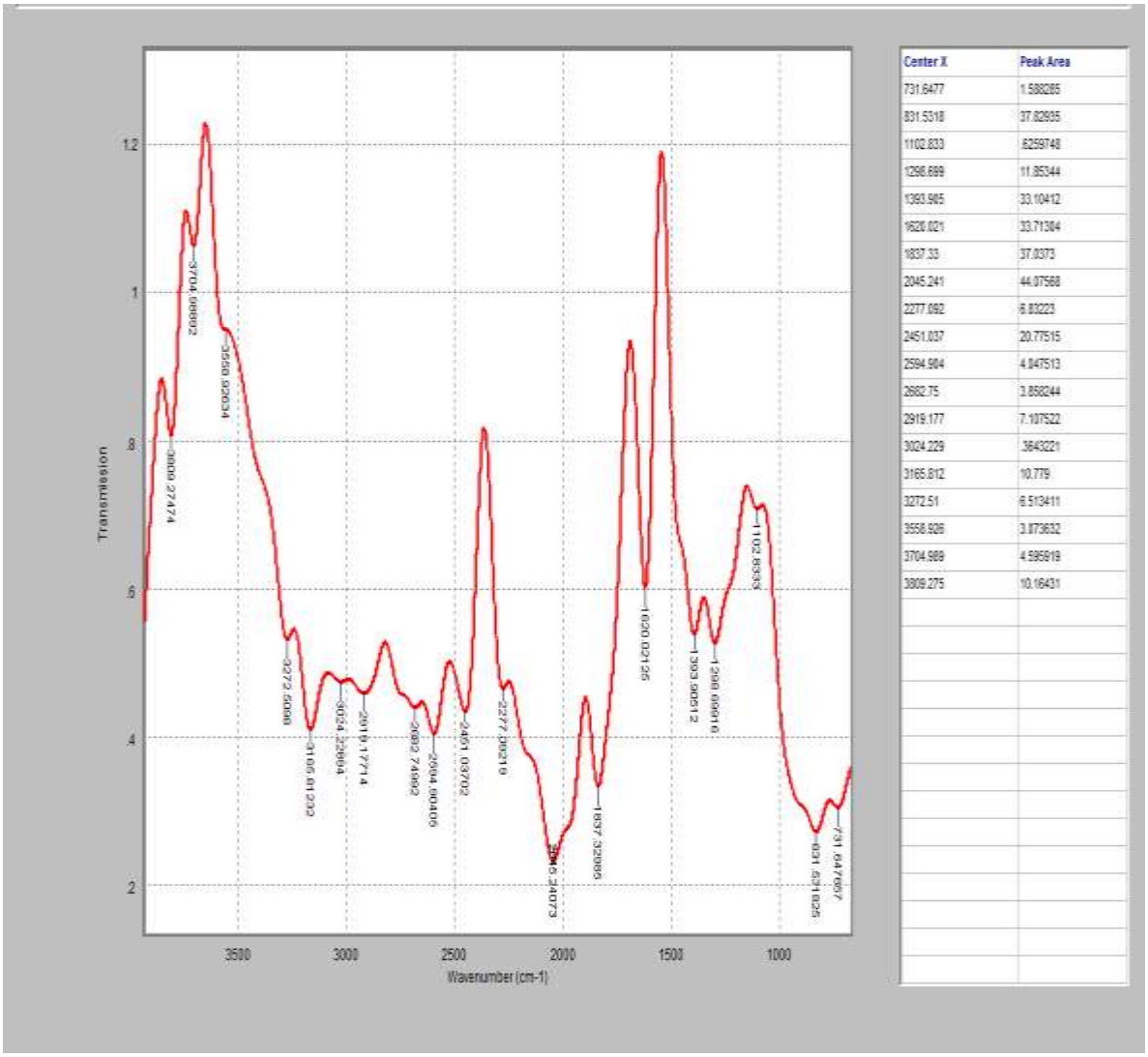


Appendix 4: FTIR analysis of vinegar



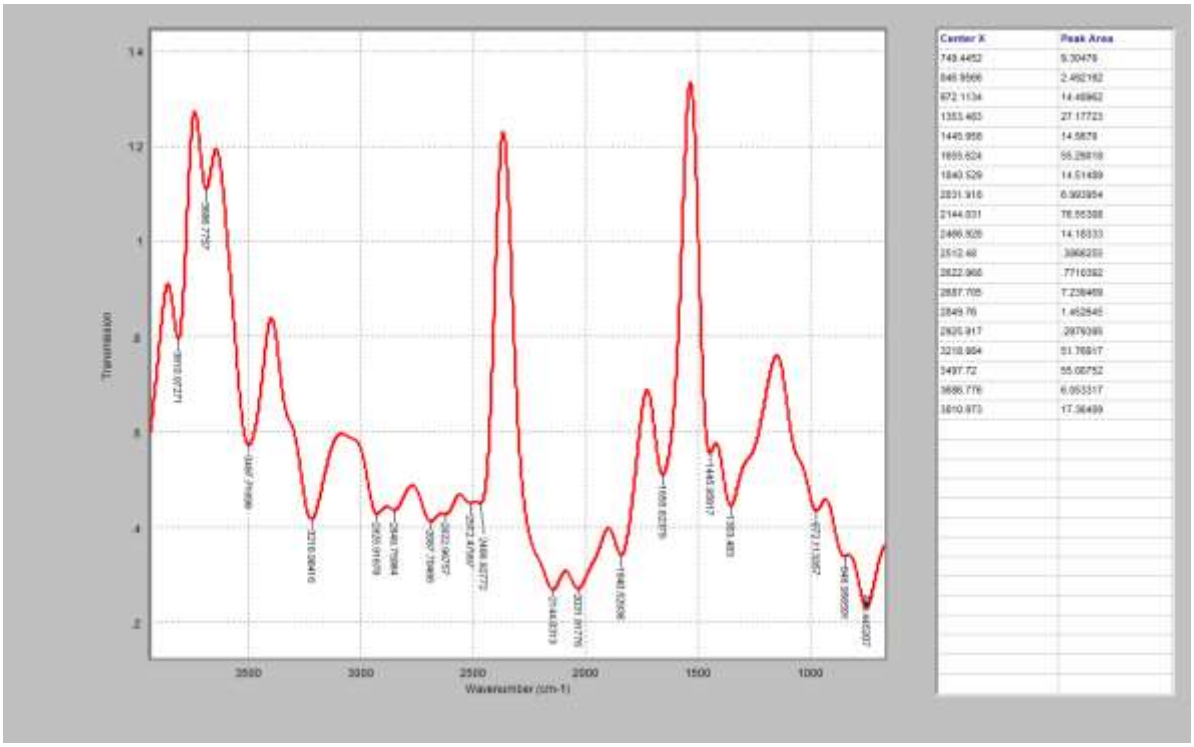
Center X	Peak Area
719.0585	1.844627
830.9062	23.5068
1101.292	1.38959
1322.171	23.80197
1437.724	25.39538
1626.265	44.56494
1889.889	28.6859
2026.817	1.403335
2113.309	1.596272
2201.73	48.06722
2449.36	60.31556
2619.551	9.94952
2742.866	3.454415
2911.437	16.03067
3184.373	10.06694
3374.118	30.15077
3575.131	22.33388
3803.59	8.470231

