Jimma University

Jimma Institute of Technology

School of Chemical Engineering

Process Engineering Stream

Synthesis and Characterization of Bioplastic Film from Banana Peel Starch Blending with Banana Pseudo-Stem Fiber

A Thesis Submitted to Research and Graduate Studies in Partial Fulfillment of the Requirements of Degrees of a Masters science in Chemical Engineering under Process Engineering Stream

By: Workiye Getnet

January, 2019

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Advisor: Dr. K.Ramachandran

Co-Advisor: Mr. Gebeyehu Abu

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Jimma, Ethiopia

DECLARATION

I declare that this thesis entitled “**Synthesis and characterization of bioplastic film from banana peel starch blending with banana pseudostem fiber**” for the M.Sc. Degree at Jimma University, hereby submitted by me, is my original work and has not previously been submitted for a degree at this or any other university, and that all resources of materials used for this thesis have been duly acknowledged.

Name: Workiye Getnet Abera

Signature: __________

Date of Submission: __________

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APPROVAL

The thesis entitled “**Synthesis and Characterization of Bioplastic Film from Banana Peel Starch Blending with Banana Pseudo-Stem Fiber**” submitted by Workiye Getnet Abera is approved and accepted as a Partial Fulfillment of the Requirements for the Degree of Masters of Science in Chemical Engineering under Process Engineering Stream.

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Main Advisor:	Dr. K.Ramachandran	___________	___________
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As members of the examining board of MSc. thesis, we certify that we have read and evaluated the thesis prepared by Workiye Getnet Abera. We recommend that the thesis could be accepted as a Partial Fulfillment of the Requirements for the Degree of Masters of Science in Chemical Engineering under Process Stream.

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ABSTRACT

Biomass has the potential to reduce greenhouse gas emissions by replacing fossil fuel.The exponential growth of the human population has led to the accumulation of non-degradable waste materials across our planet. Bioplastics synthesized from only starch has less mechanical strength and thermal stability, and can be improved by the addition of fillers. In this study, banana peel starch and banana pseudostem fiber were used for the synthesis of bioplastic film. Proximate analysis of banana peel was moisture (10.5%), total ash (8.5%), crude lipids (6.66%), crude protein (2.17%), crude fiber (34.37%), and carbohydrates (37.8%).The optimum process variables for starch yield were 27.66 minutes, 37.92°C temperature which gives 12.06% maximum yield. The extracted starch was characterized as pH (6.31), moisture (7.2%), loss on drying (11.2%), amylose (19.3%), amylopectin (80.7%) and ash content (0.8%). The thermal resistance of fiber was characterized by TGA. For fiber density (1.43g/cm^3), yield (8%), moisture content (61.3%), water absorptions (4.6%), and cellulose (55%-60%) has been investigated. Film was synthesized by conducting 20 experiments and analyzed by design expert 11, by considering three factors of dry oven temperature(35-65°C), glycerol concentration (20-50%)w/v and fiber concentration (5-20%) w/w of starch(5g) basis. Three responses were tensile strength (TS), water absorption (WA), and elongation at the break (EA) of the synthesized bioplastic film. The result obtained in ranges were (0.2-7.25Mpa) TS, (4.9-31.01%) EA and (18.6-35.3%) WA. The obtained results at the optimal point were 7.23Mpa TS, 5.46% EA and 18.60% WA at a combination of 56.5°C dry oven temperature, 28.6%glycerol, and 18.26% fiber concentrations. Moisture, solubility, transparency, and thickness of the film were determined. The film also characterized by TGA, DSC and FTIR. This study indicates that higher strength, low water absorbent, and good thermally stable bioplastic film can be produced from banana peel starch via banana pseudostem fiber.

Key words: *Banana Peel Starch, Banana Pseudo-Stem Cellulosic Fiber, Bioplastic Film, Bio Polymers, DSC,TGA*

ACKNOWLEDGMENTS

I would like to thank the Almighty GOD for giving me the strength and wisdom to successfully complete this thesis for his protection and strength, an ever-present help in the entire situation and challenge that I face.

I wish to express my appreciation to my advisor, Dr. K.Ramachandran, for his guidance and advice. His understanding of attitudes and constructive suggestions, as well as his great patience, undoubtedly was the utmost support for me to succeed in my thesis work. I also thank my co-advisor Mr.Gebeyehu for his continues comment.

I am so grateful to the college of agriculture at Jimma University, which allowed me to conduct the experiment. I give deep thanks to Mr.Gelana and Ms. Nesra for their help during my laboratory work at the microbiology lab. Also, I thank the material department lab technicians Mr. Dita and Ms.Tiruework for their help. I am also deeply thankful to chemical engineering lab technicians Mr. Dingeta and Worku for their help and good friendships. I thank Bahr Dar University (poly campus, Blue Nile lab technicians), and Abay paper and printing factory quality control department workers.

I thank JIT to be my sponsor and giving me a chance to join this master's program. Dear friends, Samuel Lativo, Merid Debebe, Getachew Alemu, and Tilahun Teshome thank you for the friendship and support. I want to give my heart felt gratitude for Mamiye. Finally, I acknowledge my family with the deepest gratitude who encouraged me to strive toward academic-excellence and always wish the best for them.

May the Almighty God bless all of you!!!

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABBREVIATION

ANOVA	Analysis of variance
BIO-PE	Bio-polyethylene
BIO-PVC	Bio-polyvinyl chloride
$C_6H_{10}O_5$	Starch
$C_6H_{12}O_6$	Glucose
CCD	Central composite design
CV	Coefficient of Variation
D.oven tem	Dry oven temperature
EA	Elongation at the break
FB	Fiber
FTIR	Fourier-transformed infrared ray spectroscopy
GHG	Greenhouse gases
Gly.coc	Glycerol concentration
JIT	Jimma institute of technology
NaOH	Sodium hydroxide
PRESS	Predicted residual error sum of squares
RSM	Response surface method
SNNP	Southern nation, nationalities, and peoples
Tc	Crystal temperature
Td	Decomposition temperature
Tg	Glass transition temperature
TGA	Thermogravimetric analysis
Tm	Melting temperature
TS	Tensile strength
WA	Water absorption

1. INTRODUCTION

1.1. Background

Polymers are (Greek: poly-many, meros-particle) which are compounds with large molecular mass, built of interlinking, basic building blocks, called monomers. When those polymers bunch together form plastics. Plastics are relatively low cost, durable and versatile materials. Plastics have brought some important to society in terms of economic activity, jobs, and quality of life. However plastic wastes also impose negative environmental impacts. It is usually non-biodegradability and therefore can remain as waste disposed of in the environment for a very long time; it may pose risks to human health as well as the environment, and it can be difficult to reuse and/or recycling in practice (Luengo & Ã, 2003).

Most polymers are originated from renewable resources which are biomass (John & Thomas, 2008). Biomass has the potential to reduce greenhouse gases (GHG) emissions by replacing fossil fuel. Bio-based plastics consist of biodegradable and are recycled. The mechanical properties are similar to those derived from fossil, for example, bio-polyvinyl chloride (Bio-PVC), bio-polyethylene (Bio-PE) originated from sugarcane (Ghuttora, 2016). Biodegradation for biopolymer is breaking into smaller particles with the help of microbial activity and converted into biomass, water, and carbon dioxide but, some conventional plastics are degraded after a long period of time into methane and carbon dioxide (Vega-Castro *et al.*, 2016).

Cellulose, lignin, and starch are commonly available in nature. Cellulose is abundant in most plants, although some plants produce more than others. Wood contains lignin, and starch is commonly found in plants such as corn, potatoes, banana peels and wheat (Gadhave *et al.*, 2018). Starch is a natural biodegradable polymer that has the potential to substitute synthetic polymers used for limited-time applications (Lubis and Harahap, 2018).

Fiber from the banana plant is best in physical strength and cellulose content relative to fiber obtained from other fibrous commodity by-products, extracted from their fruit stalk, pseudostem, and leaves. Fiber from banana is environmentally friendly and alternative to produce food packaging plastics, dry good packaging plastics or other reinforcing fiber utilized in engineering composites (Kumar *et al.*, 2013). The latest studies have indicated that banana fiber possesses a lot of importance on chemical and physical properties which can be used as a very good raw material for the textile and packaging industry (Vigneswaran *et al.*, 2015).

Approximately almost 90 million tons of banana fruit is produced, mainly in tropical areas (Pyar & Peh, 2018). In Ethiopia, bananas occupy nearly 60% of the harvested area of fruit crops, in 2017/18. Ethiopia produces 494,000 metric ton banana (Bickford, 2018). It produced in the Southern Nations, Nationalities and Peoples (SNNP) and the Oromiya regional states. In the Arba Minch, about 2500 hectares of banana are produced. Another second production area in the southwestern part of the country found around Jimma, Sheka, Kaffa, and Bench Maji zones in Southern and Oromiya regional states. In Amhara, Tigray, Benishangul-Gumuz, and Gambella are mainly for domestic production. However, due to irrigation systems, it produced in Afar and Somali (Alemu, 2016). We have such resources, but our country imports plastic raw materials 100%.

Banana pseudostem cellulosic fiber also produced from the waste banana stem. Therefore, it is possible to obtain a banana sufficiently and the banana peel is rich in starch to use for the synthesis of biodegradable bioplastic films. The physical, mechanical and thermal property of the produced bio-plastic film was improved by the addition of cellulosic fiber as filler.

1.2. Statement of the problem

Currently, the world has faced side effects from fossil fuel dependence such as greenhouse gas (GHG) emissions, environmental pollution, and ocean acidification. The exponential growth of the human population has led to the accumulation of huge amounts of non-degradable waste materials across our planet (Luengo & Ã, 2003). During plastic production, there is a great emission of pollutant gasses. Since conventional plastics use the by-product of fossil fuel as the main feedstock, this causes also a depletion of a natural resource.

Many researchers have been searched to reduce the adding number of plastic wastes into the environment. Biodegradable bioplastics are being studied to aid the pollution of plastics, since synthetic plastics not degraded for a long period of time, consume more energy during production, and their non-recycling property. During the degradation of conventional plastics, there is an emission of pollutant gases such as carbon dioxide and methane to the soil and into the atmosphere. Conventional plastics affect plants, animals, micro-organisms and pollute the entire water bodies after disposal.

To overcome the above challenges it is advisable to synthesis clean, renewable, biodegradable, eco-friendly and almost sustainable bioplastics. The biodegradable bioplastics, synthesized from only starch has less mechanical strength and thermal stability. Now biodegradable bioplastics are synthesized from any starch-rich biomass sources such as orange peel, potato, sweet potato, banana peel, and maize starches. To improve the mechanical strength and thermal stability of the film, starch was reinforced with cellulosic fiber. The raw material used was waste products that do not contradict any food security, utilizing waste for the production of usable products also advantageous to reduce waste disposal on the environment. In this case, banana peel starch blending with banana pseudo-stem fiber was used to synthesis the plastic film by using solution cast technics.

1.3. Objectives of the study

1.3.1. General objective

The General objective of this thesis is to synthesize and characterize bio-plastic film from banana peel starch blending with banana pseudo-stem fiber.

1.3.2. Specific objective

The specific objectives for this thesis are to;

- Determine the proximate composition of the banana peel, extract and characterize starch, and determine the optimum parameters which give a maximum yield.
- Extract and characterize fiber from banana pseudo-stem.
- Optimize the tensile strength, elongation at the break and water absorption of the synthesized bioplastic film.
- Determine solubility, transparency, thickness, and density for the synthesized plastic film, and characterize the best film by FTIR , DSC and TGA.

1.4. Significance of the study

The results of this study were aimed to provide essential baseline information about the potential of banana peel starch and banana pseudostem cellulosic fiber for the synthesis of bioplastic film. Investigating the conversion capacity of this low-value biomass into the more valuable product, particularly bioplastic film, was aimed to provide the researchers with an insight towards;

- The possibilities of using agricultural residues, particularly banana peel and banana pseudostem fiber, as raw material for the synthesis of degradable bioplastics.
- Reducing the emission of greenhouse gases to the environment as a result of replacing petroleum by alternative renewable raw materials.
- Finding raw materials that have a high amount of starch and excellent fibers, best for the synthesis of degradable bioplastics but not contradict with any food security.
- Production of value-added products in innovative ways of waste resource management.
- To increase the awareness to use environmentally friendly and degradable bioplastics for food packaging and dry good packaging alternatives.

Generally, this study contributes to show the direction of finding an alternative raw material for the synthesis of degradable bioplastics which is technically feasible, environmentally acceptable and easily available raw material.

2. LITERATURE REVIEW

2.1. Introduction

Banana (*Musa Cavendish)* belongs to family Musaceae is the most known plant in tropical and subtropical countries. It approximately contains 75%water and 25% carbohydrate besides a trace amount of protein and fat. It also contains higher levels of calcium, phosphorous and carbohydrate. However, starch is considered to be the major constituent in a green unripe banana. The starch consists of a long chain of covalently bonded glucose molecules. Banana is the second most-produced fruit after citrus, covers nearly 16% of the world's total fruit yield. India covers 26% of the world's banana production (Joshi & Sarangi, 2014). In fact, banana peel covers approximately one-third of the overall weight of banana fruit. One bunch of banana consists of 13 banana fruits, the length of one banana fruit is ±40 cm, and the thickness of banana fruit is 1± 0.5 cm (Hadisoewingyo *et al.*, 2017).

Plastic materials (plastics) considered as the key component of synthetic polymers, which are characterized by higher molecular mass. Due to easy processing and choice for making cost-effective products that elevate the living standard and the quality and comfort of life, polymeric materials successfully penetrated the global market (Joshi & Sarangi, 2014). Agro-industrial waste can be the production source of biopolymers such as bioplastics (Vega-Castro *et al.*, 2016). Plants, wood, corn, potatoes, banana, and wheat are all raw materials that are renewable, biodegradable and easily available souse of lignin, cellulose, and starch (Gadhave *et al.*, 2018). Biopolymers are polymers which able to bio-degrade with the action of micro-organisms, heat, and moisture (Vega-Castro *et al.*, 2016).

The different types of plant sources are starch-based sources like wheat, banana peel, corn starch, rice, sweet potato, barley, sorghum and cellulose derivatives which account for almost 80% of the bioplastics market (Puppala *et al.*, 2012).

Due to inherent chemical complexity, fruit waste can be utilized for the production of high-value materials, such as organic acids, biodegradable plastics, and enzymes (Uçkun *et al.*,2014).

Bioplastics are a type of biopolymer that can be produced from hydrolyzed polysaccharide materials and could eventually replace polypropylene and polyethylene, being biodegradable, biocompatible and produced from renewable carbon sources (Naranjo *et al.*, 2014).

The process of recycling is also much easier with biodegradable plastic. Furthermore, they cause no harm to the environment, as there is no toxins or chemicals residue produced as a byproduct (Ghuttora, 2016). Plastic materials are currently considered very important materials due to their properties and performance over other materials such as metal and wood (Arikan & Ozsoy, 2015). Biodegradable bioplastics are fully degraded by a microorganism (Arikan & Ozsoy, 2015).

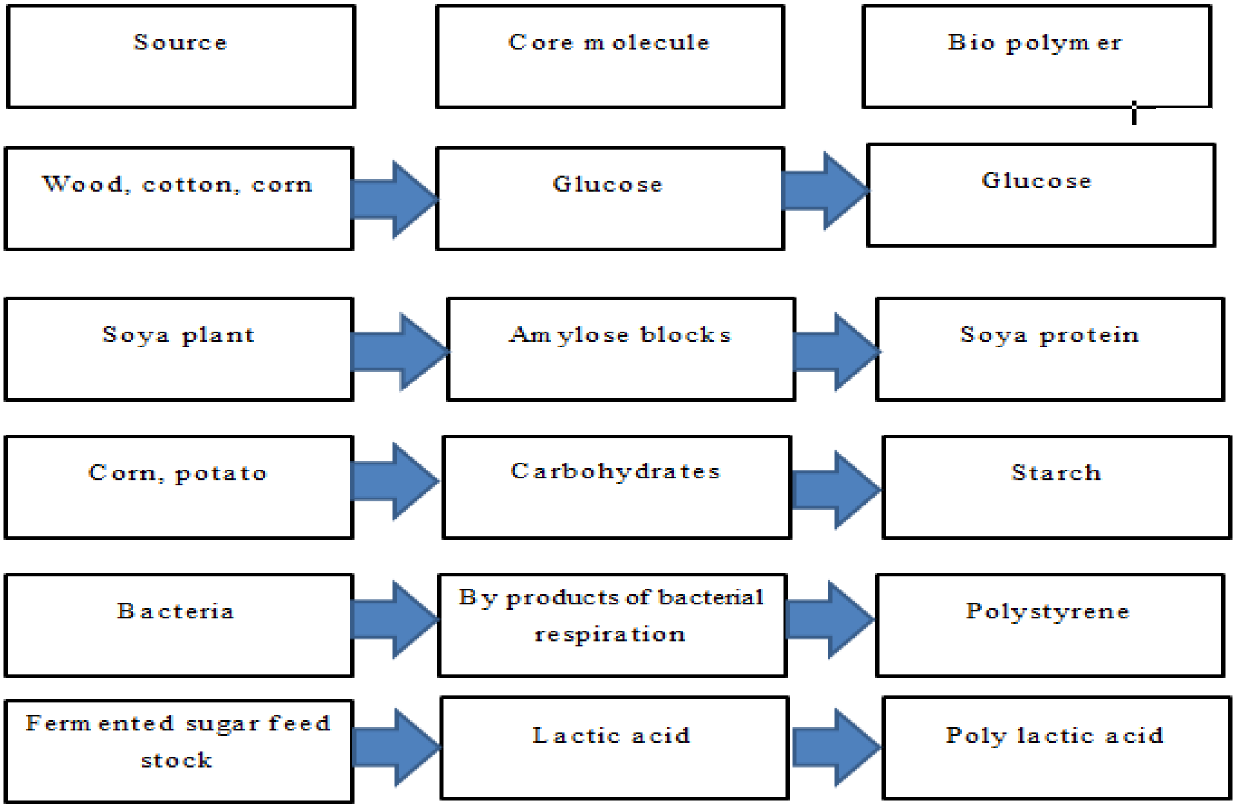


Figure: 2. 1.A different source of bioplastics (Arikan & Ozsoy, 2015)

2.2. Plastic additives used in plastic production

Plastic additives are a combination of an extremely diverse type of material. Some are complex organic molecules (antioxidants and light stabilizers) designed to achieve dramatic results at very low loadings. An example would incorporate the large surfactant type materials based on fatty acid chemistry which could impart lubricant, antistatic, mold release, and/or slip properties to a plastic matrix, based upon the materials involved, loading level, processing conditions, and application (Kattas *et al.*, 2018). Antioxidants are important in a variety of resins to protect oxidative degradation. Degradation is initiated by the action of highly reactive free radicals caused by heat, radiation, mechanical shear, or metallic impurities. The initiation of free radicals may occur during polymerization, processing, or fabrication (Kattas *et al.*, 2018). Plasticizer: are dispersants or additives that increase the plasticity or fluidity of a material.

The dominant applications are for plastics, especially polyvinyl chloride (PVC), glycerol, and sorbitol. The properties of bioplastics film are improved when blended with plasticizers including concrete, clays, fibers and related products. Plasticizers are used to get improved compound processing characteristics, while also providing flexibility in the end-use product. Glycerol (also called glycerin) is a simple polyol (sugar alcohol) compound. Basically, glycerol is a colorless, odorless, viscous liquid that is sweet-tasting and non-toxic substances. Physical properties of glycerol include chemical formula $C_3H_8O_3$, molecular mass (92.10g/mol), melting point (18^oC), boiling point (290^oc) and density (1.26 g/cm^3) (Mohsen *et al.*, 2015). For this research glycerol was chosen as a plasticizer due to its availability, potential and has no health impact. Most of the time the concentration of plasticizer/glycerol used is (10-60)% dry starch basis (Wittaya, 2012).

2.3. Biomass starches

The glucose produced by photosynthesis forms bonds to grow to a macromolecule which is starch (Schon and Schwartz, 2018).

Plant starch includes bast (or stem or soft sclerenchyma) starch, leaf or hard starch, seed, fruit, wood, cereal straw, and other grass starch (John & Thomas, 2008). Starch is important as starting material for a wide range of green materials. 75% of most organic material on earth is found in the form of polysaccharides. Starch is obtained in seeds and in tubers or roots of the plant's parts. Most of the starch produced worldwide is derived from corn (Gadhave et al., 2018). Linear amylose linked by α-l, 4-bonds and branched amylopectin linked by α-l, 6-bonds (Ebnesajjad, 2017).

At a given molecular weight, amylose swells to a much larger volume in solution than amylopectin, but the more amorphous amylopectin absorbs the more water than amylose at elevated temperatures because it is branched and not stable than that of amylose. Linear amylose polymer is capable to align their chains faster than branched amylopectin polymers. The branched amylopectin can have an infinite variety of structures, depending on the frequency of branching and the length of the branched chains. Different physical properties are associated with these various structures. These molecules can be cross-linked by themselves, or with other multi-functional reagents. As the cross-linking increases, the cross-linked polymer becomes less water-soluble (Ebnesajjad, 2017).

$$nC_6H_{12}O_6 \longrightarrow (C_6H_{10}O_5)_n + (n-1)\,H_2O$$

For amylopectin ‘n’ in the above equation can vary between 2000 and 200 000, which means that the amylopectin chains are formed by between 2 000 and 200 000 glucose units, which form branching every 24 to 30 glucose units. For amylose ‘varies between 300 and 3000 and there is no branching in the chain (Schon and Schwartz, 2018) Amylose is a chiefly linear polymer composed of α(1-4)-linked glucose units and a molecular weight (Mw) of about 105–106g mol^{-1}. Amylopectin is a larger molecule (Mw = 107–109g mol^{-1}) and is a branched polymer with α(1-4)-linked glucose backbone and typically 5% α (1-6)-linked branches (Sagnelli *et al.*, 2017).

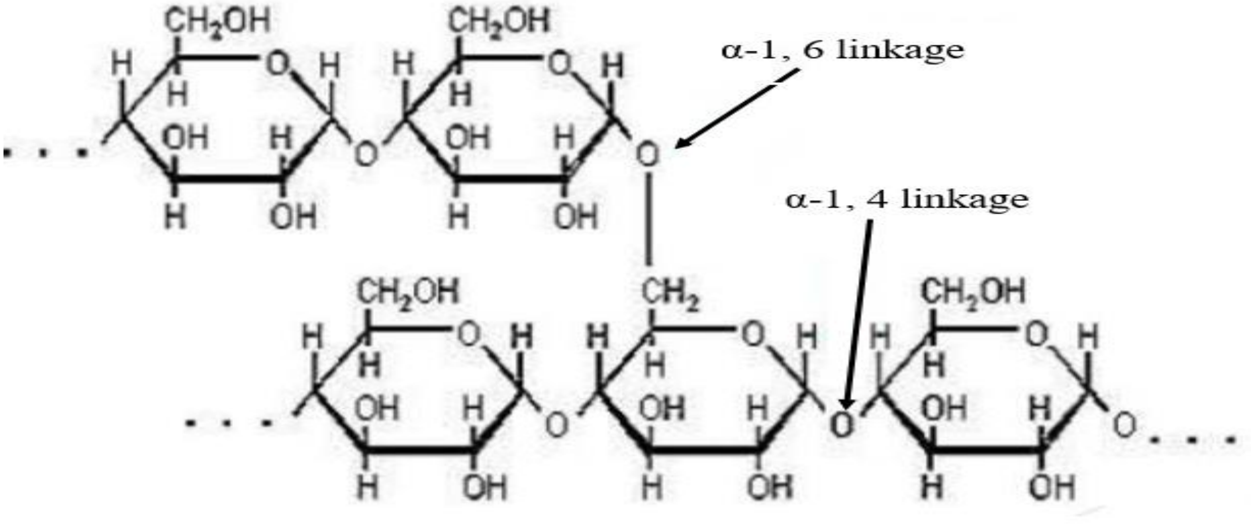


Figure: 2. 2.Molecular structure of starch (source; www1.isbu.ac.uk)

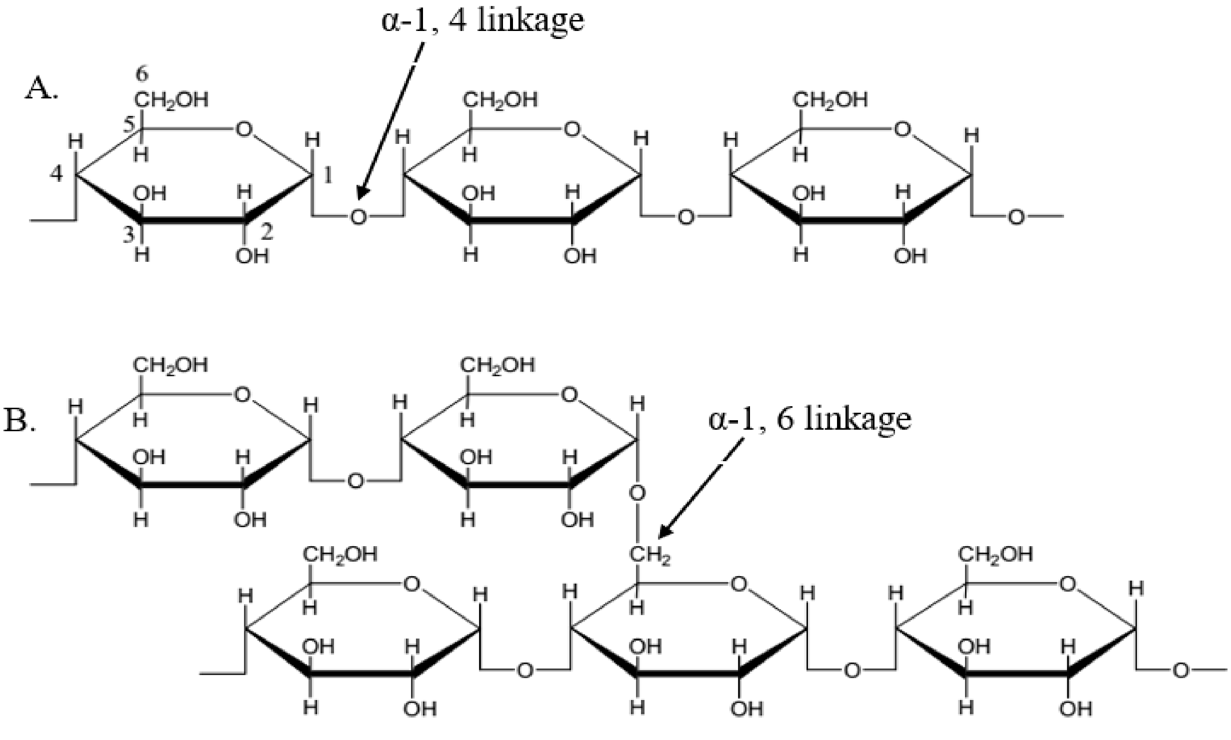


Figure:2.3.Amylose (A) and Amylopectin (B) structure(source;www1.isbu.ac.uk)

Physical properties of starch include chemical formula $(C_6H_{10}O_5)_n$, variable molecular mass, decompose at the melting point, and Density (1.5 g/cm^3) (Mohsen *et al.*, 2015).

2.4. Banana production capacity

2.4. 1. Banana production capacity in the world

Banana (Musa spp.) is one of the most popular types of fruits grown throughout the tropics and subtropics worldwide. It is consumed both as an energy-yielding food and as a dessert. Banana is a very important fruit commodity in many developing countries and is the main fruit in international trade which ranks first in terms of the volume exported and second after citrus fruit in terms of monetary value (Alemu, 2014).

Banana, the largest herbaceous plant in the world, is one of the ten most important fruits. It mainly produced in tropical areas such as Africa (13%), South and Central America (28%) and South-Eastern Asia (47%) (Pyar & Peh, 2018). So, it is possible to obtain banana peel adequately and application depends on its chemical compositions. In addition, peels and seeds can present higher nutrient content. They have a wide range of vitamins and minerals present in both pulps and peels (Pyar & Peh, 2018).

Table: 2. 1.Top banana producing countries in the world

Rank	Country	production in tons
1	India	27,575,000
2	China (mainland)	12,075,238
3	Philippines	8,645,749
4	Brazil	6,892,622
5	Ecuador	5,995,527

Source: (FAO, 2017)

2.4.2. Banana production capacity in Ethiopia

Banana is the most widely produced fruit crop. Production is estimated closely at 500,000 metric tons.

The largest share of production comes from the Arba Minch district and the Gamo Gofa zone in the SNNP region. Bananas occupy nearly 60% of the harvested area of fruit crops in the country. According to industry contacts, the type of banana cultivar farmers widely produce is dessert banana. Cooking banana varieties are less known among Ethiopian banana producers (Alemu, 2016). The major varieties of dessert banana cultivated include Giant Cavendish, Dwarf Cavendish, and Poyo. Giant and Poyo varieties are reported to bear the good size of fruit having better quality. The dwarf banana cultivar has a short plant height allowing farmers to manage the plantation easily (Bickford, 2018). Ethiopia invests 41 million euro to import plastic raw materials, 100% of the raw materials are imported from the middle east, Europe, and Asia in 2018 (Euromap). We have enough resources so; we have to use and utilize our resources and save the money invested in the case of importing raw materials.

2.5. Review on bioplastic production from different biomass sources

Previously different source was used to synthesis biodegradable plastics. A different source of starch with different fillers was also done. The technique to synthesis bioplastics at lab scale is solution cast method; which is whereby the film-forming solution is spread on a level surface and then dried to produce films (Altemimi, 2018).

2.5.1. Biodegradable plastic production from corn starch

Bioplastics were produced using a mixture of glycerol, cornstarch, vinegar, and food color (Keziah *et al*, 2018). Corn starch-based antimicrobial packaging film reinforces with plumbago zeylanica (Amira) root crude extract was done. The methanol extract of plumbago zeylanica root was used as antimicrobial agents (Muche,2018). Bioplastics were produced from corn starch by incorporating the cellulose obtained from sugarcane bagasse. The maximum and minimum value of tensile strength (26.81 and 11.55MPa), water absorption (39.02% and 20.45%) and elongation at break (25.99% and 4.32%) was obtained respectively.

This was done by considering two factors dry oven temperature (30-50^{0}C) and fiber amount (5-15 w/w) (Mosisa, 2017). Starch films were prepared from starch extracted from cassava, corn, potato, and yam with and without plasticizers to evaluate the effects of the type and quantity of plasticizer on the mechanical properties of the starch films (Adeyefa, 2015).

2.5.2. Biodegradable plastic production from potato starch

For potatoes starch, which is one of the best polysaccharides, highlighting the importance of the amylose/amylopectin ratio during production and leading to improved strength and lowered strain (Priedniece *et al.*, 2017). Thermoplastic Starch film was prepared from sweet potato blend with polyvinylalcohol (PVOH), at varying compositions by gelatinizing and plasticizing it with water and glycerol, the melting and glass transition temperatures of the TPS increased from 146 ^{o}C to 167 ^{o}C and 50.8 ^{o}C to 71.8 ^{o}C respectively, with the addition of PVOH (Offiong & Ayodele, 2016). The potato-based bioplastic sheet was obtained the highest tensile strength percentage (4.87 Mpa) followed by a cassava-based bioplastic sheet (4.5 Mpa) and a corn-based bioplastic sheet (3.59 Mpa) (Al *et al.*, 2016).

2.5.3. Biodegradable plastic production from Banana peel

Tensile strength of approximately 0.443 Mpa (0.22mm thick), 1.098 Mpa (0.15mm thick) and 1.596 Mpa (0.37mm thick) were observed for banana peels treated with $Na_2S_2O_5$, untreated banana peel + agar and banana peel treated with $Na_2S_2O_5$ + sorbitol composites (Mukhopadhyay *et al.*, 2017). Antimicrobial packaging film also produced from banana peel starch via ginger oil (Zerihun *et al.*, 2016).

2.5.4. Bioplastics production from cottonseed

Cotton-seed protein bioplastics from cottonseed flour were successfully prepared by hot-press molding in the presence of urea, aldehydes, and glycerol *(*Yue *et al.*, 2012).

A novel class of environmentally friendly protein-based bioplastics cottonseed protein plastic sheets (CP-sheets) was prepared by compression molding in the presence of cottonseed flour as the main raw material, aldehydes as crosslink agent and glycerol as plasticizer (Yue *et al.*, 2014).

2.5.5. Bioplastics production from Cassava Starch

The aim of the study was done to analyze the effect of glycerol on microbial degradation, that complete degradation could be achieved on the 9th day (Wahyuningtiyas & Suryanto, 2017). Bioplastics were prepared from cassava peel starch plasticized using sorbitol with a variation of 20; 25; 30% (wt/v of sorbitol to starch) reinforced with microcrystalline cellulose (MCC) with a range of 0 to 6% (wt/wt of MCC to starch). The results showed improvement in tensile strength with higher MCC content up to 9.12 Mpa compared to non-reinforced bioplastics (Wahyuningtiyas& Suryanto, 2018). Bioplastic component of cassava starch, glycerol as plasticizer and nanoclay as reinforcement were also investigated, The results show that the addition of nanoclay into bioplastic results increasing the tensile strength of bioplastic also increases from 5.2 MPa to 6.3 Mpa (Wahyuningtiyas & Suryanto, 2018).

Production of Biodegradable Plastic Packaging Film from Cassava Starch were also investigated, obtained result was a biodegradability of 41.27% compared to 10.33% and 85.99% for polythene and paper respectively, the film also has a tensile strength of 24.87N/mm^2 compared to 10.86N/mm2 and 8.29N/mm2 for polythene and paper (Ezeoha & Ezenwanne, 2013). Mechanical properties of bioplastics cassava starch film with Zinc Oxide nanofillers as reinforcement was done, the result showed that the Tensile strength (TS) was improved significantly with the addition of zinc oxide but the elongation at break (EB%) of the composites was decreased. The maximum tensile strength obtained was 22.30 N / mm on the addition of zinc oxide by 0.6% and plasticizer by 25% (Harunsyah *et al.*, 2017).

2.5.6. Bioplastics production from chitosan

Bioplastics production from starch and chitosan blends shows that the glycerin has an important role at the flexibility of the film, the acetic acid influences on the consistency, elasticity and shape, and the chitosan influences on the stiffness and thickness (Catálise *et al*., 2017).

Bioplastic from chitosan and yellow pumpkin starch with castor oil as a plasticizer, the characterization of the optimum tensile strength test was obtained on the 40/60 composition of 6.787 ± 0.274 Mpa and the fastest biodegradation test process within 5-10 days occurred in the 50/50 composition. The more chitosan content the higher the value of the tensile strength test obtained, while the fastest biodegradation rate occurred in the composition of yellow pumpkin starch and chitosan balanced 50:50 (Sarinigsh *et al.,* 2018).To finalize, bioplastics are produced on a large scale by molding, extrusion, and blowing. In a lab-scale, the most common and effective technique to produce bioplastic is a solution casting method.