Appendix 5: FTIR analysis of Glycerol

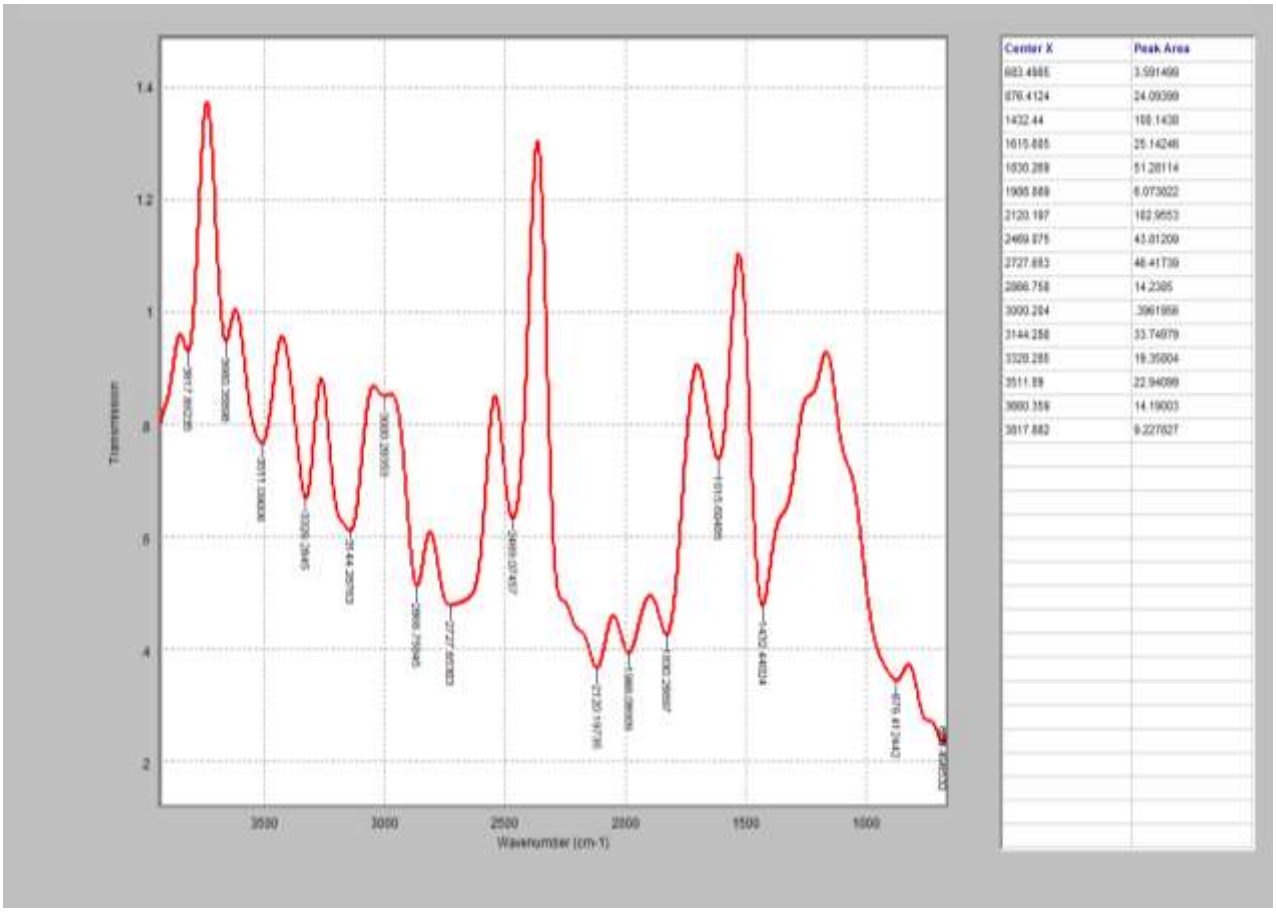


Center X	Peak Area
731.6477	1.598285
831.5318	37.82935
1102.833	.6259748
1296.699	11.85344
1393.905	33.10412
1626.021	33.71384
1837.33	37.0373
2045.241	44.97568
2277.092	6.83223
2451.037	20.77515
2594.904	4.947513
2682.75	3.858244
2919.177	7.107522
3024.229	.3643221
3165.812	10.779
3272.51	6.513411
3558.926	3.873632
3704.989	4.595919
3809.275	10.16431