Table: 2. 2.Tensile strength obtained from different biomass sources

S.no	Raw material	Tensile strength	Reference
1	Corn starch	26.81Mpa	(Mosisa, 2017)
		3.59 N/mm^2	(Al *et al.*, 2016)
2	Potato starch	4.87 N/mm^2	(Al *et al.,* 2016)
3	Banana peel starch	1.596 Mpa (0.37mm thick)	(Mukhopadhyay *et al.*, 2017)
4	Cassava starch	4.5 N/mm^2	(Al *et al.*, 2016)
		24.87N/mm^2	(Ezeoha et al., 2013)
		22.30 N / mm^2	(Ezeoha *et al.*,2017)
5	Chitosan	6.787 ± 0.274 Mpa	(Sarinigsh *et al* ., 2018)

2.6. Banana peel starch extraction and specifications criteria

2.6.1. Review on chemical and proximate composition of banana peel

A banana peel, called banana skin in British English, is the outer covering of the banana fruit. Banana peels are used as food for animals, in water purification, for the manufacturing of several biochemical and bioplastics products as well as for jokes and comical situations. Bio-plastic biomaterials utilizing banana peel waste biomass in the pharmaceuticals, medical, biomedical and bio-engineering aspects (Saad, 2016). Crude lipid, ash, carbohydrates, crude fiber, moisture, and crude protein content is determined by proximate analysis (Pyar & Peh, 2018). One of the most common waste forms of starch is the banana peels (Fatimah *et al.*, 2017). Different researchers determine different values on the chemical composition of banana peel. This variation might be due to the difference of extraction method used, variety difference, handling method, age on the maturation of the peel and other reasons.

Table: 2. 3. The chemical and proximate properties of banana peel in previous work

Parameter	Result (%)[a]	Result (%)[b]	Result (%)[c]
Ash (%)	8.8	9.83	8.5
Moisture (%)	14.5	13.5	6.7
Crude protein (%)	5.3	5.53	0.9
Crude lipid (%)	1.6	24.00	1.7
Crude fiber (%)	29.2	14.83	31
Organic matter (%)	_	_	_
Carbohydrate (%)	40.6	32.4	59

Source: [a](Pyar & Peh, 2018)[b](Abubakar *et al.*, 2016)[c](Anhwange & Ugye, 2014)

2.6.2. Extraction of starch from banana peel

The preparation method of banana peel starch used a modification of the preparation method of durian seeds starch (Joshi & Sarangi, 2014). The banana peel was weighed and cleaned, cut into small pieces using a knife and then was homogenized with distilled water containing sodium bisulfate solution (0.25g/L) at a proportion of banana peel: distilled water (1:2 w/v) (Joshi & Sarangi, 2014). The resulting homogenate was heated at 20-40 ^{0}C for 15- 30 minutes filtered, squeezed using a cotton filter cloth. The filtrate of the resulting suspension was kept for 24 h, yielding complete starch sediment. The starch was collected by decantation and it was subsequently dried in an oven (40-60ºC) for 24-40 h. The drying process took place until the moisture content of starch is within the specified range. The dried starch was grounded and screened through a 24-0.52µ m sieve.

Table: 2.4.Characteristics of banana peel starch

Types of test	Specification
1. Qualitative test	Bluish violet color
2. Physical test	
Form	Powder
Color	White
Odor	Odorless
Test	Tasteless
3. PH	4-7
4. Moisture content	<20%
5. Loss on drying	<15%
6. Ash content	<1%
7. Amylose content	17-25%

Source: (Hadisoewignyo and Tjandrawinata, 2017)

Table: 2. 5. Transparency of banana peels paste and banana peels starch powder

Products	Transparency (% transmittance)
Banana peels paste	0.6±0.057
Banana peels starch powder Bps	77.23±1.26.

Source:(Zerihun *et al.*, 2016)

2.6.3. Factors that affect the yield of starch

The conditions that increase the yield of starch production from banana peel wastes are the extraction parameters, such as antioxidant concentration, extraction time, and temperature affect the starch production yield and purity. The wet extraction method is used for starch extraction from banana peel (*Musa species*) peels. The starch yield varied from 3.25 to 13% starch obtained from the original 154g (Lambis M. & Pasqualino, 2018).

The ripping stage also affects the yield of starch. The main component of unripe bananas is starch. Green bananas contain up to80% starch measured in dry weight. During ripening, the starch is converted into sugars and ends up being less than 1% when the banana is fully ripe. The most common types of sugar in ripe bananas are sucrose, fructose, and glucose (Lambis M. & Pasqualino, 2017). Another study also shows as the starch content of peel for matured banana drops from approximate 75% to 1%, while the content of soluble sugar increases from approximate 1% to 20% (Li *et al.*, 2018). The apparent amylose and amylopectin content of the peel starch was 25.7% and 74.3 respectively (Li *et al.*, 2018).

2.7. Natural fibers used for composite application

Natural fibers can be classified according to their origin and grouped into the following classification (Kalia, *et al*, 2009).

Leaf: abaca, cantala, curaua, date palm, henequen, pineapple, sisal and banana.

Seed: cotton

Bast: flax, hemp, jute and ramie

Fruit: coir, kapok, oil palm

Grass: alfa, bagasse, bamboo

Stalk: straw (cereal)

The bast and leaf (the hard fibers) types are the most commonly used in composite applications. Natural plant fibers are constitutes of cellulose fibers, consisting of helically wound cellulose microfibrils, bound together by an amorphous lignin matrix.

2.7.1. Banana pseudostem fiber

Different varieties of bananas are available worldwide, only the cultivated and the edible banana are of economic importance. The edible clones are Musa sapientum (dessert banana) and Musa paradisiaca (Culinarytype). It belongs to the genus Musa of the family Musaceae, which has two genera Musa and Ensete. There are different varieties of banana differing in size, color, and taste (Kumar et al., 2013).

All types of banana trees are bounded with fiber (Vigneswaran et al., 2015). Banana, pineapple, palm, and coconut are the most usable fibers for reinforcement (Gopinathan *et al.*, 2017). Manual fiber extraction is a cumbersome process. In this process, Pseudostem is initially cut into pieces of about 60cm length and 7.5cm width. Then the Pseudostem is scraped and the fiber is separated by using a scraper or a flat blunt blade. In the manual process, skilled labor can produce the only 500600g of dry fiber in 8 hours' time (Mohiuddin *et al.*, 2014). The physical and chemical properties of banana fiber are considered below: The chemical composition of banana fiber is cellulose (50-60%), hemicelluloses (25-30%), pectin (3-5%), lignin (12-18%), water-soluble materials (2-3%), fat and wax (3-5%) and ash (1-1.5%) (Mohiuddin *et al.*, 2014). The yield of fiber is about 2 - 4 % of the dry basis of banana Pseudostem (Kumar *et al.*, 2013).

2.7.2. Chemical composition of banana pseudostem fiber compared with other fibers

The banana fiber has an important such as low density, appropriate stiffness, and mechanical properties and high disposability and renewability. Moreover, they are recyclable and biodegradable. Banana fiber, a lingo-cellulosic fiber, obtained from the pseudo-stem of banana plant (Musa sapientum), is a leaf fiber with relatively good mechanical properties (Kumar *et al.*, 2013). Recent studies have indicated banana fiber possesses a lot of advantages chemical and physical properties which can be used as a very good raw material for the textile and packaging industry (Vigneswaran *et al.*,2015). According to (Kassew, 2018) reported the proximate chemical analysis of Musa sapientum was cellulose (41.45%), hemicellulose (23.37%), lignin (10.46%) and extractive (12.72%). The proximate chemical analysis of banana pseudostem fiber and its comparison with other fibers species were presented in Table 2.6.

Table: 2. 6. Proximate / Chemical analysis of banana pseudostem fiber & it's a comparison with other fibers

Particulars	Banana fiber	Rice Straw	Sugarcane	Jute	Wheat Straw
Ash (%)	2-3	15-20	1.5-5.0	0.5-2	4.5-9
Pentosan (%)	10-12	23-28	27-32	18-21	26-32
Lignin (%)	11-14	12-16	19-24	21-26	16-21
Cellulose (%)	82-85	28-48	32-48	45-63	29-51

Source:(Kumar *et al.*, 2013)

2.7.3. Alkali treatment of fiber

Alkali treatment enhances surface roughness resulting in better mechanical interlocking and the amount of cellulose exposed on the fiber surface.

This increases the number of possible reaction sites and allows better fiber wetting. The possible reaction happened for the fiber and NaOH which is represented as shown below (Ebisike *et al.*, 2013).

Fiber-OH + NaOH----- Fiber-O-Na^+ + H_2O.

2.8. Bioplastic film synthesis technics

There are three common types of plastic synthesis techniques (Wittaya, 2012), these are (1) Casting, which is setting hot solutions on a surface upon cooling and drying a solution or a gel is a simple method for producing films with controlled thickness. (2) Extrusion or the thermo pressing process is a process used to create objects of a fixed cross-sectional profile. A material is pushed or drawn through a die of the desired cross-section. Used to create very complex cross-sections and work materials that are brittle, because the material only encounters compressive and shear stresses. It also gives finished parts an excellent surface finish. (3) Electro-hydrodynamic atomization referred to as electrostatic atomization or electro-spraying. This is a process in which a liquid is forced through a capillary and a potential difference of the order of kilovolts is applied between the capillary and the collection electrode. In this case, the first technique was selected due to its effectiveness and simple to conduct at the lab scale.

3. MATERIAL AND METHOD

3.1. Sample collection and preaparations

This part of the thesis intends to collect all the necessary raw materials, the collected samples were prepared and conditioned for further characterizations. Banana plant (Musa Cavendish) species for both banana pseudostem and banana peel were collected randomly from Jimma town. Sample preparation process includes; extraction of starch from banana peel by soaking with sodium bisulfate antioxidant, blending the soaked peel using hand blender, filtration and centrifugation finally drying and grinding to get starch in powder form. Characterize the starch and also extraction of fiber from the banana pseudo stem manually, treat the raw fiber by sodium hydroxide to remove the non cellulose part, and characterize the extracted fiber. Finally bioplastic film was synthesized using solution casting method, and then the physico chemical properties and thermal properties were determined. The best film was characterized by FTIR, TGA and DSC.

3.2. Chemicals

Major chemicals that were used in the experiment were; Sodium hydroxide solution (0.3M NaOH) for the treatment of raw fiber such that for removal of non-cellulosic components of raw fiber. Sodium bisulfate used as an anti-oxidant/preservative, (Glycerol) Propan-1, 2, 3-triol as a plasticizer, hydrogen peroxide for bleaching, (NaOH, HCl, H_3BO_4 (Boric acid), H_2SO_4 (96-98%), catalyst tablet VST (cod. A00000277) used for protein determination), for lipid determination; (oilseed or groundnut or maize flour) and solvent (petroleum ether, bp 40-60^{o}C or anhydrous diethyl ethyl ether or n-hexane) for fiber determination, etc.

3.3. Materials

The banana pseudostem was used as the raw material to extract the fiber. Distilled water used to dissolve the fiber and starch. Fiber is used to make ropes and mats and uses range from fencing to house and other constructions, etc.

For this thesis work, false banana stem cellulosic fiber was used as reinforcement. Starch was extracted from the banana peel. Starch and fiber were the main raw materials to undertake this particular study, the study areas were in Jimma town, Oromiya reign southwestern part of Ethiopia.

3.4. Equipment

The equipment used to conduct this study was: Hand blender, ball mill, Petri dish, cotton cloth, centrifuge, digital weighing balance (0.1-999 gram), thermometer, hydrometer, Viscometer, polyethylene bag (250gram), silica crucible, desiccators, drying oven, electrical muffle furnace, beaker(500cm^3), water bath, heating mantle, measuring cylinders(10-500ml), stopwatch, scissor, glove, goggles, spoon, stirring rod, magnetic stirrer, Kjeldahl digestion unit, Kjeldahl automatic distillation unit, digester (UDK 139), condenser, 124µm-1mm size mesh sieve, tensile machine, FTIR, TGA, DSC, UV photometer, For extraction of lipid the main equipment used were Soxhlet extraction apparatus, extraction thimbles (Whatman cellulose), filter paper (Whatman No. 42), desiccator, evaporating dish, water bath, analytical balance and etc.

3.5. Processing methods

3.5.1. Methods for proximate analysis of banana peel

Banana (Musa Cavendish) fruit peels were washed and allowed to dry at room temperature. Then fresh peels were dried at 50°C, then ground to obtain a fine powder (Pyar & Peh, 2018).

Proximate analyses: The proximate analysis was carried out using the standard methods of analysis of the association of official analytical chemists (Abubakar *et al.*, 2016). The parameters determined were: moisture content, crude protein, crude fat, crude fiber, total ash and nitrogen-free extract (carbohydrate). Triplicate was done for each parameter.

Moisture content determination: Two grams of fresh samples were placed in a pre-dried and weighed crucible. Oven combustion was used to dry the samples at a temperature of 105°C until a constant weight was obtained.

After removing the samples from the oven, they were allowed to cool for 60 min in a desiccator and then re-weighed. The moisture content was calculated as a loss in the weight of the original sample and expressed as a percentage. The moisture content was determined according to the equation below:

$$\text{Moisture content}(\%) = \frac{w_1 - (w_2 - w_o)}{w_1} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3.1)$$

Where: w_o = container constant weight; w_1 = fresh sample weight and w_2 = container and dry sample weight.

Crude protein determination: Kjeldahl analysis was used to determine the crude protein content by using a Kjeltec analyzer which measures the total nitrogen content in the sample. This is then converted to crude protein by using the factor 6.25 based on the assumption that the average of protein contains about 16% nitrogen. The principle of this analytical technique is similar to the conventional micro-Kjeldahl method and can be divided into three stages: i) digestion, ii) distillation and iii) titration.1.0 g of sample were taken for each trial. The protein content (%) of the samples was calculated using the following equation:

$$\text{Nitrogen in sample } (\%) = \frac{w_2 * \text{normality of acid} * 14 / w_1}{w_1} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3.2)$$

Where w_1 = Sample weight measured by milligram and w_2= volume of HCl. A conversion factor of 6.25 will be used to convert total nitrogen to protein for all the waste substrates as follow, Crude protein (%) = Nitrogen (%) x 6.25.

Crude lipid determination: Exhaustive Soxhlet extraction was used to determine the crude lipid by petroleum ether on a Soxhlet extraction system. A total amount of 1.0 g of the sample was put into pre-weighed thimbles and covered with a wad of cotton. A Soxhlet extraction cup was weighed and 100 mL of petroleum ether was added to each sample.

This process started in the controller unit and consisted of the following steps: boiling for 20 min, rinsing for 35 min, and recovery for 10 min and finally drying for 5.0 min. After the extraction, the solvent was evaporated.

The extraction cup was removed and placed in an oven for drying at 105°C. The extraction cup was removed from the oven after 60 min and allowed to cool in desiccators. The lipid was measured by re-weighing the extraction cup and the crude lipid content was determined by using the following formula:

$$\text{Crude lipid content}(\%) = \frac{(w_3 - w_1)}{w_2} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.3)$$

Where w_1= container weight; w_2= sample weight and w_3= lipid and container weight.

Total ash determination: The ash content is defined as the inorganic residue that remained after the organic matter was burnt away. The porcelain crucible was dried in the oven at 105°C for 60 min and then weighed after being cooled in the desiccator. As much as 1.0 g of the sample was weighed and placed overnight in a muffle furnace which was set at 550°C. After ashing, the sample was removed from the furnace. The crucible was cooled in a desiccator and then re-weighed. The total ash content was calculated using the following equation:

$$\text{Ash content}(\%) = \frac{(w_2 - w_o)}{w_1} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.4)$$

Where w_o= crucible weight; w_1 = sample weight and w_2 = crucible and ash sample weight.

Crude fiber determination: Crude fibers are the indigestible carbohydrate consists principally of cellulose, hemicelluloses, and lignin. It can be defined as the loss on ignition of the remaining dried residues after the consecutive digestion of feed sample with dilute acid and alkali under specific conditions. Triplicate fat-free dried samples of 1.0 g were weighed in a clean pre-weighed filter crucible. The crucible, together with the sample, was transferred to the hot-extraction unit and the sample was left to digest for 30 min with 150 ml of the solution containing 12.5% sulfuric acid and 0.25 mL of octanol. The condenser was switched off after 30 min and allowed to cool. The acid solution was filtered and washed with hot distilled water by suction. Then, the sample was digested for 30 min with 150 mL alkali solution (12.5% sodium hydroxide) and 0.25 mL of octanol to dissolve alkali-soluble matter from the sample.

The porcelain crucibles together with final residue were dried at 105°C in an oven for 60 min, cooled in a desiccator and then weighed. The ashing of the residue was ignited in a pre-heated muffle furnace at 550°C for 180 min. The percent of crude fiber content was calculated using the following equation:

$$\text{Crude fiber}*(\%) = \frac{w_2 - w_3}{w_1} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3.5)$$

Where w_1= sample weight; w_2= crucible with residue weight and w_3 = empty crucible with ash residue weight.

Determination of nitrogen-free extract: Nitrogen-free extract (carbohydrate) was determined according to the equation below:

$$\text{NFE} = (100 - (\text{WC\%} + \text{CP\%} + \text{CL\%} + \text{CF\%} + \text{ash\%})) \ldots\ldots\ldots\ldots\ldots\ldots \quad (3.6)$$

Where NFE = nitrogen-free extract; WC = water content; CP: crude protein; CL = crude lipid and CF = crude fiber.

3.5.2. Methods for the extraction of starch from banana peel

The preparation method of banana peel starch used in this study was a modification of the preparation method of durian seeds starch (Joshi & Sarangi, 2014). The banana peel was weighed and cleaned, cut into small pieces using a knife and then was homogenized with distilled water containing sodium bisulfate solution (0.25g/L) at a proportion of banana peel: distilled water (1:2 w/v) this was done by hand blender machine. 77g/144mL (peel: distilled water) was taken for each trial. The mixing was taken out with the aid of a mechanical stirrer for (15-30) minutes at (25-45^{o}C) temperature. After that slurry was filtered through a cotton cloth, the waste solution was cleaned.

The starch suspension was gone away overnight in refrigeration at 6°C yielding complete starch sediment and centrifuged for 15 min at 3000 RPM. The white sediment was then dried at 40°C in a convective oven for up to (40) hrs. The drying process took place until the moisture content of starch is within the specified range. Then starch in dried form was triturated with the supporter of a motor pestle to obtain the starch in powder form.

The dried starch was grounded and screened through a 0.125mm mesh sieve. Twelve experiments were done to observe the effect of main factors (by varying extraction time and temperature by fixing antioxidant concentration).

Figure: 3. 1.Banana peel

The yield of starch present in the peel of Musa Cavendish banana peel was calculated by measuring the mass of banana peel before extraction of starch and measuring the mass of extracted starch and then the two masses were compared. 12 experiments were done and the D-optimal response surface method was used to analyze the yield response.

$$\text{Yield (\%)} = \frac{\text{Mass of starch extracted }(\text{g})}{\text{Mass of peel used }(\text{g})} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.7)$$

3.5.3. Extraction of banana pseudostem fiber

The plant was cut down as soon as the fruits were harvested. The outer most covers were peeled and brown-green skin was thrown away retaining the cleaner or white portion which can process into knotted fibers.

The extraction process of the fiber was preceded by cutting the pseudostem at the bottom to avoid non-fibrous materials with a sharp blade. Then the fiber is extracted manually by scraping away the plant tissue using an iron ribbon till fibers were fully separated. Fiber is being extracted to determine the possible yield. To remove non-cellulosic component five grams of sample was taken and treated with a concentration of (0.3M) of 200 ml sodium hydroxide at 90^{o}C treatment temperature and at the duration of one hour. It was washed individually with distilled water and allowed to dry in the atmosphere for 48 hrs (Ebisike et al., 2013).

Figure: 3. 2. Banana pseudo-stem cross-sections

The yield of fiber present in the stem was calculated by measuring one segment of the stem before extraction of fiber and measuring of extracted fiber and then the two masses were compared.

$$\text{Yield (\%)} = \frac{\text{Mass of fiber extracted (g)}}{\text{Mass of stem bundle used (g)}} * 100 \ldots\ldots\ldots\ldots \quad (3.8)$$

3.5.4. Characterization of the extracted starch from banana peel

The banana peel starch was evaluated qualitatively for its properties including pH, moisture content, ash content, density, amylose content, loss on drying and amylose content (Hadisoewingyo, 2017).

The presence of starch was identified: The appearance of bluish violet color upon the addition of iodine was observed by adding iodine (Zaini, 2012).

PH measurement: Five grams of starch was mixed with 25 ml of CO_2-free distilled water for 1 min and kept for 15 min. The pH value was subsequently measured using a pH meter. In general, the pH of starch varies between 4 and 7 (Alcázar *et al.,* 2015).

Moisture content starch powder: Three grams of starch was weighed into a crucible and placed in an oven with a temperature of 105°C and dried for 24 hours to constant weight. Moisture content in the dried starch was determined by keeping the weighed quantity of sample in a thermostat-controlled oven at 105°C for 24 hours. The dry weight of each sample was taken on a weighing balance (A. O. A. C., 1975). The percentage of the moisture content and dry matter was then calculated by the formula as presented below:

$$\text{Moisture content}(\%) = \frac{(\text{Initial weight} - \text{Final weight})}{\text{Initial weight}} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3.9)$$

The moisture content of starch should be approximately less than 20% (Hadisoewingyo, 2017).

Determination of loss on drying: One gram of starch was weighed and put into a tarred pre-heated (105°C for 30 min) weighing bottle with its lid, and then placed into an oven (105°C) and dried until a constant weight is achieved.

$$\text{Loss on drying } (\%) = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3.10)$$

Determination of ash content: The sample was heated at the temperature of 550°C as such that the organic compound and its derivatives were destroyed and evaporated, yielding mineral elements and organic compound residue.

The ash content should be less than 1%. The ash content was calculated using the following formula:

$$\text{Ash content } (\%) = \frac{(w_2 - w_o)}{w_1} * 100 \text{.................................. } (3.11)$$

Where w_o= crucible weight; w_1 = starch weight and w_2 = crucible and ash sample weight.

Determination of amylose content: One hundred milligrams of amylose was dispersed using 1 mL absolute ethanol, and 9 mL 1M NaOH was added to the solution. The solution was positioned in a boiling water bath for 15 min with frequent shaking to obtain a clear solution; then the entire volume was adjusted to 100 mL using distilled water to obtain the stock solution.

To generate absorption spectra, 4 mL amylose stock solution or 2.6 mL amylopectin stock solution was added to 30 mL distilled water and the pH was adjusted to 3.6 using 0.5M HCl. Then, 0.5mL potassium iodide–iodine (I_2-KI) solution (2g/L I_2, 20g/L KI) was added, and the total volume was adjusted to 50mL using distilled water. This reaction solution was mixed and placed at room temperature for 30min, after which it was scanned using a spectrophotometer at the 600nm wavelengths. To generate the amylose standard curve, the amylose stock solution was diluted to obtain reaction solutions with final concentrations of 10, 20, 30, 40, 50, 60 and 70 µg/mL, respectively (Ma & Deng, 2017). Based on this amylose standard calibration curve, the maximum value of the amylose that is found in banana peel starch was determined using the following formula.

$$\text{Amylose content } (\%) = 3.06 * \text{Absorbance} * 20 \text{..................................} (3.12)$$

$$\text{Amylopectin content}(\%) = 100 - \%\ \text{Amylose content} \text{.........................} (3.13)$$

Amount of amylose and amylopectin present in starch affects the mechanical property of the starch polymer. High amylose starch presents higher stiffness and hardness than high amylopectin starch due to the comparatively less branching structures (Alobi *et al*, 2017).

3.5.5. Characterizations of Banana Pseudo Stem Fiber

Moisture Content: The moisture content of the fiber was measured with the principle of the mass difference between the mass of fiber before dried and after dried. The process was done by drying the wet raw fiber up to constant weight, the decrease in the fiber weight can be calculated in the following formula (Ebisike et al., 2013).

$$\text{Moisture content}(\%) = \frac{(\text{Initial weight of fiber} - \text{Final weight of fiber})}{\text{Initial weight of fiber}} * 100 \ldots\ldots (3.14)$$

Cellulose and Non-Cellulose Content: Cellulose in banana fiber has been used for further processing; it is enough to know cellulose content and non-cellulose content. The cellulose content of raw banana fiber was analyzed by soaking the raw fiber with 0.3 M of 7.5% sodium hydroxide solution for 1 hour at a temperature of 90^{o}C. After the process, soluble components were removed and the fiber was washed with distilled water to the neutral condition that happened and dried. The mass difference of raw fiber before and after soaking was taken to calculate the cellulose and non-cellulose content of the fiber by gravimetric analysis.

$$\text{Cellulose }(\%) = \frac{(\text{weight befor soaking} - \text{final weight after soaking})}{\text{weight befor soaking}} * 100 \ldots\ldots (3.15)$$

Water Absorption of banana pseudostem fiber; Water absorption test was done by soaking grams of dried fiber with water for 24 hours and finally, the soaked fiber was measured. After immersion, the excess water on the surface of the fiber was removed up using a filter cloth and the final weights of the specimens were then taken. The increase in the weight of the fiber was calculated using the following equation (Ebisike *et al.*, 2013).

$$\text{Fiber water absorption } (\%) = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} * 100 \ldots\ldots\ldots\ldots\ldots (3.16)$$

The density of extracted fiber: Density was measured using the Archimedes principle by using water as liquid. The mass of extracted fiber was measured and immersed in a measured volume of the water. This shows that the volume of the fiber was probably the volume difference of water after immersion and before immersion of the fiber. And density was calculated with a mass over volume formula.

$$\text{Density} = \text{Mass of fiber} \,/\, \text{Volume of fiber} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \ (3.17)$$

Thermogravimetric analysis (TGA): The thermal stability of fiber was tested by measuring the mass loss during a heating ramp rate by the Thermogravimetric analyzer (TGA instrument) using temperature programming from 25 to 600^oC at the heating rate of $20^oC/min$ to which the sample was heated. The thermal degradation onset temperature and the thermal degradation weight loss of the fiber were recorded and analyzed.

3.5.6. Synthesis of bioplastic from the extracted starch via fiber

The sample was prepared by the procedure adapted from the method described by *(*Salleh *et al,* 2009*)*. A film-forming dispersion was prepared by mixing the starch (5g) extracted from banana peel and distilled water (100ml). The dispersion was stirred manually on the shaker water bath set at 75°C for 20 minutes while stirring at the same rate until it becomes gelatinized. This step helps to provide homogeneous dispersion by disintegrating the starch granules. Then glycerol was added at different concentrations (20%, 35%, and 50%) w/v starch basis. Cellulosic fiber extracted from the banana pseudo-stem was then added at (5%, 12.5%, and 20%) w/w based on a starch weight basis. Each mixture was stirred for homogeneity and to make the gelatin very strong and then allowed to cool to 50 °C before being cast on a Petri dish. Film-forming solutions (40mL) still in hot were transferred into casting in glass Petri-dishes having 15 cm diameter. The glass Petri-dishes served as casting surfaces, enabling the film to have a smooth and flat surface. Then dishes were placed in an oven set at 65°C, 50^0C and 35^0C until the film was dry. Subsequently, the dishes were removed from the oven and the films were peeled off, and then stored at room temperature in polyethylene bags for further analysis.

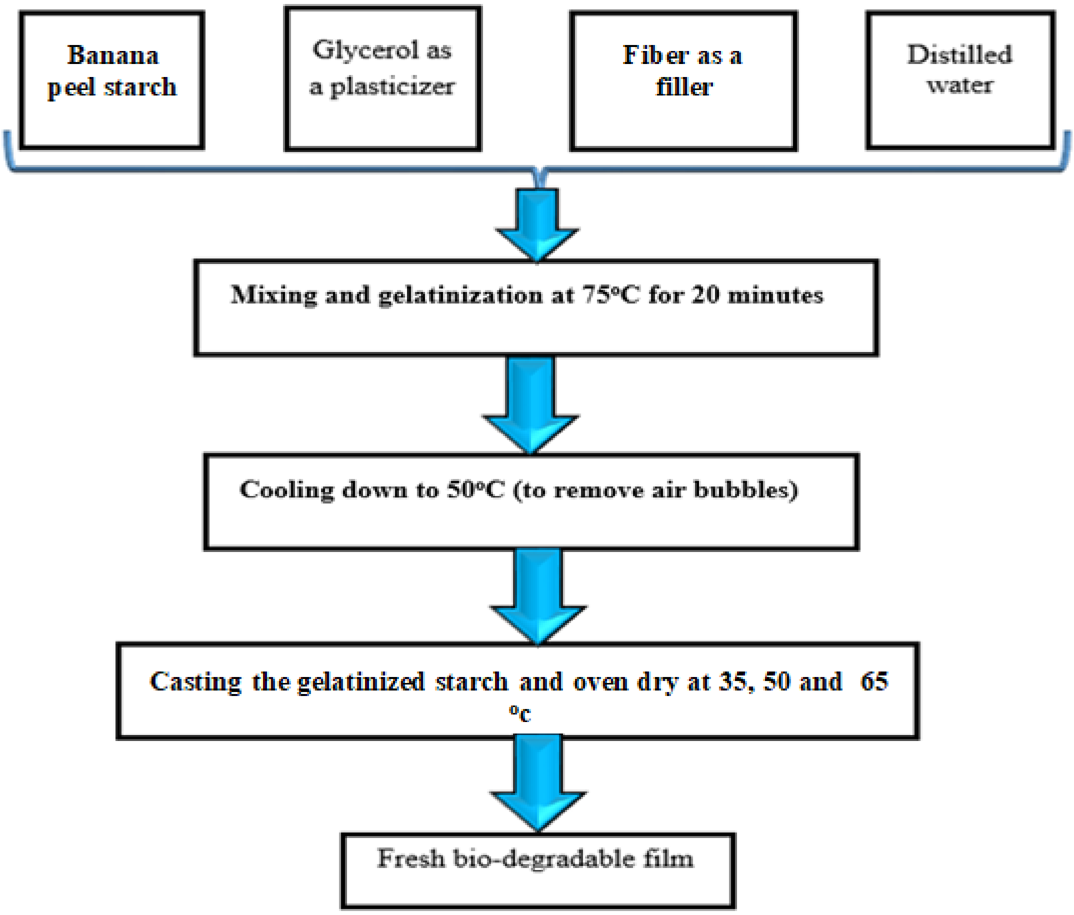


Figure: 3. 3.Film formation process

3.5.7. Characterization of Physical properties of plastic film

For every physical testing, before starting any test the sample was conditioned for 48hr in a specified temperature and humidity. For this purpose, we use the standard test method (ISO 2418:2005 and ISO2419:2005, Sampling and Conditioning).

Water solubility test: The film's solubility in water was determined according to the method reported by (Yehuala & Emire, 2013). Disks of the film (2cm x 2 cm) were cut, weighed (M1), and immersed in a beaker containing 50 ml of distilled water. After 24 h of immersion at 25^{o}C with a slow agitation, the pieces of the sample were taken out and dried to constant weight (M2) in an air circulated oven set at 105^{o}C for 24hr.

$$\text{Water solubilty} = \frac{M1 - M2}{\text{M1}} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(3.18)}$$

Where M1 = is the initial mass and

M2 = is the final mass of the sample

Water absorption test: Water uptake was investigated by cutting film with a size of approximately 2x2 cm and then weighed the mass. The film was put into a container filled with distilled water for 24 hours. After immersion in water, the film was removed from the water and weighed to measure the wet weight. Water uptake was calculated as follows

$$\text{Water absorption}(\%) = \frac{\text{Wet weight} - \text{Dry weight}}{\text{Wet weight}} * 100 \ldots\ldots\ldots\ldots \text{(3.19)}$$

Density test: The bulk density (D) was determined by dividing the mass (m) per unit volume (v) of the plastic sample.

$$D = \frac{m}{v} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(3.20)}$$

Transparency of produced bioplastics: The transparencies of the films are determined using a spectrophotometer (UV 7804C). The transmittance of films was determined at 600nm as described by (Yehuala & Emire, 2013). The film samples are cut into rectangles and make it as a solution form, then put into the internal side of the spectrophotometer cell.

$$\text{Transparency}(\%\text{T}) = \frac{\text{-logT600nm}}{X} \ldots\ldots\ldots\ldots\ldots\ldots \text{(3.21)}$$

Where T600 = is the transmittance at 600nm and X is the film thickness (mm).

Moisture: Prior to determining film properties, samples were conditioned at 25^{o}C and 53% relative humidity (RH) for 48 h. Film moisture content was determined through the weight loss through which the film went after a 24hrs oven drying at 90^{o}C. Preliminary experiments showed us that 24hrs was enough to dry up samples. The temperature was chosen to avoid the loss of plasticizer (Khazaei *et al.*,2014).

$$\text{Moisture content}(\%) = \frac{\text{Final weight} - \text{initial weight}}{\text{Final weight}} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.22)$$

The thickness of Films: The film thickness was measured using digital micrometers and measurements were made in at least, three random locations.

Tensile strength and elongation at break: Tensile strength is defined as the strength of a material in terms of force per unit area of cross-section while applying force in a linear direction. The test was carried out at the Amhara paper and printing factory at Bahir Dar. The test specimens were conditioned to conform to standard requirements of thin plastic sheeting in accordance with ASTM D882 equipped with a 1 KN load cell at a crosshead rate of 50 mm/min. From the developed plastic sheet, film strips of uniform width and thickness (10mmx50mm) were cut.

$$\text{Tensile strength at yield (MPa)} = \frac{\text{Force (N)}}{\text{Area(width*thickness)mm}^2} \ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.23)$$

$$\%\ \text{Elongation} = \frac{\text{Increase in length}\left(\text{mm}\right)}{\text{Original length}\left(\text{mm}\right)} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.24)$$

FTIR analysis of the plastic film: The functional groups (chemical bonds) of produced bioplastic were identified by Fourier transforms infrared spectroscopy (FTIR).Then the FTIR spectrum was allowed to pass through the prepared sample and the spectrum responses were recorded.

Finally, the peak plot of wavenumber (400-4000cm^{-1}) versus Transmittance (%) was plotted using origin software and identified the functional groups.

Thermogravimetric analysis (TGA): The thermal stability of the film was studied by using Thermogravimetric Analysis (TGA) and were performed on the Shimadzu TGA-50 equipment under nitrogen atmosphere, the flow rate of 50 mL/min, a heating rate of 20°C/min and temperature range of 20ºC to 450°C. Approximately 5mg was placed on the platinum pan. DSC also done from 20-200 °C to determine different operating tempreture of the film.