Appendix 6: FTIR analysis of P-BF

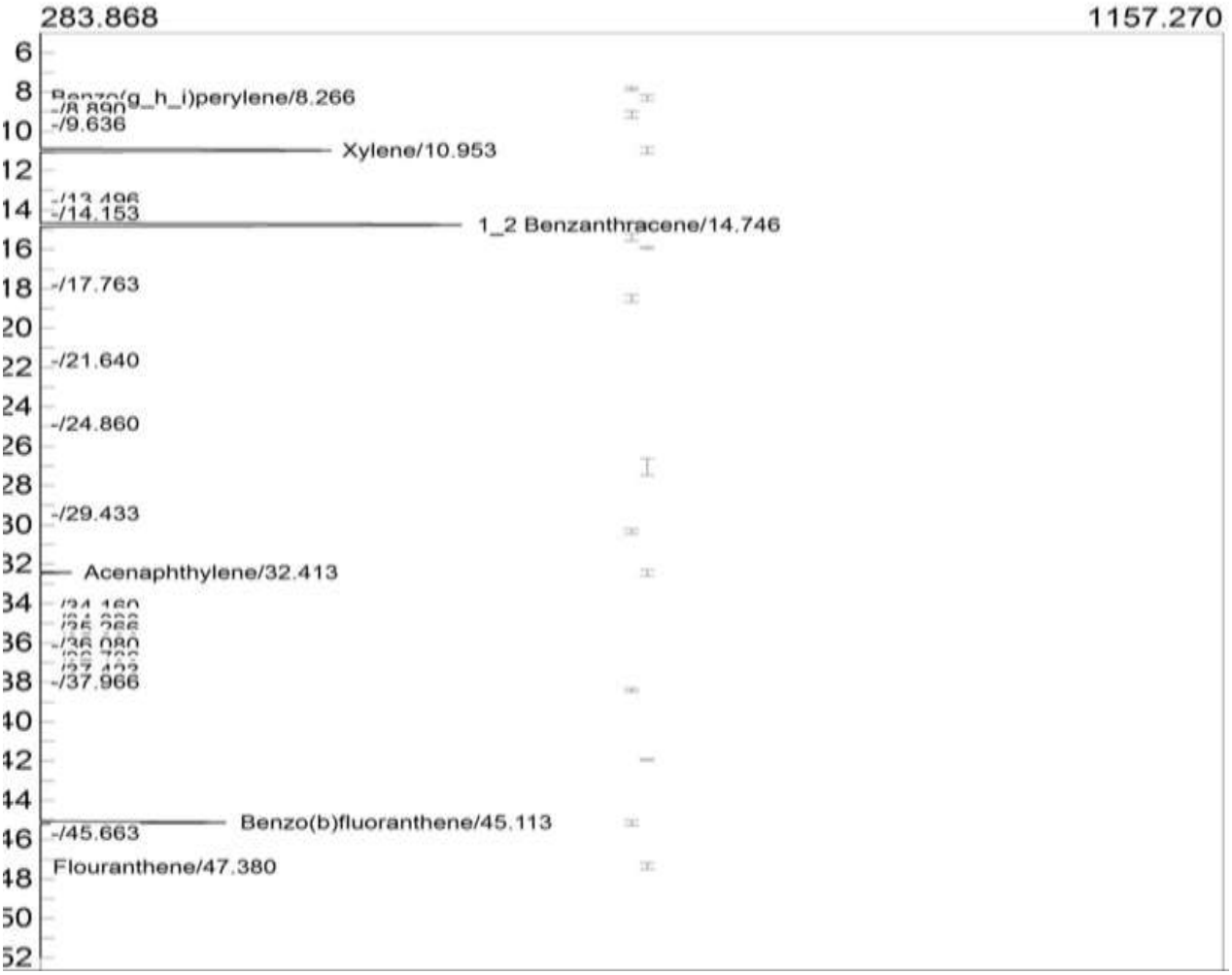


Appendix 7: FTIR analysis of NP-BF



Center X	Peak Area
683.4985	3.591469
876.4124	24.09399
1432.44	108.1438
1615.685	25.14246
1630.289	51.28114
1986.889	6.073822
2120.197	182.9653
2469.975	43.81209
2727.683	48.41739
2886.758	14.2385
3000.284	.3961956
3144.258	33.74979
3328.285	19.35804
3511.89	22.94099
3680.359	14.19003
3817.882	9.227027