3.5.8. Experimental Design and statistical analysis

For experimental design and analysis of the data, the design expert 6.0.8 and 11 were used. By using the design expert of the response surface, central composite design (CCD) of quadratic model of the total experiment to be conducted was 20 for the synthesized film. Randomization of the experimental runs as well as appropriate analysis technique was ensured through proper utilization of software Design Expert 6.0.8 and 11 versions. Three independent variables and three coded levels (-1, 0 and +1) with three responses were used in this study. A (dry oven temprature, ^{o}C) ranged from (35 to 65), B (glycerol concentration, %) ranged from (20 to 50) and C (fiber concentration,%), while the dependent variables (response variables) were the tensile strength,water absorption and elongation of the film. The response variable was fitted to the following second-order polynomial model which is generally able to describe relationship between the responses and independent variables. A second order polynomial model was selected by taking into account the benefit of order and hierarchy. The order of the polynomial model is kept as low as possible. If this is not satisfactory, then second order polynomial is tried. A model is said to be hierarchical if it contains the terms x, x^2, x^3, etc. in a hierarchy. It is expected that all polynomial models should have this property because only hierarchical models are invariant under linear transformation. The general quadratic model / second order equation is shown below.

$$\mathrm{Yi} = \mathrm{b_o} + \sum_{\mathrm{i-1}}^{\mathrm{n}} \mathrm{b_i X_i} + \sum_{\mathrm{i-1}}^{\mathrm{n}} \mathrm{b_{ii} X_i^2} + \sum_{\mathrm{i-1}}^{\mathrm{n}} \sum_{\mathrm{j-i+1}}^{\mathrm{n-1}} \mathrm{b_{ij} X_i X_j} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(3.25)$$

Where Yi is the predicted response, b_0 the offset term, b_i the linear effect, b_{ii} the square effect, and b_{ij} is the interaction effect. It is a polynomial regression model in one variable and is called as second order model or quadratic model. For statistical calculations the variable x_i was coded X_i according the following equation:

$$X_i = \frac{\mathrm{xi - xo}}{\Delta \mathrm{x}}, \mathrm{I} = 1;\ 2;\ 3 \ldots \mathrm{K} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(3.26)$$

WhereX_i is coded (dimensionless) value of the variable x_i, x_0 is the value of x_i at the center point and Δx is the step change. The following second-order polynomial models were used analogically to describe the relationship between the three independent variables and the three response variables:

$$TS = a_0 + a_1A + a_2B + a_3C + a_{12}AB + a_{13}AC + a_{23}BC + a_{11}A^2 + a_{22}B^2 + a_{33}C^2 ......(3.27)$$

$$EA = b_0 + b_1A + b_2B + b_3C + b_{12}AB + b_{13}AC + b_{23}BC + b_{11}A^2 + b_{22}B^2 + b_{33}C^2 ......(3.28)$$

$$WA = c_0 + c_1A + c_2B + c_3C + c_{12}AB + c_{13}AC + c_{23}BC + c_{11}A^2 + c_{22}B^2 + c_{33}C^2 ......(3.29)$$

Where TS, EA and WA are the predicted response which are tensile strength,elongation and water absorption respectively; a_0, b_0 and c_0 are an intercepts; [a_1, a_2 , a_3] , [b_1, b_2 , b_3] and [c_1, c_2 , c_3] were the estimated linear effects; [a_{11}, a_{22} , a_{33}], [b_{11}, b_{22} , b_{33}] and [c_{11}, c_{22} , c_{33}] were quadratic effects; and [a_{12}, a_{13}, a_{23}], [b_{12}, b_{13}, b_{23}] and [c_{12}, c_{13} , c_{23}] were interaction effects of independent variables which were ‘A’ (dry oven temprature), ‘B’ (glycerol concentration) and ‘C’ (fiber concentration).

The following second-order polynomial model also used to describe the relationship between extraction time, extraction temperature and the response variable(yield of extracted starch from the peel):

$$Y_i = e_0 + e_1X_1 + e_2X_2 + e_{12}X_1X_2 + e_{11}X_1^2 + e_{22}X_2^2 ....(3.30)$$

where Y_i is the yield of starch; e_0 is an intercept; e_1 and e_2 are the estimated coefficients of time (X_1) and temperature(X_2), respectively; e_{11} and e_{22} are quadratic effects; and e_{12} is interaction effect of independent variables.

Table: 3. 1.Factors and ranges of the CCD design quadratic model

Factors/independent variables	Range and levels	
	Ranges	Levels
Fiber conc.	(5-20%)w/w	5, 12.5 and 20
Oven dry temperature	(35-65)^{0}C	35, 50 and 65
Glycerol conc.	(20-50%)w/v	20, 35 and 50

4. RESULT AND DISCUSSION

4.1. The result on proximate composition of raw banana peel

The proximate analysis was done using the standard methods of analysis of the association of official analytical chemists (Abubakar *et al.,* 2016). The parameters determined were: moisture content, crude protein, crude fat, crude fiber, total ash and nitrogen-free extract (carbohydrate). The obtained result compared with other researchers was summarized in Table 4.1.

Table: 4. 1. The result on proximate composition of the raw banana peel on a mass basis

Parameter	Result (%)[a]	Result (%)[b]	Result (%)[c]	Result (%)[d]
Ash (%)	8.5	8.8	9.83	8.5
Moisture (%)	10.5	14.5	13.5	6.7
Crude protein (%)	2.17	5.3	5.53	0.9
Crude lipid (%)	6.66	1.6	24.00	1.7
Crude fiber (%)	34.37	29.2	14.83	31
Organic matter (%)	81	_	_	-
Carbohydrate (%)	37.8	40.6	32.4	59

Source: [a](current study)[b](Pyarthe and Peh, 2018) [c](Abubakar *et al.*,2016) [d](Anhwange & Ugye, 2014)

The moisture content: The result shows small when compared to the value obtained (Abubakar *et al.,* 2016) and (Pyar & Peh, 2018). But relatively higher than the value obtained (Anhwange & Ugye, 2014). This variation may be due to handling problems and level of ripping. And high moisture content basically causes food items to the enhanced microbial spoilage and short shelf life, which can lead to its deterioration. As much as possible the moisture content should be small and the above result was somewhat good and acceptable.

Total ash content: The lower ash content may suggest low mineral content. On the other hand, the high values of the ash were indicative of high mineral content. The above result shows almost similar with (Anhwange & Ugye, 2014) and (Pyar & Peh, 2018) but lower than (Abubakar *et al.*, 2016).

The crude lipid content: It was low and may be good sources of carbohydrate and can also contribute significantly to the energy content of the feeds that can be prepared with the wastes. The obtained result shows higher compared (Pyar & Peh, 2018) and (Anhwange & Ugye, 2014) but smaller when compared with the value done (Abubakar *et al.,* 2016). This variation may be resulted in due to differing in varieties of banana.

The crude fiber: The values of crude fiber (34.37%) reasonably higher with the first two studies but similar (Anhwange & Ugye, 2014). The high fiber content in diets has been reported to result in increased removal of potential mutagens, steroids, and Xenobiotic by binding or absorbing to dietary fiber components and thereby aids digestion. Basically, fibers are an indigestible portion of carbohydrate (cellulose, lignin, and hemicellulose).

Protein: It is an essential component of the diet needed for the survival of animals and human beings; its basic function in nutrition is to supply an adequate amount of required amino acids. The Crude protein content of the samples was very low when compared to other studies but higher than the result obtained (Anhwange & Ugye, 2014).

Organic matter: It is the total composition of the above constitute excluding the moisture content and ash contents. It comparatively depended on the amount of nitrogen-free extract.

Carbohydrate content: The samples were very high in carbohydrate content and can be good sources of energy for the animals. The obtained result of carbohydrate (37.8) was almost higher than the value determined (Abubakar *et al.,* 2016) and lower when compared with the two other studies (Anhwange & Ugye, 2014) and(Pyarthe and Peh, 2018).

The result shows the peel has a higher amount of carbohydrates indicates the peel constitutes a large amount of starch. Starch is a complex carbohydrate. The ripping stage of the peel also affects its carbohydrate yield. The amount of carbohydrate was calculated by the subtraction of the summation of each composition from 100%.

4.2.Starch extraction and yield results

Extraction time and extraction temperatures were the two factors and starch yield was the response. And D-optimal RSM was taken to analyze the variance for 12 experiments. The obtained result with each trial is summarized in Table 4.2.

Table: 4. 2.Values of the yield variables associated with the two factors

Std.	Run	Factor 1 A: Extraction time (minutes)	Factor 2 B: Extraction temperature(^{0}C)	Yield (%)
1	5	15	45.00	10
2	4	15	25.00	4
3	11	30	35.00	11.5
4	7	22.5	35.00	11.8
5	8	30	45.00	10.82
6	2	30	25.00	8.5
7	9	22.5	25.00	6.15
8	1	15	35.00	9.5
9	12	22.5	45.00	10.5
10	10	30	25.00	9.2
11	6	15	25.00	3.8
12	3	15	45.00	10.2

As shown in Table 4.2 the maximum yield was obtained at 35^{o}C and 22.5 minutes which is 11.8% from the 7th run. As the temperature rises beyond the range, the starch becomes, jel-like and it might be gelatinized and almost at a difficult stage to re-crystal. So care should be given during operating the process. As extraction time increases the yield to some extent slightly increase but it is much significantly affected by extraction temperature. The temperature also affects the yield as shown in Table 4.3, the temperature very significantly affects the yield.'A' implies time and 'B' implies temperature. As temperature increases up to 35^{0}C the yield increase(maximum), then it decreases. But starch gelatinization which might start to occur at high temperatures > 40 ^{0}C may also increase the viscosity. Considering the efficiency of starch separation, the medium temperature should be used (Been *et al.*, 1987). Medium extraction temperature is mostly advisable because it reduces the viscosity of the starch suspension, and make easy for centrifugation (Been *et al.*, 1987).

Response: yield ANOVA for response surface quadratic model analysis of variance table [partial sum of squares].

Table: 4. 3. ANOVA results for starch extraction /yield from banana peel

Source	**Sum of Squares**	**Mean Squares**	**F -Value**	**Prob > F**	
Model	80.98	16.20	66.11	< 0.0001	significant
A	14.53	14.53	59.31	0.0003	
B	35.43	35.43	144.61	< 0.0001	
A^2	0.25	0.25	1.03	0.3491	
B^2	13.89	13.89	56.69	0.0003	
AB	7.74	7.74	31.61	0.0014	
Residual	1.47	0.24			
Lack of Fit	1.18	0.39	4.16	0.1362	not significant
Pure Error	0.28		0.095		
Cor Total	82.45				

The Model F-value of 66.11 implies the model is significant. There is only a 0.01% chance that a "Model F-value" this large could occur due to noise. Values of "Prob > F" less than 0.0500 indicate model terms are significant.

In this case, A, B, B^2, AB are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. If there are much insignificant model terms (not counting those required to support hierarchy), model reduction may improve your model. "Lack of Fit F-value" of 4.16 implies the Lack of Fit is not significant relative to the pure error. There is a 13.62% chance that a "Lack of Fit F-value" this large could occur due to noise. Non-significant lack of fit is good - we want the model to fit.

Table: 4. 4.Model adequacy measures for starch extraction yield response

Std. Dev.	0.49	R-Squared	0.9822
Mean	8.83	Adj R-Squared	0.9673
C.V.	5.60	Pred R-Squared	0.9134
PRESS	7.14	Adeq Precision	23.615

The "Pred R-Squared" of 0.9134 is in reasonable agreement with the "Adj R-Squared" of 0.9673."Adeq Precision" measures the signal to noise ratio. A ratio greater than 4 is desirable. Your ratio of 23.65 indicates an adequate signal. This model can be used to navigate the design space.

Effect of temperature on yield: Figure 4.1 below shows as the temperature increases up to 35^0C, the yield increase, then it decreases when time is kept constant 22.5 minutes. Temperature is a very significant factor that affects the amount of starch extracted because the starch molecules were sensitive to temperature. Starch yield depends on extraction time and temperature; longer time and extraction temperature with higher alkali concentration is not recommended to extract starch. At lower alkali concentration, the longer time the starch yield might slightly increase.

At extraction, temperature increases the starch yield increase up to a certain instant and finally, it decreases (Mistry, *et al*., 1992). In starch, the bond exists between amylose and amylopectin in the crystal and amorphous region. Temperature directs the swelling process along the amorphous regions and since the crystalline regions do not expand, stress increases at the interface between the crystalline and amorphous regions. Thus, at a certain point in the swelling process, the crystalline regions are rapidly and irreversibly broken and gelatinization is initiated (Usman *et al.*, 2017).

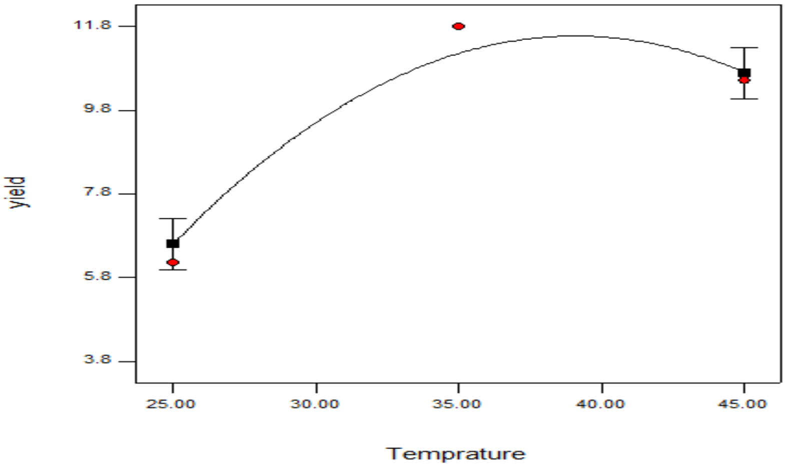


Figure: 4. 1. Effect of temperature on yield

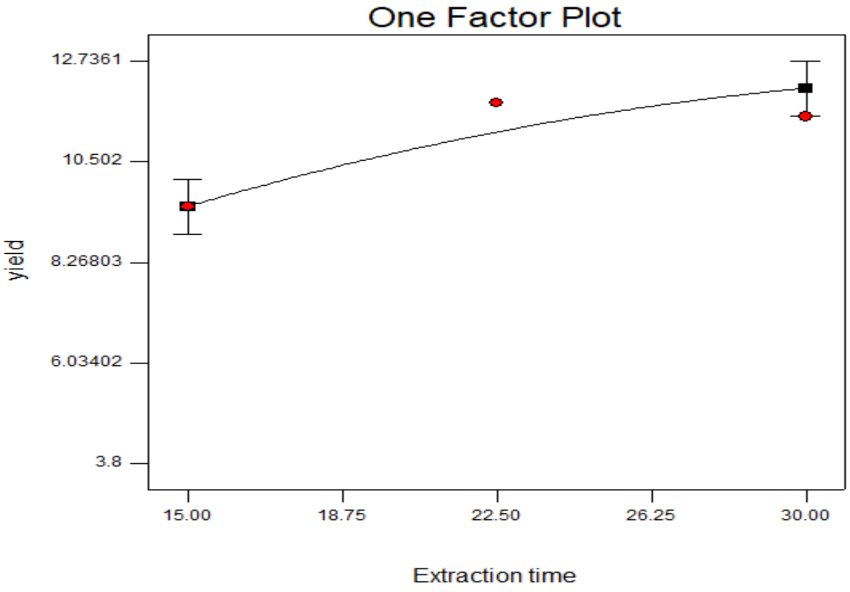


Figure: 4. 2.Effect of time on yieldEffect of time on yield:

Figure 4.2 shows as the extraction time increases, the yield slightly increases, finally it decrease very slightly when the extraction temperature is kept constant 35^{o}C. This is due to the amylose and amylopectin fragments and the capacity of the alkali used to soak the peel and extract the starch. At higher level of extraction extraction time starch decrease, because it is consumed initially, and aproches to end at the extraction time procceds.

4.2.1.Development of a regression model for starch yield

The model equation that correlates the response to the process variables in terms of coded value and actual is given below.

yield=+11.16+1.31* A+2.04 * B-0.34 * A^2-2.51* B^2-1.09* A * B…………(4.1)
where,A=extraction time, B= extraction temprature.

Final Equation in Terms of Actual Factors:
yield=-45.20157+0.95382*time +2.29084*temprature-6.02756E-003*$time^2$ -0.025141*$temprature^2$-0.014515*time*temprature …………………..(4.2)

4.2.2. Optimization of process variable and yield

The main objective of the optimization is either to minimize effort or to maximize benefit. The effort or benefit can be usually expressed as a function of certain design variables. Hence, optimization is the process of finding the conditions that give the maximum or the minimum value of a function. Under this, the process variable both extraction time and temperature should be in range with yielding maximum starch.

Table: 4. 5. Summary of constraint responses and goals of optimizations

Constraints Name	Goal	Lower Limit	Upper Limit	Lower Weight	Upper Weight	Importance
Extraction .time	Is in range	15	30	1	1	3
Extraction.Tmp	Is in range	25	45	1	1	3
Yield	Maximize	3.8	11.8	1	1	3

Using numerical optimization 10 solutions were found, the possible combination which contains a maximum yield was selected.

Table: 4. 6.Predicted optimum operating points

Extraction Time(minutes)	Extraction Temprature(^{0}C)	Yield (%)	Desirability
27.66	37.92	12.063	1.000

The predicted result by using numerical optimization was validated by doing an experiment (triplicate) at the predicted parameter and the yield obtained was 12.08%, which is near to the predicted yield.

Table: 4. 7. Model validation for yield

Number	Extraction Time(minutes)	Extraction Temprature(^{0}C)	Yield (%)
Pridicted	27.66	37.92	12.06
Experimental	28	38	12.08

The result 12.06% shows as the yield are reduced due to the ripping stage. As already mentioned in the literature part, the yield of starch affected by anti-oxidant concentration, extraction time, temperature, ripping stage of the peel and methods of extraction(wet or dry), The raw banana peel used was the skin of the banana which is thrown as waste. A green unripe banana might have yielded up to 80%, but it becomes reduced as approaches to the age of maturation, the reason is that because of the starch is converted into sugar like sucrose and fructose. Finally, the yield of starch may reach 1% (Lambis M.& Pasqualino, 2017). The optimum conditions obtained were 27.66 minutes of extraction time and 37.92^{0}C extraction temperature with yield 12.06%. The maximum starch recovery had been achieved at 40^{0}C and the minimum starch recovery had been achieved at 22^{0}C (Usman *et al.*, 2017).

This result almost agrees with the author (Lambis M. & Pasqualino, 2017) which is 13%. The result obtained was comparable to potato starch (7.5-16.5%) (Altemimi, 2018).

4.3. The result of the characteristics of the extracted starch

The resulting starch in this study is as shown in Table 4.8. The starch was obtained in powder form after drying and grinding.

PH determination: Banana peel starch powder was mixed with distilled water yielding a suspension. The pH of the mixture was found to be 6.31, which falls within the ranges of (4-7) (Alcázar-alay et al., 2015). In general, a starch suspension in water has a pH of (4 – 7) (Alcázar-alay *et al.*, 2015).

Table: 4. 8.The result on characteristics of banana peel starch

Types of test	[a]Results	[b]Specification
Qualitative test	Bluish violet color	Bluish violet color
Form	Powder	Powder
Color	Brownish white	White
Oder	Odorless	Odorless
Test	Tasteless	Tasteless
a. PH	6.31	4-7
b. Moisture content	7.2 %	<20%
c. Loss on drying	11.20 %	<15%
d. Ash content	0.8%	<1%
e. Amylose	19.278%	(17-25)%

Source: ([a]current study and [b]Alcázar-alay *et al.*, 2015).