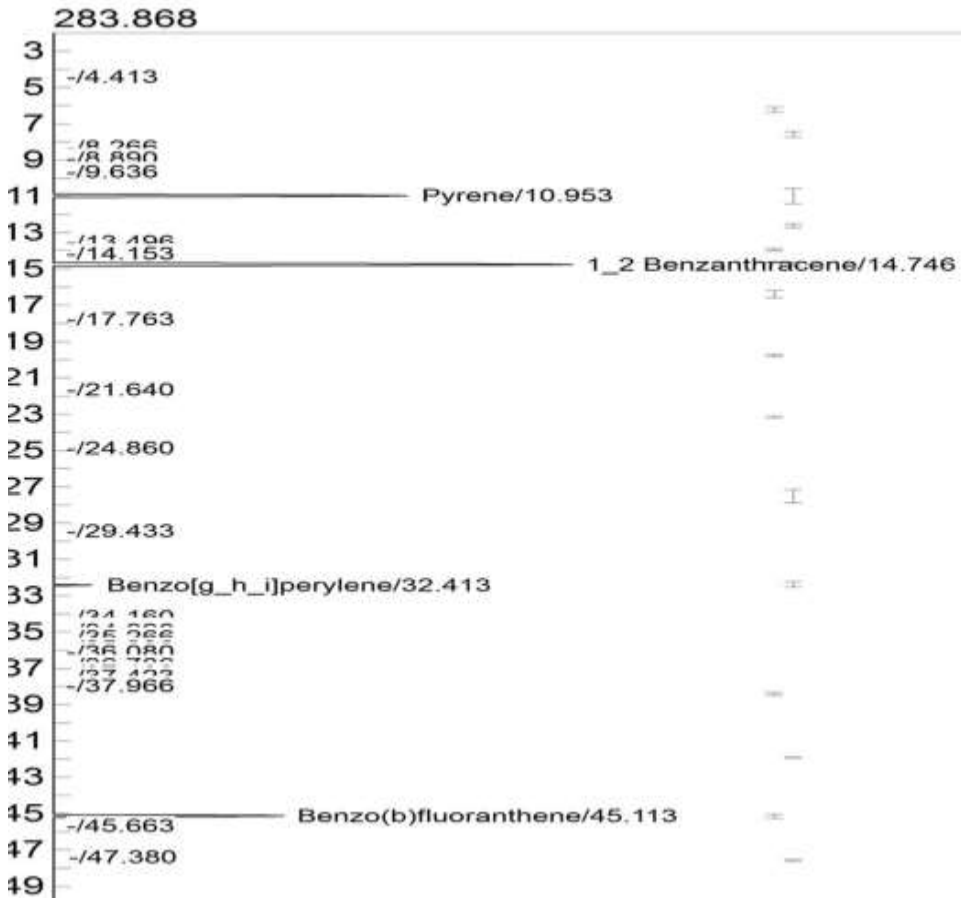
Appendix 7: PAH analysis of plantain peels



Appendix 9: PAH analysis of acetic acid (vinegar)

Component	Retention	Area	Height	External	Units
Fluoranthene	10.793	4775.5038	375.479	0.1492	mg/ml
1_2 Benzanthracene	12.450	4743.7844	372.795	0.4299	mg/ml
Phenanthrene	14.040	4579.5053	360.005	0.3231	mg/ml
Anthracene	19.756	3930.6483	309.146	0.0968	mg/ml
Benzo[e]pyrene	21.553	5025.1523	394.848	1.3854	mg/ml
Pyrene	24.580	4953.9757	389.494	0.4166	mg/ml
Benzo[g_h_i]perylene	31.363	4199.9884	329.743	0.2871	mg/ml
		32208.5582		3.0880	