Moisture content: The moisture content of starch powder may determine its quality and stability. Starch powder with high moisture content is vulnerable to bacterial growth, compared with that with low moisture content.

The moisture content of starch should be less than 20% (Hadisoewingyo, 2017). The moisture content of banana peel starch powder was found to be 7.2%. This result met the above specification. As shown in Table 4.8.

Loss on drying: Testing of loss on drying is performed to measure the loss of water upon heating. The result showed that the loss on drying of banana peel starch was 11.20%, meeting the required specifications (Table 4.8). In general, the loss on drying of starch is less than 15%.

Ash content: It was below 1% that is 0.8% which satisfies the above specification**.** The color of the resulting banana peel starch was brownish-white by visual inspection. It is also possible to change into pure white by using hydrogen peroxide as a bleaching agent.

Amylose determination: A Standard amylose calibration curve (appendix h) is a mathematical tool that provides a set of reference points that an unknown concentration of samples can be compared too. Once the curve is generated and all other values that pertain to techniques like linear regression (for example, the correlation coefficient (R^2)) were determined, the curve can also be used to find the concentration of the unknown sample.

It was calibrated using spectrophotometry that the absorbance of the sample at 620nm wavelength was obtained 0.314 and the unknown sample concentration (using the slope and the intercept from the standard calibration equation) was also determined which is within the range of the standard absorbance and it implies that the unknown solution certainly contains amylose which was about 19.3% and amylopectin 80.7% based on the calibration curve in the appendix(h) and equation (3.12 and 3.13) chapter 3. This result agrees with the researcher which determines banana peel starch consists of 20% amylose and 80% amylopectin (Hadisoewingyo, 2017).

Since the specification criteria of banana peel starch completely met the above criteria the extracted starch was efficient during plastic film production.

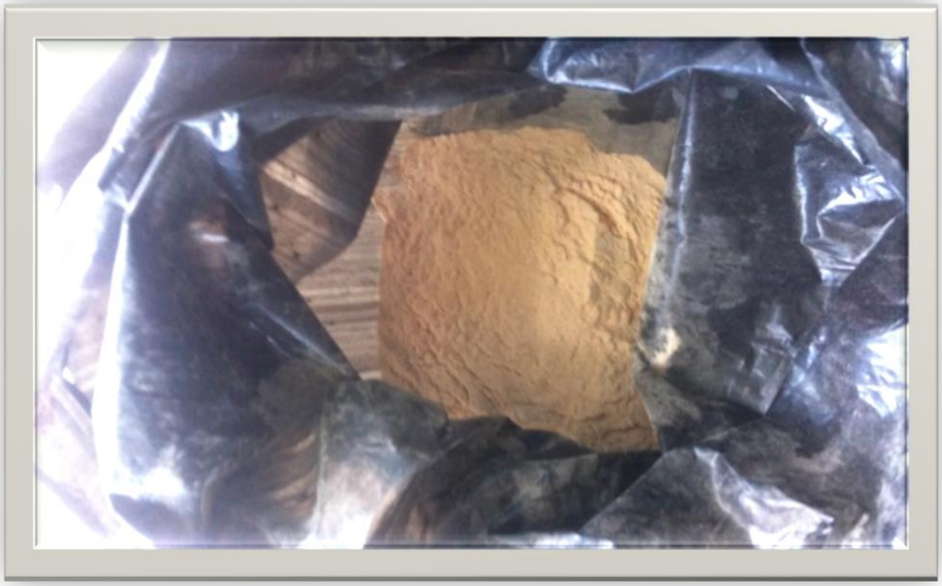


Figure: 4. 3.The extracted starch powder from banana peel

4.4. Chemical and physical properties of banana pseudostem fiber

The density of extracted fiber: Density was measured using the Archimedes principle by using water as liquid (Amiri, 2017). The density of the fiber after treatment was 1.4gm/cm^3. The results described the density was increased as the fiber treated more because of more polymerization gives higher molecular arrangements. Lignin and hemicellulose were removed and the denser component (cellulose) probably described the content of treated fiber. Before treatment, density was determined as 1.37 gm/cm^3.

Yield for banana pseudostem fiber: The yield of raw fiber from the pseudostem was analyzed by taking a bar of steam with known weight and taking the wet mass of fiber after extracted. Three replicates were recorded and calculated with equation 3.8. The average percentage yield of fiber in the pseudo steam is around 8%. (Vigneswaran *et al.*, 2015) describes the yield of banana stem fiber from bundle stem was around 10%.

which is almost approximated with the experimental result of 8% and the reason in the decrement of yield was probably the extraction process did not use a modern machine and simply with a knife. But this result is greater than the other author 2-4% (Kumar et al., 2013).

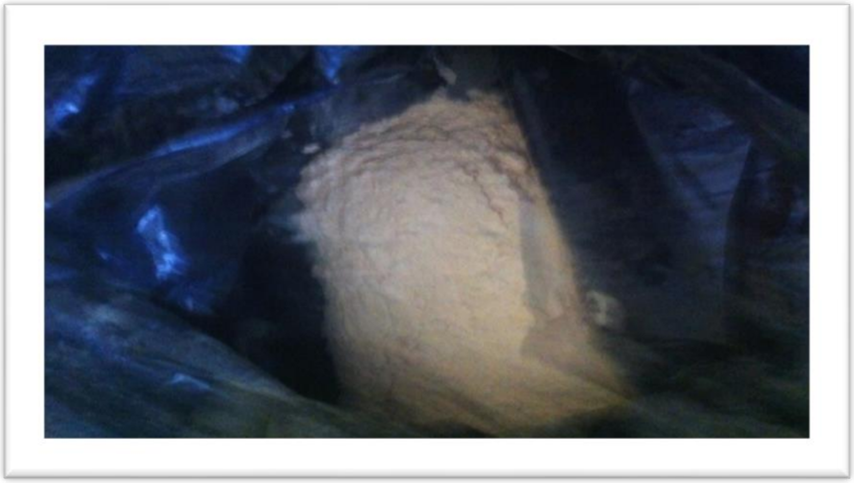


Figure: 4. 4.Extracted fiber from banana pseudo-stem

The moisture content of the extracted fiber: The moisture content of raw fiber used to determine amounts of water in the raw banana stem fiber was measured by taking three replicates of wet fibers the moisture content was calculated using equation 3.14. The average percentage of moisture content obtained was around 61.3% weight percent which is less moisture content than the work of (Ebisike *et al.*, 2013).

Which shows the moisture content of fresh banana fiber was around 90% which is higher than the value 61.3%? This may probably cause by the removal of moisture during transportation and the extraction process.

Water Absorption of the extracted fiber: The water absorption capacity of fiber was measured. The absorption capacity of fiber was around 4.6%. It is possible to minimize this number by treating with soda; during this, the hydroxyl group is substituted by sodium during the treating process (Kalia *et al*, 2009).

Cellulose and Non-Cellulose Content of the extracted fiber: Since the cellulose part of a banana, fiber has used for further processing; it is enough to know cellulose content and non-cellulose content.

Two trials were taken for test and the percentage cellulose content was around 55%-60% which matches the authors: (Mohiuddin *et al.*, 2014), describe the cellulose content of banana pseudostem fiber was around 50-60%.

And the other authors have also quantified the cellulose content of banana stem fiber between 80 and 85% (Kumar *et al.*, 2013). The cellulosic fiber was used for this work. The cellulose content of the raw fiber is higher than the other wood and non -wood source of fiber as already mentioned from the literature review part.

Thermal Characterization (TGA) for the extracted fiber: The thermal stability of fiber was tested by measuring the mass loss during a heating ramp rate by the thermogravimetric analyzer (TGA instrument) using temperature programming from 25 to 600^oC at the heating rate of $20^oC/min$ to which the sample was heated. The thermal degradation onset temperature and the thermal degradation weight loss of the fiber were recorded and analyzed as follows. The TGA analyses of fiber are shown in fig.4.5.

The fiber had three main weight loss regions. The initial small weight loss, which was caused by the loss of free water adsorbed in the fibers, occurred in the range of 45^0Cto 135 ^{0}C. The weight loss percentage was about 0.5% to 9.5%. The second weight loss in the temperature range of 260^0C to 360^0C. The weight loss percentage was about 15% to 60%. And it mainly contributed to the depolymerization of hemicelluloses and thermal decomposition of α-cellulose. The third weight loss region was due to the degradation of non-cellulosic substances such as lignin. Given that lignin is the most difficult constituent to decompose, its decomposition usually covers the whole temperature range of 200^0 C to 600^0C (S. Das.*et al.*, 2018). The above result obtained was almost similar to the literature done by (Teli & Terega, 2017) and *(*Shuying .*et al*, 2015). The final residue in the form of ash obtained was 9.88% at 604 ^{0}c, but 22% of residue was obtained by (Teli & Terega, 2017), this is due to the difference in operating temperature, the author uses up to 500^0c. The obtained result was summarized by the Figure 4.5 as shown below.

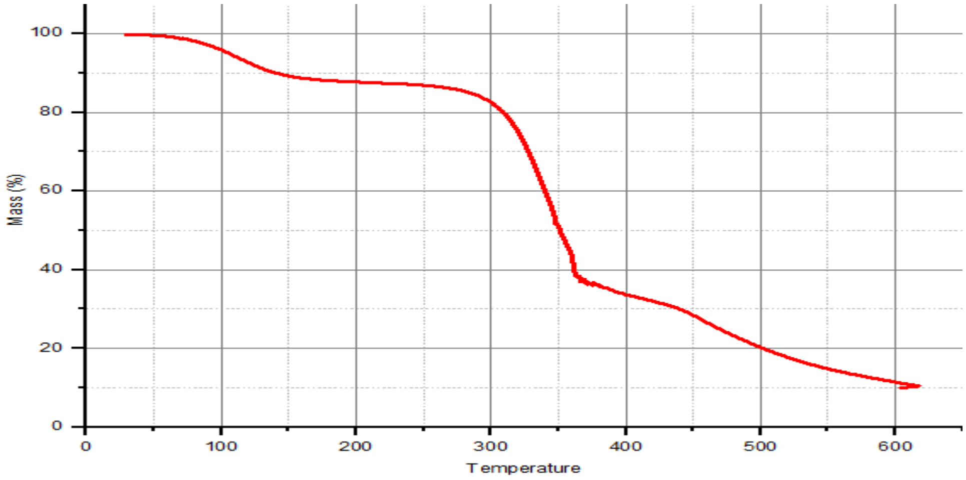


Figure: 4. 5.Thermograph (TGA) of banana pseudo-stem fiber

4.5. Synthesis of bioplastics and analysis on tensile strength, elongation at the break and water absorption

The experimental values of the tensile strength (TS), elongation at the break (EA) and water absorption (WA) obtained under different conditions are presented in Table 4.10.

These results were inputs to the DESIGN EXPERT® software version 6.0.8 and version 11 for further analysis and the statistical analysis of the conditions is discussed in the following section. The factors were oven-dry temperature, glycerol, and fiber concentration. The overall design summary is summarized in Table 4.9.

Table: 4. 9. Design Summary

Study Type	Response Surface
Initial Design	Central Composite
Experiments	20
Model	Quadratic

Table: 4. 10.Values of the three response variables associated with the three factors

Std	Run	Factor1 A: Dry. Temp (^{0}C)	Factor2 B: Glycerol conc. % (w/v)	Factor3 C: fiber conc. % (w/w)	Response1 TS(Mpa)	Response 2 EA (%)	Response 3 WA (%)
1	16	35	20	5	1.05	15.2	30
2	6	65	20	5	3.8	5.9	22.36
3	19	35	50	5	0.2	31.01	35.3
4	12	65	50	5	2.02	18.8	27.54
5	5	35	20	20	1.52	13.5	25.3
6	3	65	20	20	5.01	4.9	20.69
7	4	35	50	20	0.32	26.1	30.5
8	17	65	50	20	2.86	12	25.7
9	7	35	35	12.5	3.02	20.1	24
10	15	65	35	12.5	5.7	7.8	18.6
11	13	50	20	12.5	5.55	6.5	20
12	10	50	50	12.5	3.9	17	25
13	18	50	35	5	6.3	13.3	23
14	8	50	35	20	6.9	8.5	20.8
15	20	50	35	12.5	7.25	10	19.2
16	9	50	35	12.5	6.9	9.6	19.22
17	11	50	35	12.5	6.8	11	20.5
18	14	50	35	12.5	7.01	9.4	18.9
19	1	50	35	12.5	7.18	9.5	19.2
20	2	50	35	12.5	7.21	9.6	20.1

4.5.1. Statistical analysis of factors affecting the response variables

The design matrix and the corresponding results of RSM/CCD experiments to determine the effects of the three independent variables dry oven temperature, glycerol, and fiber concentration on tensile strength, elongation at the break and water absorption were shown in Table 4.10. The results were analyzed by using ANOVA (analysis of variance) appropriate for the experimental design used and shown in Table 4.11 and in Appendix Table B_1, B_2. Empirical models for the output responses of films in terms of the dry oven temperature, glycerol concentration and fiber concentration in actual and coded factors were developed by using RSM/CCD methodology. The sequential model sum of Squares are given from appendixes table B_9 for tensile strength response as a sample and was found that quadratic model was the most suitable model for the present study, the model is significant, had higher polynomial order, had no aliased terms, high R-squared, adjusted R-squared and predicted R-squared for all response variables. The ANOVA of the quadratic regression model indicates that the model to be significant. The Model F-value of 439.37, 182.1 and 159.97 implied the model to be signed for tensile strength, elongation at the break and water absorption respectively. There is only a 0.01% chance that a "Model F-value" this large could occur due to noise. Model P-value (Prob>F) is very low [<0.0001]. This reiterates that the model is significant. The P values are used as a tool to check the significance of each of the coefficients, which in turn are necessary to understand the pattern of the mutual interactions between the test variables. The smaller the magnitude of the P, the more significant is the corresponding coefficient. The values of P less than 0.0500 indicate the model terms to be significant. For tensile strength, the coefficients estimate and the corresponding P values suggest that, among the test variables used in the study, in this case, A, B, C, A^2, B^2, $C^{2,}$ AB, and AC are significant model terms as shown in Table 4.11. For elongation, at the break, the coefficients estimate and the corresponding P values suggests that, among the test variables used in the study, In this case, A, B, C, AB, BC, A^2, and B^2 are significant model terms and for the water absorption the coefficients estimate and the corresponding P values suggests that, among the test variables used in the study, In this case, A, B, C, AC, A^2, B^2, and C^2 are significant model terms. Values which greater than 0.1 indicate the model terms are not significant. The "Lack of Fit F-value" of 0.7923, 1.94 and 0.4192 implies the lack of fit is not significant relative to the pure error for tensile strength, elongation at the break and water absorption respectively. There is a 59.770%, 24.29% and 81.90% chance for tensile strength, elongation at the break and water absorption respectively that a "Lack of Fit F-value" this large could occur due to noise. Non-significant lack of fit is good because the model needs to fit.

Table: 4. 11. Analysis of variance [Partial sum of squares], for tensile strength

Source	Sum of squares	Mean squ.value	F-Value	Prob>f	
Model	119.55	13.28	439.37	< 0.0001	Significant
A-oven T.	17.64	17.64	583.33	< 0.0001	
B-gly.conc.	5.82	5.82	192.56	< 0.0001	
C-FB	1.05	1.05	34.72	0.0002	
AB	0.4418	0.4418	14.61	0.0034	
AC	0.2665	0.2665	8.81	0.0141	
BC	0.0648	0.0648	2.14	0.1739	
A^2	17.27	17.27	571.19	< 0.0001	
B^2	12.60	12.60	416.91	< 0.0001	
C^2	0.1944	0.1944	6.43	0.0296	
Residual	0.3023	0.0302			
Lack of Fit	0.1336	0.0267	0.7923	0.5977	not significant
Pure Error	0.1687	0.0337			
Cor Total	119.85				

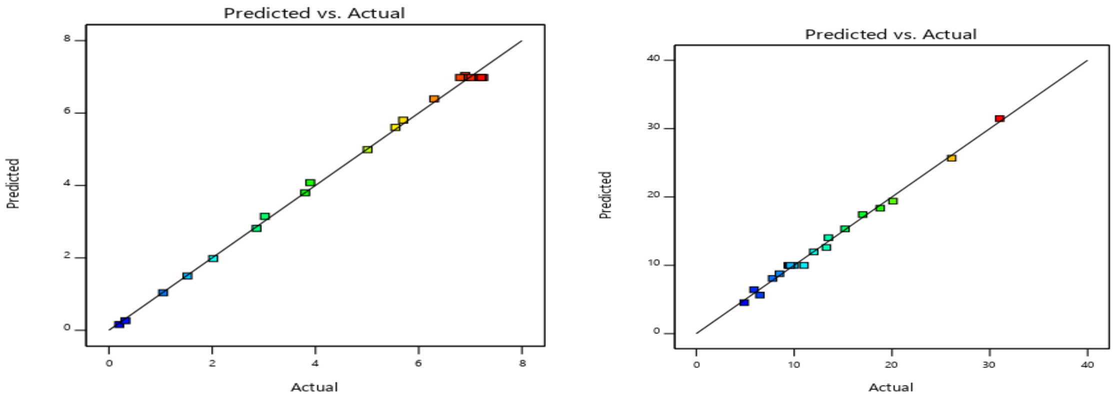
4.5.2. Adequacy check for the developed response surface quadratic models

The predicted R^2 of 0.9932, 0.9529 and 0.9789 is in reasonable agreement with the adjusted R^2 of 0.9952, 0.9885 and 0.9869 for tensile strength, elongation at the break and water absorption respectively. Adequate precision measures the signal to noise ratio. A ratio of greater than 4 is desirable. This model can be used to navigate the design space. The fit of the model was also expressed by the coefficient of regression R^2, which was found to be 0.9975, 0.9939and 0.9931 indicating that 99.75%,99.39% and 99.31% the variability in the response could be explained by the model for tensile strength, elongation at the break and water absorption respectively. The closer the value of R^2 (correlation coefficient) to 1, the better is the correlation between the experimental and predicted values. Here the value of R^2 (be 0.9975, 0.9939and 0.9931) being close to 1 indicated a close agreement between the experimental results and the theoretical values predicted by the model equation. This implies that the prediction of experimental data is quite satisfactory. The coefficient of variation (CV) indicates the degree of precision with which the experiments are compared. Generally, the higher the value of the CV is, the lower the reliability of the experiment. Here a lower value of CV (3.84, 5.59 and 2.27 for tensile strength, elongation at the break and water absorption respectively) indicates greater reliability of the experiments performed. Press is the predicted residual sum of squares for the model which is a measure of how well a particular model fits each point in the design; Adequate precision is a measure of the range in predicted response relative to its associated error, in other words, a signal to noise ratio and its desired value is 4 or more. In this case, the ratio of 55.9441, 52.4515and 44.5626 for tensile strength, elongation at the break and water absorption respectively indicates an adequate signal which could be used to navigate the design space or decide whether the model can be used or not. Model adequacy measures for tensile strength, elongation at the break and water absorption shown in Table 4.12 and in Appendix part, Table B_3 and B_4.

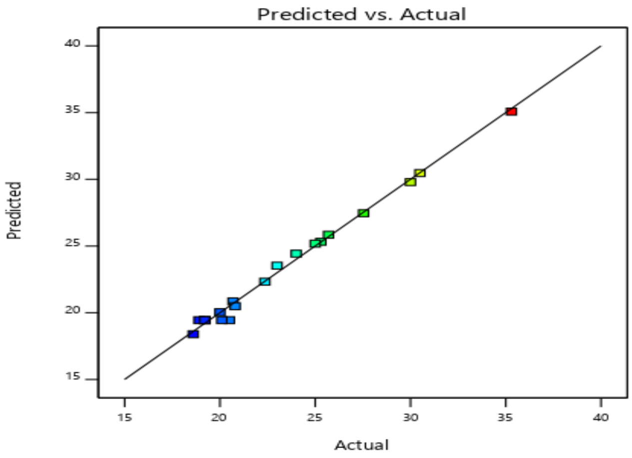
Table: 4. 12.Model adequacy measures for tensile strength

Std. Dev.	0.1739	R²	0.9975
Mean	4.52	Adjusted R²	0.9952
C.V. %	3.84	Predicted R²	0.9932
PRESS	0.82	Adeq Precision	55.9441

The adequacy of the model was further checked by through graphical analysis of predicted versus actual, outlier T versus run number, normal probability plot of residuals versus studentized residual, and residual versus run number.



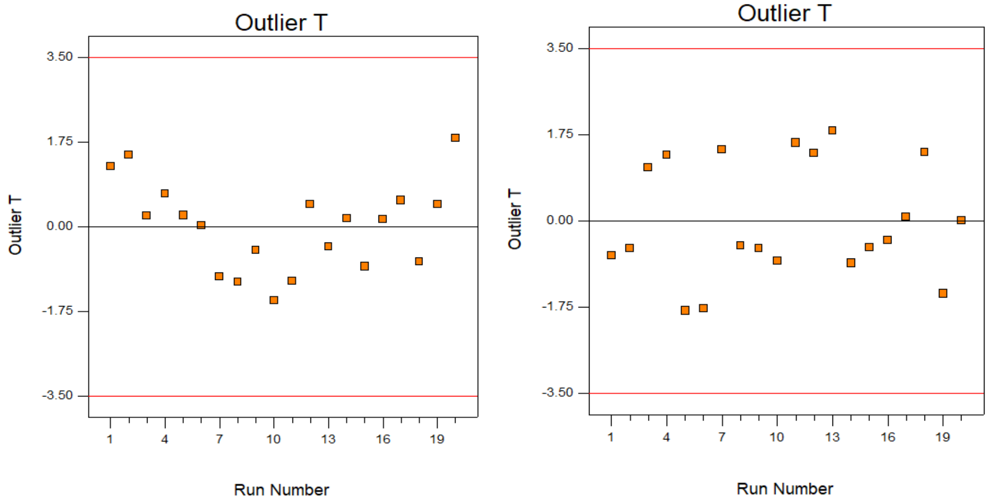
A. Tensile strength B. Elongation



C. Water absorption

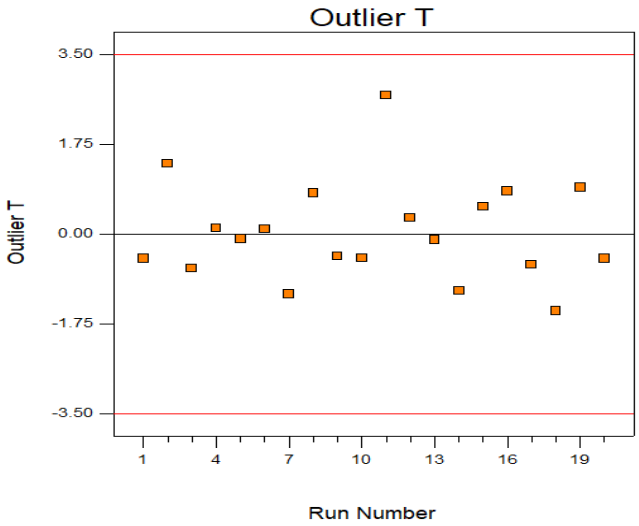
Figure: 4. 6.Predicted versus Actual plots

From the above Figures 4.6, the plots represent the line of a perfect fit with points corresponding to zero error between predicted values and actual values and demonstrated that the regression model equation provided an accurate description of the experimental data for all responses, in which all the points are close to the line of a perfect fit. Also, model adequacy can be checked by the plot of outlier T vs. run number which gives information about whether experiments were done in appropriate condition or not.



A. Tensile strength

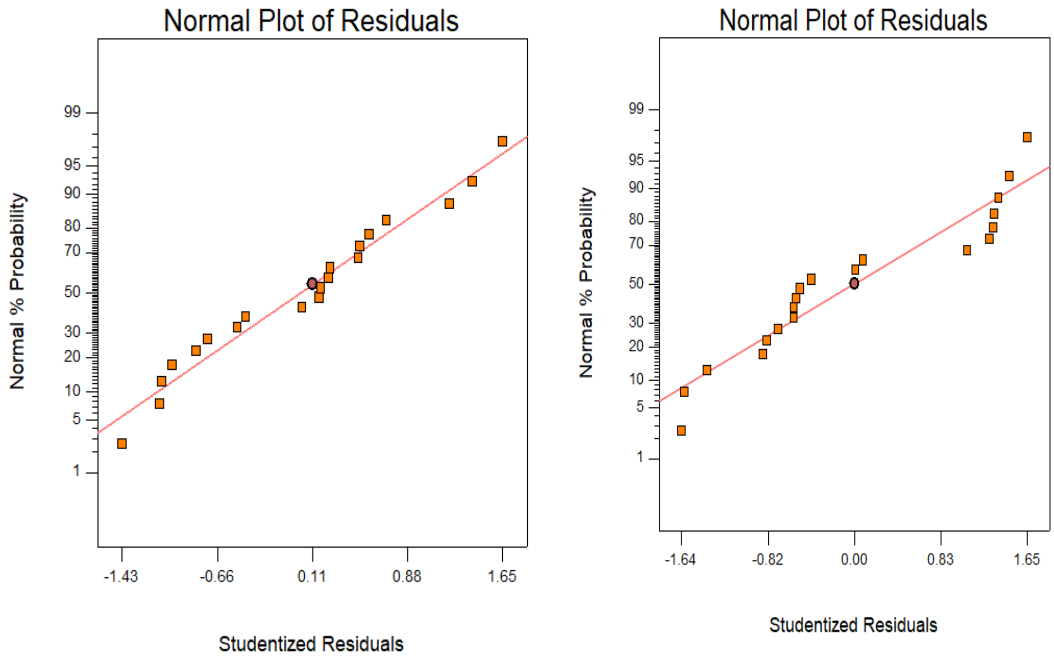
B. Elongation



C. Water absorption

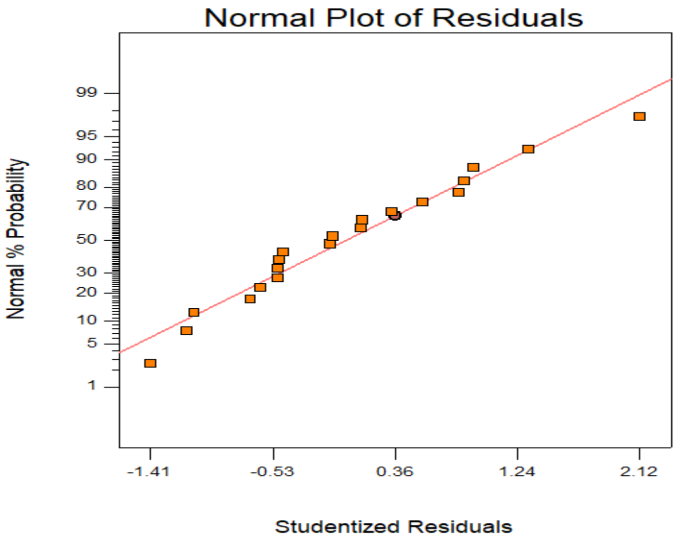
Figure: 4.7.Outlier versus Run Number Plot

The plots of outlier T vs. run numbers presented in Figure 4.7 above. Hence from the outlier vs. run number show, each experiment is within the outlier T boarder. It implies that no bad data and all experimental runs were done in the appropriate condition. Hence no experiments repeat again.



A. Tensile strength

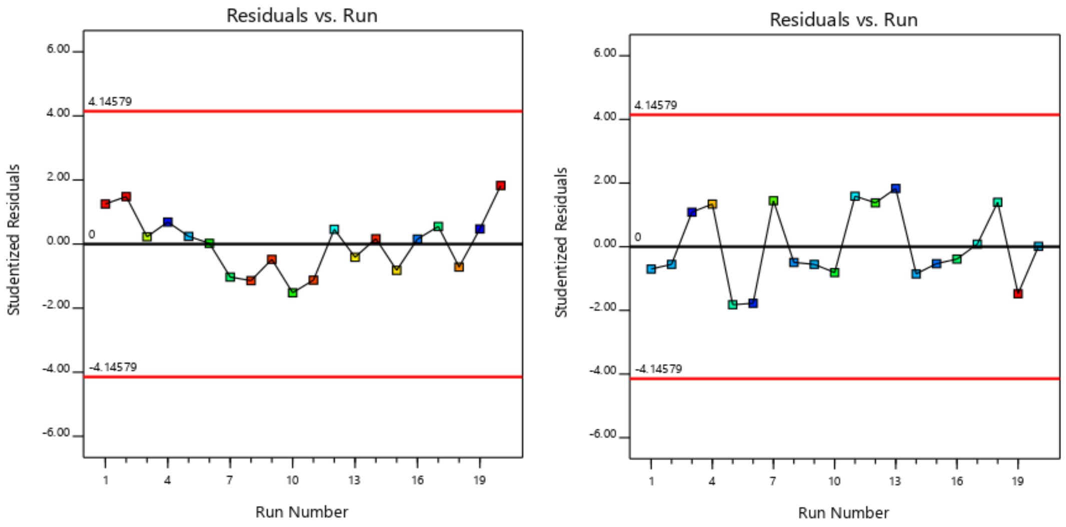
B. Elongation



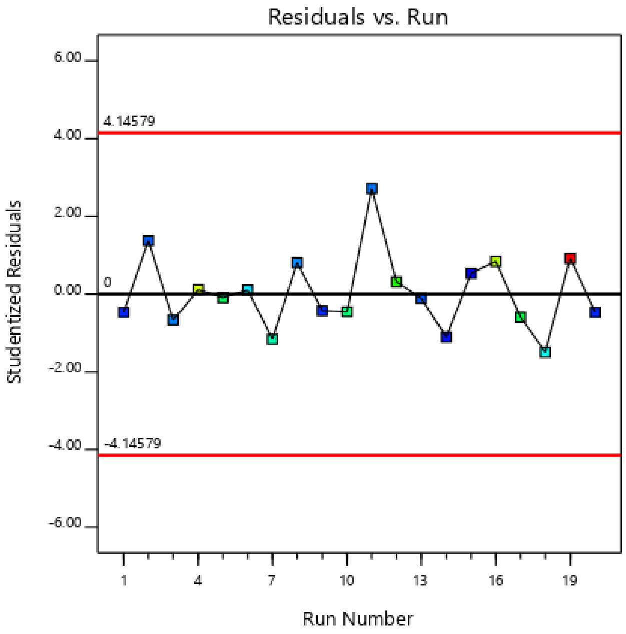
C. Water absorption

Figure: 4. 8. Normal Probability Plots

Data have been collected in a randomized run order or some other order that is not increasing or decreasing in any of the predictor variables used in the model to emphasize in experiment design. Hence it is better to check whether the data collection method has an effect on the adequacy of the model by the graphical method as shown the Figure 4.9 below.



A. Tensile strength B. Elongation



C. Water absorption

Figure: 4.9.Residual versus Run Number Plots

4.5.3. Development of Regression Model Equation

The regression coefficients of the developed model were determined from the regression analysis. As indicated in fit summery which is given from the appendixes Table B_8, for each factor, the quadratic models are suggested, as the p-value of this model is smaller than that of other models and no aliased terms. The model equation that correlates the response to the process variables in terms of actual value and codded was formulated. Final equations in terms of actual and coded factors for all responses are given in appendix C.

4.5.4. Effects of individual process variables

The effect of oven-dry temperature, glycerol and fiber concentration on tensile strength, elongation, and water absorption was studied and evaluated for best bioplastic film synthesis conditions. Based on the analysis of variance, plastic synthesis was significantly affected by main individual factors and various interactions between the process variables. In addition to the interaction effect, individual process variables significantly affect the three responses.

1. Effects of dry oven temperature on tensile strength (TS), elongation (EA) and water absorption (WA).

In Figure 4.10 the dry oven temperature varies from 35 to 65^0C and both fiber and glycerol concentration kept constant at 12.5 and 35% respectively. The increase in dry oven temperature improves the tensile strength and provides better quality for the produced film. But as the temperature is beyond the range or rises, it was difficult to peel off the film at the Petri dish. As shown in Table 4.10 the minimum tensile strength was obtained at 35^0C which is 0.2Mpa and the maximum tensile strength was 7.25Mpa at 50^0C. From the Figure 4.10 (A), it can be observed that at dry oven temperature is small (35^0C), the tensile strength of the film was minimum (3.02Mpa), when drying temperature rises into 50^0C, tensile strength of the film increased into 7.03Mpa, and finally, when drying temperature rises into 65^0C, tensile strength reduced from 7.03 to 5.7Mpa.

This can be caused by the influence of higher temperatures that can cause intermolecular bonds in starch chains becoming weaker. The hydrogen bonds between amylose chains undergo termination of the bond. Then further heating will break glucosidic bonds (bonds between monomers) in amylose. Increasing the heating temperature can cause depolymerization in the amylose chain, the straight-chain amylose falters and becomes shorter, thus decreasing amylose content(Halley *et al.*, 2001). Generally, as drying temperature increased, tensile strength increased up to a center point (50^0C) then tensile strength decreased and finally it becomes parabolic, this implies the center point favors the tensile strength of the film.

Table 4.10 shows that, the increase in the dry oven temperature decrease elongation at the break from 31.01% to 4.9 % and water absorption of the film from 35.3 % to 18.6%. From the Figure 4.10 (B), it illustrated that percent elongation at the break was 20.1% at 35^0C when temperature increase to 50^0C %EA reduced from 20.1 into 10%, finally when drying temperature reaches in 65^0C, % EA decrease into 7.8%. This was due to increasing the tensile strength of bio-plastic as well as increasing drying temperature from the lower level up to a center point. The result of elongation at break decreases with increasing the temperature and as shown from the figure. This is because the heat that is given causes an increase in the kinetic energy of the molecules in which the molecules vibrate and create a free volume to allow larger molecular chain rotation (Wojciechowska, 2014). So, as the temperature increases %EA decreases, this implies lower elongation is a favor for the synthesized film.

As the temperature rises both the distilled water and glycerol evaporate to some extent the film dries very well and the water absorption of the film reduced. The effect of drying temperature on water absorption of bio-plastic was shown in figure 4.10 (C). As shown in table 4.10, the lowest water absorption was observed at a higher level or at 65℃ which is 18.6%. Figure 4.10 (C) indicates at minimum dry temperature (35^0C) water absorption (WA) is 24%, when drying increases to 50^0C, WA decrease into 19.7%, then decreased into 18.6% when the temperature is kept at a high level (65^0C).

This tells us the higher drying temperature gives the lower water uptake of the film. As shown in appendix C, the coded equation of the regression model, dry oven temperature affects the TS positively and it affects the EA and WA negatively. Notice that the value of the three responses at the center (50^0C drying temperature) was taken the average of the replicate.