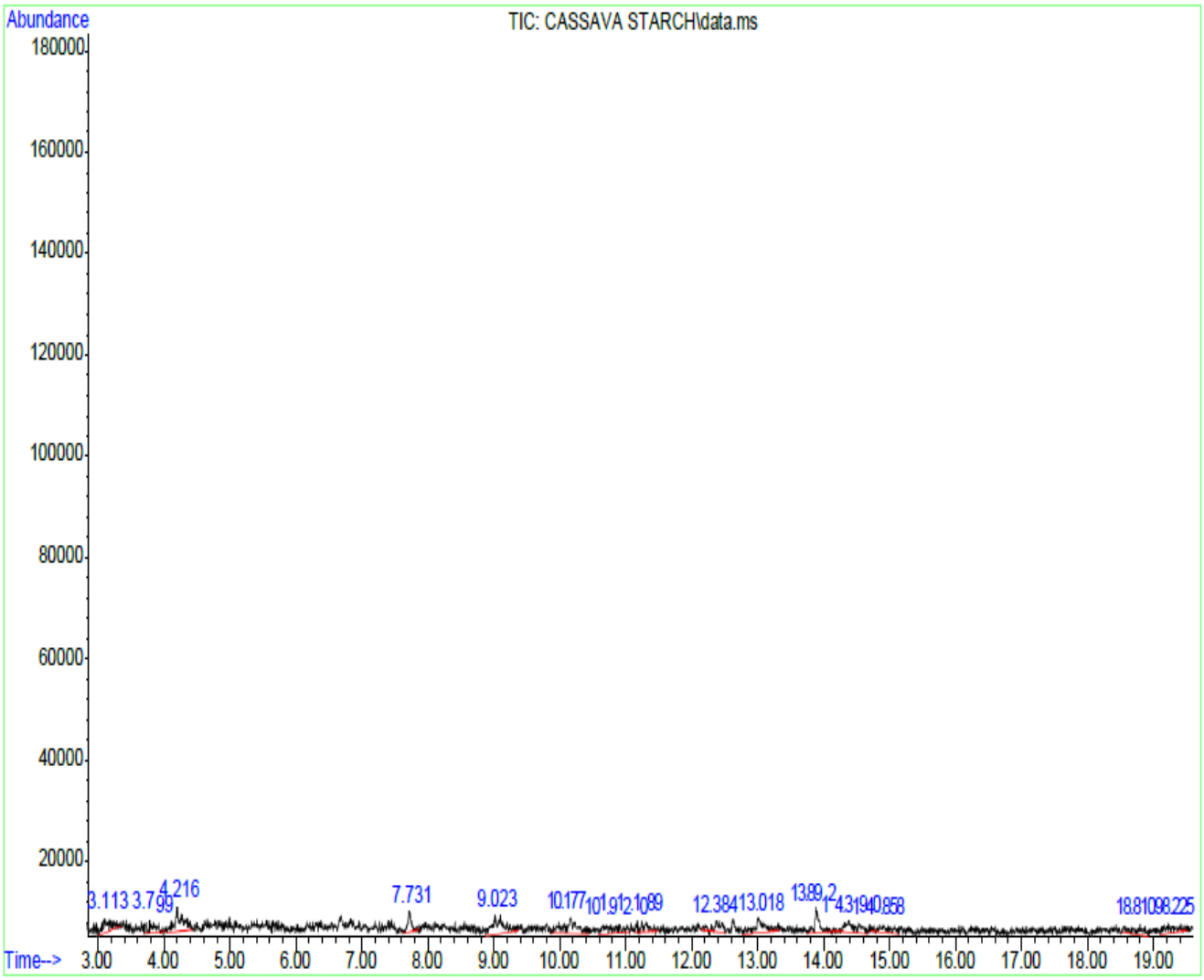
Appendix 10: PAH analysis of cassava starch



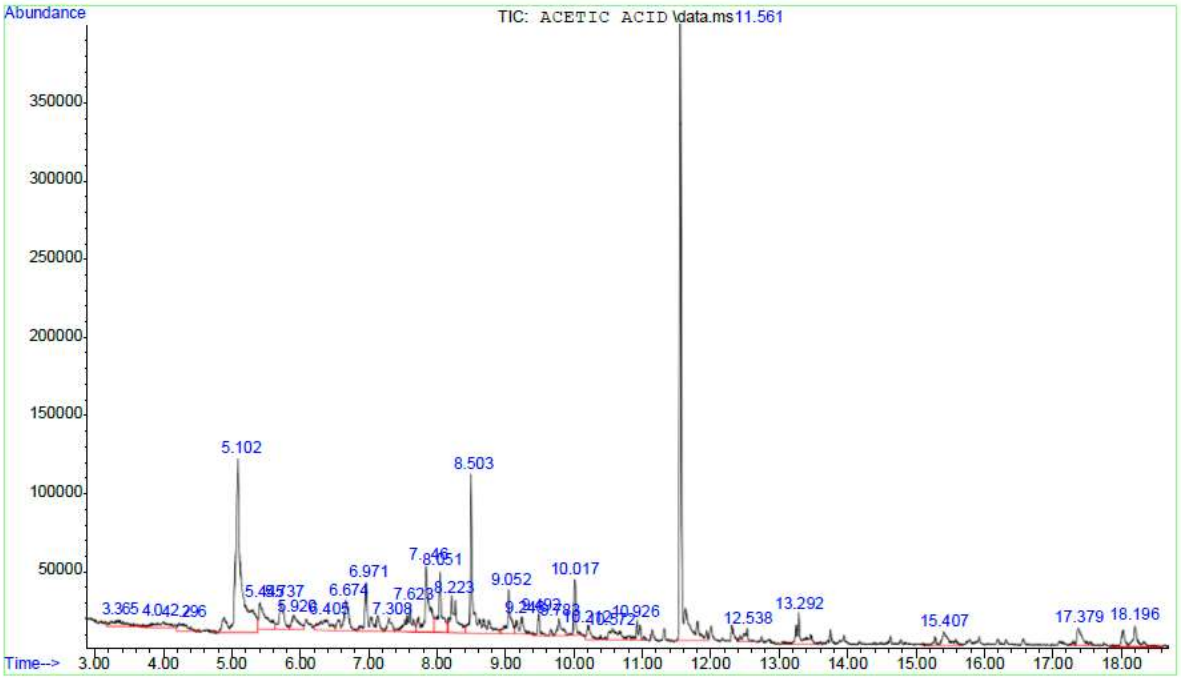
Appendix 11: PAH analysis of glycerol

Component	Retention	Area	Height	External	Units
Benzo[a]pyrene	5.793	5819.4134	456.746	0.4254	mg/ml
Perylene	8.436	4721.8424	371.188	0.0014	mg/ml
Fluoranthene	10.793	4775.5038	375.479	0.7674	mg/ml
1_2 Benzanthracene	12.450	4743.7844	372.795	1.3833	mg/ml
Phenanthrene	14.040	4579.5053	360.005	0.0219	mg/ml
Dibenzyl(a_h) anthracene	19.756	3930.6483	309.146	0.2034	mg/ml
Benzo[e]pyrene	21.553	5025.1523	394.848	0.0033	mg/ml
Pyrene	24.580	4953.9757	389.494	0.3207	mg/ml
		38549.8256		3.1269	

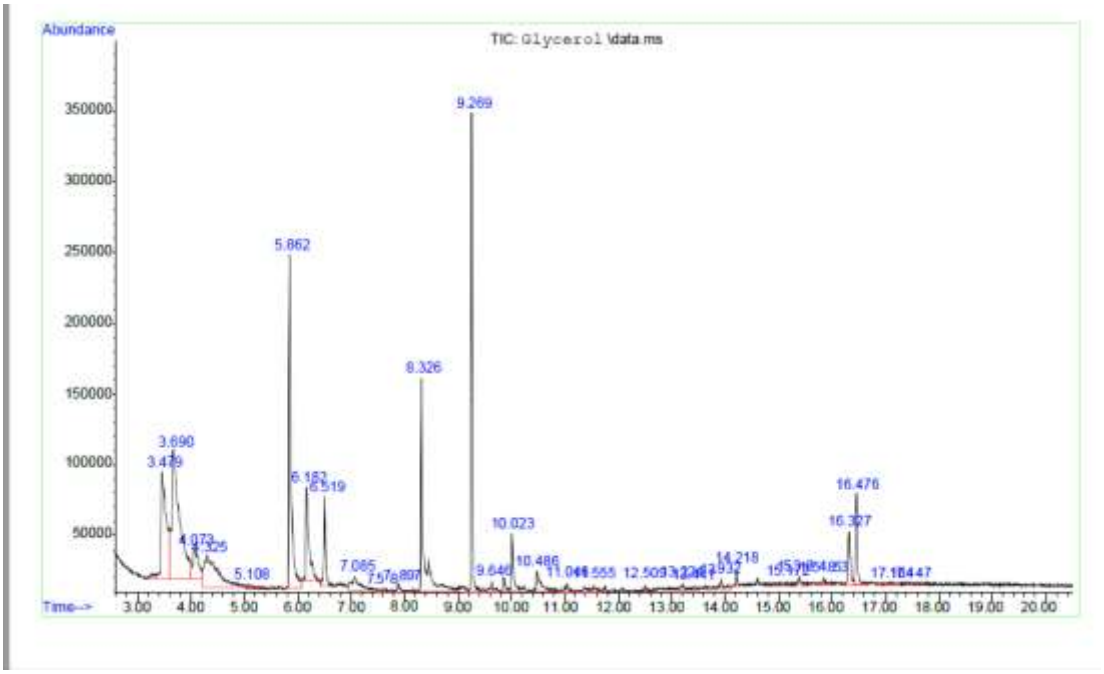
Appendix 12: GC.MS chromatogram of cassava starch



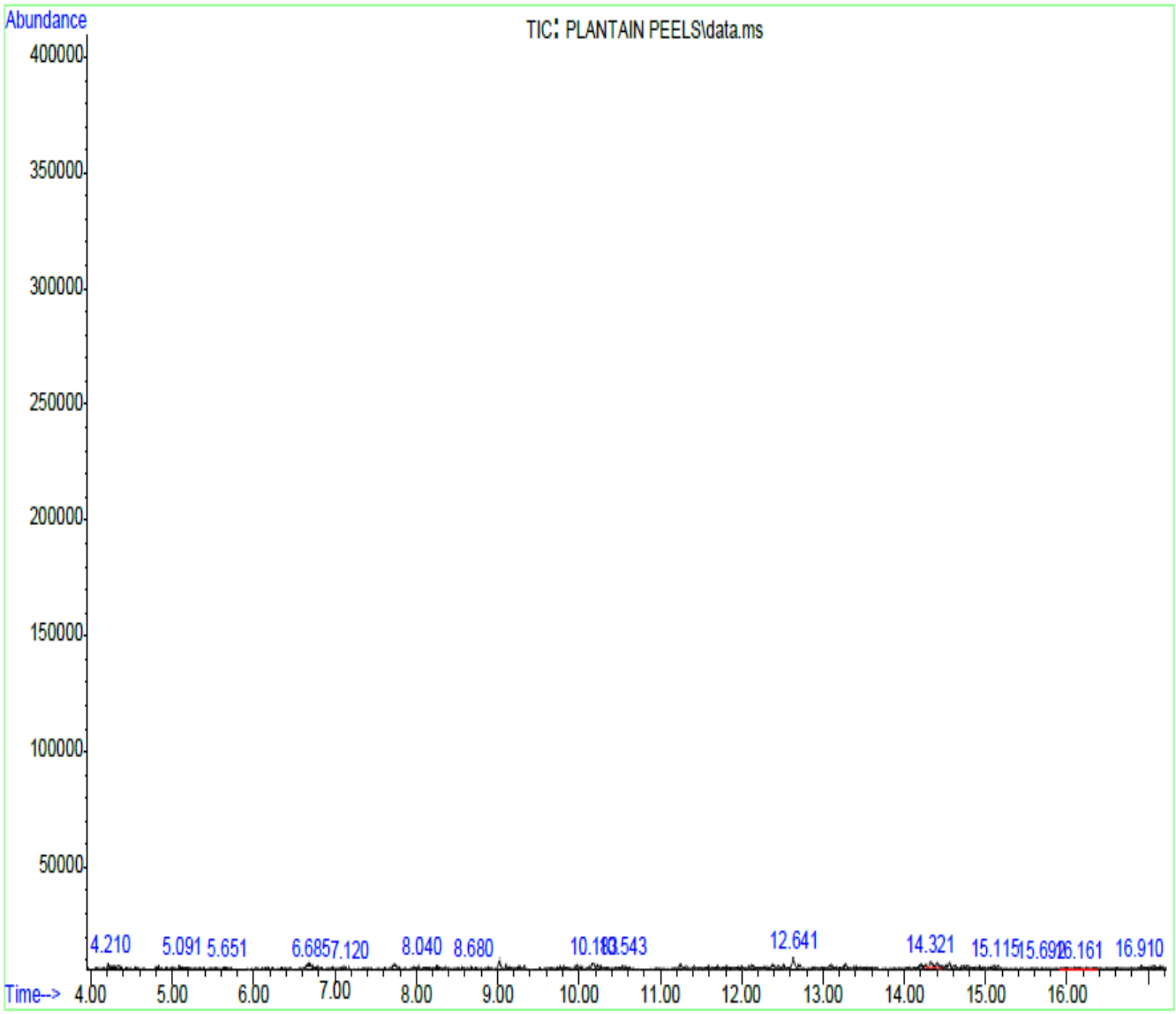
Appendix 13: GC.MS chromatogram of acetic acid (vinegar)



Appendix 14: GC.MS chromatogram of crude glycerol



Appendix 15: GC.MS chromatogram of plantain peels



Appendix **16: GC.MS chromatogram of eggshell**