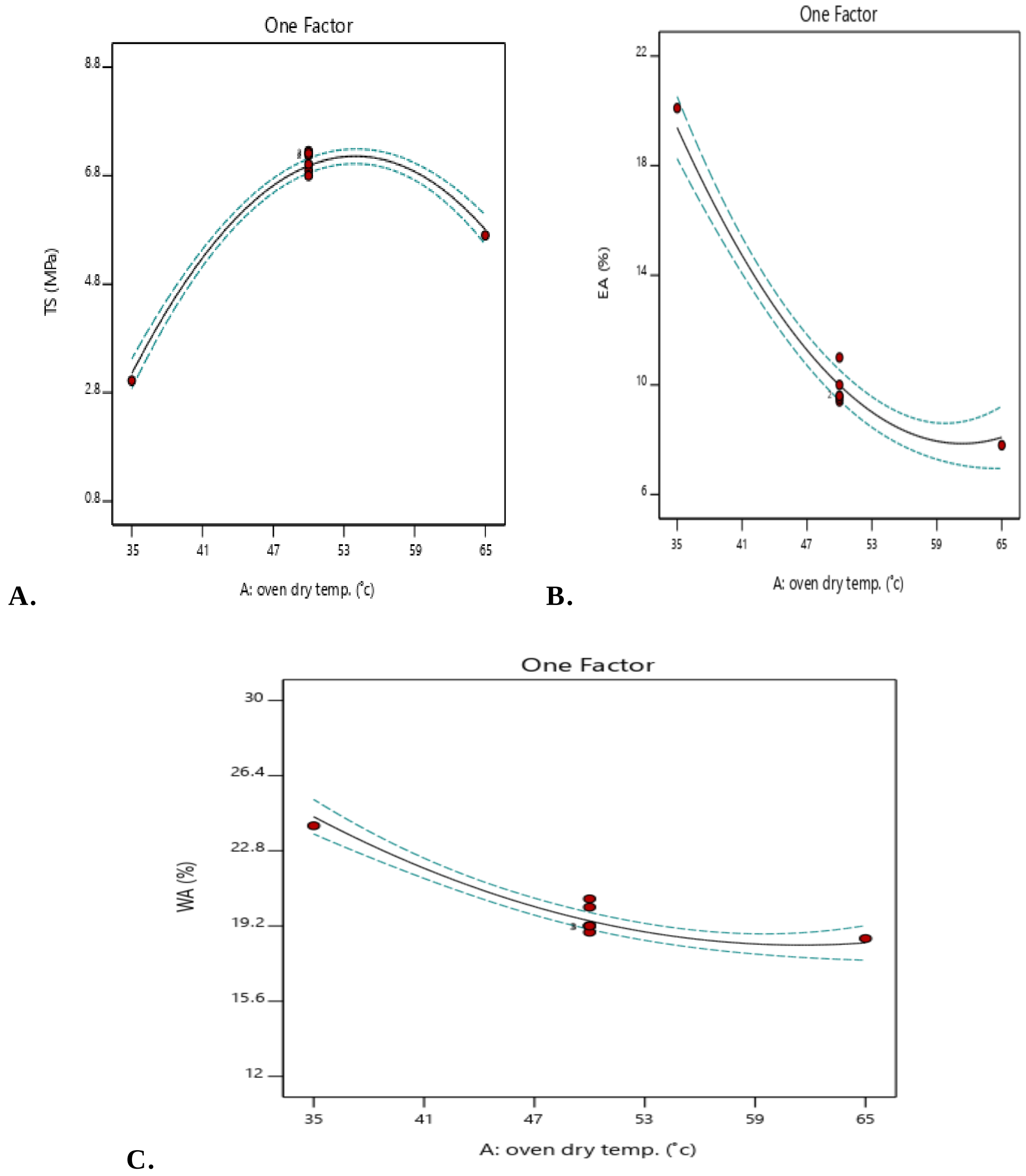


Figure: 4. 10.Effects of dry oven temperature on responses (A) tensile strength, (B) elongation and (C) water absorption.

2. **Effects of glycerol concentration on tensile strength, elongation and water absorption.**

In Figure 4.11 the glycerol concentration varies from 20 to 50% w/v starch basis but fiber concentration and dry oven temperature were taken constantly at 12.5% and 50^0C respectively. Figure 4.11(A) at a minimum glycerol concentration (20%) the tensile strength of the film was 5.55Mpa, when glycerol concentration increased from 20 to 35%, the tensile strength of the film was increased from 5.55 to 6.92Mpa, and finally when glycerol concentration raises into 50% the tensile strength of the film decline into 3.9 Mpa. As the glycerol concentration is beyond the range or rises more it was difficult to peel off the film and the film becomes highly flexible. So controlling the glycerol concentration is important. As shown in table 4.10, the minimum tensile strength was obtained at 50% gly.conc which is 0.2Mpa and the maximum tensile strength was 7.25Mpa at 35% glycerol concentration, the tensile strength increase due to increment of dry oven temperature up to 50^0C. These results agree with the previous works on cassava starch by (Tegangan & Air, 2011). As the glycerol concentration reaches beyond the saturation/increases, tensile strength declines (Mardhiah *et al.*,2017) and (Gozan & Noviasari, 2018). Generally, the medium glycerol concentration (35%) favors getting maximum tensile strength (6.92Mpa) of the film.

Table 4.10 shows the increase in glycerol concentration from 20 to 50%, an increase in both elongation at the break from 4.9 % to 31.01%, and water absorption of the film from 18.6% to 35.3 %. As shown in figure 4.11 (B) at glycerol concentration (20%), EA is 6.5%, and when glycerol concentration increased into 35% EA also increased into 9.25%, finally when glycerol reaches to 50% EA increased into 17%. Figure 4.11(C) shows at 10% glycerol concentration water uptake of the film was 20%, but as the glycerol concentration rises into 35% water absorption becomes 19.7%, finally, as glycerol concentration reaches at its higher level(50%) water absorption become maximum(25%).

The possible reason for the high tensile strength at low plasticizer concentration is the domination of strong hydrogen bonds produced by a starch-starch and starch-fiber intermolecular interaction over starch-plasticizer attraction (Sanyang *et al.,* 2015) and (Lutfi *et al.,* 2017).

Notice that the value of the three responses at the center (35% glycerol) was taken the average of the replicate. When glycerol concentration is increased, the hydrogen bond weakened the polymer mobility increase. From the result, it was observed that glycerol concentration affects the tensile strength negatively, but it affects EA and WA positively. As shown in the figure below glycerol concentration increase, elongation increase linearly, this situation is similar to (Leblanc, 2012) and (Lutfi *et al.*, 2017) justification. The decrease in extension at break value was probably due to more plasticizer content which may lead to lower interaction between polymer chains. An increasing the amount of glycerol will increase the composite mobility but it will affect the mechanical properties by lowering the strength of the samples. The three response value at the center point was taken as the average of the replicate. The effect of glycerol concentration at constant dry oven temperature (50^0C) and fiber conc. (12.5%) is shown below. The individual effect of the whole factor for each response also found in appendix J, of the perturbation graph.

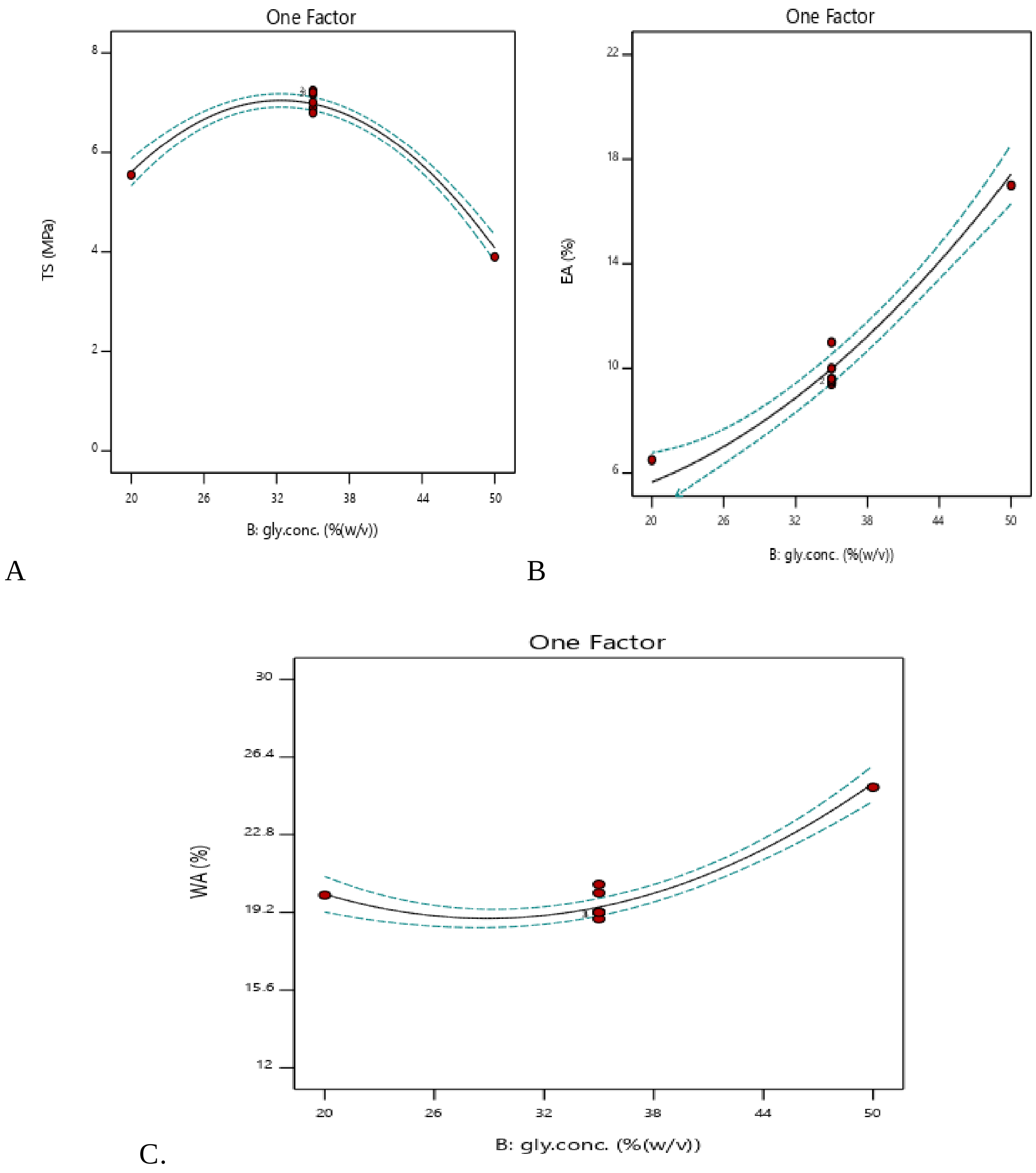
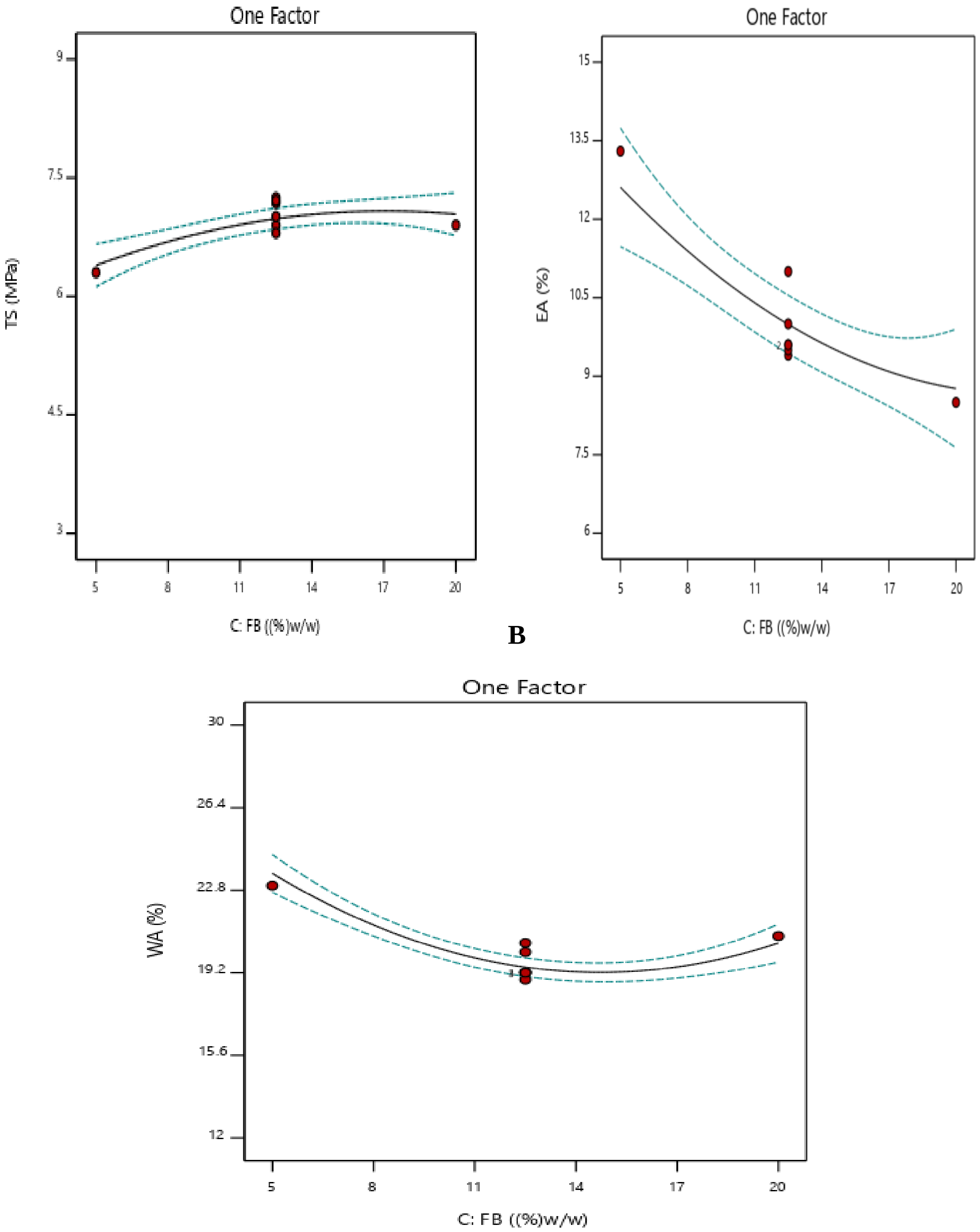


Figure: 4.11.Effect of glycerol concentration on response; tensile strength (A), elongation (B) and water absorption (C).

3. **Effects of fiber concentration on tensile strength, elongation and water absorption.**

Both drying temperature and glycerol concentration were kept constant as 50^0C and 35%. As shown from Figure 4.12(A) when fiber concentration is at lower (5%) tensile strength is 6.3 Mpa.

When fiber concentration increased in center point (12.5%), the tensile strength of the film enhanced (7.03 Mpa), finally as fiber concentration increased into the higher level (20%) the tensile strength decreased into 6.9 Mpa. Generally, as fiber concentration increased up to a center point, tensile strength increases, beyond the center point it decreases. This increment is due to the stronger bonding between the fiber and the starch matrix, beyond the center point to some extent decrease smoothly, Probably this effect is due to an interaction of the fiber with the starch, decreasing the molecular mobility and resulting in more rigid and less flexible materials (Mekawy, 2016). The researcher describes that at 0 % matrix tensile strength was 3Mpa and at 15% fiber, 9Mpa was recorded (Guadalupe *et al.*, 2011). As shown from Figure 4.12 (B) the fiber content 5% elongation at the break was maximum (13.3%) and as fiber concentration increased up to a center point (12.5%) elongation decreased into 10%. Finally, as fiber concentration increased into a higher level (20%) elongation was reduced to 8.5%. As the matrix of synthetic or biological polymers increased the values of the tensile strength of bioplastics increased (up to a center point) and decreased the values of elongation at fracture. This is in agreement with other literature (Guadalupe *et al.*, 2011*)*. The sensitivity of the composites to water absorption from the environment may be one of the most important parameters to be undermined for some applications, as water absorption affects the properties of starch-based composites (Gadhave *et al.*, 2018). As shown from the Figure 4.12 (C) the fiber content 5% water absorption was maximum (23%) and as fiber concentration increased up to a center point (12.5%) water absorption decreased into 19.7%, finally as fiber concentration increased into a higher level (20%) water absorption was slightly increased into 20.8%. This water absorption reduction in bio-composites can be attributed to stronger hydrogen bonds between the matrix and the reinforced material. The three response value at the center point was taken as the average of the replicate. In fact, the incorporation of fibers in thermoplastics as an alternative for reducing water uptake in bio-composite exposed to high humidity environments, but the ratio of fiber to starch should be considered (Wittaya, 2012).



A.

B

C.

Figure: 4. 12.Effects of fiber concentration on responses; tensile strength (A), elongation (B) and water absorption(C).

4.5.5. **The interaction effect between process variables and responses**

To study the interactive effect of factors on the tensile strength, elongation, and water absorption, the response surface methodology was used and the 3D surface was drawn. Response surface plots as a function of two factors at a time, maintaining another factor at fixed levels are more helpful in understanding the interaction effects of these factors. The 3D response surface graphs as shown in Figure 4.13 to 4.15 shows that a significant interaction between every two variables, as it can be seen from coded equation (appendix C) the interaction factor effects on tensile strength, elongation and water absorption can be understood easily by the coefficients of interaction factors. In this section, the significant interaction effect of the factors is clearly discussed. There are three interaction factors analyzed by the model equation. These are AB (dry oven temperature and glycerol concentration), AC (dry oven temperature and fiber concentration) and BC (glycerol concentration and fiber concentration). Among these three interaction factors, the one which consists of higher coefficients at the code regression models is the most significant interaction factor for the response. The sign of the coefficient of the interaction factor indicates the effect of the interaction factor on responses. Therefore, the interaction factors with positive signs have a positive effect on the responses. Whereas, interaction factors with negative signs have a negative effect on the responses (TS, EA, and WA). The interaction graph was easily shown by the 2D graph in the appendix part of appendix E. The significant interaction factors are clearly discussed in the following sections.

1. Interaction effects of dry oven temperature and glycerol concentration on tensile strength, elongation and water absorption

Figure 4.13(A, and B) of the 3D represents the interaction effects of dry oven temperature and glycerol concentration on tensile strength and elongation at constant fiber concentration (12.5%). Varying dry oven temperature and glycerol concentration had a significant (P-values less than 0.0500) effect on tensile strength and elongation value.

The increase in dry oven temperature and the glycerol concentration up to a center point enhance the tensile strength of the film. This is also true according to (Gujar *et al.*, 2014) justification, this author also mentions the maximum TS is obtained at 1 mL. And at the lower temperature and higher glycerol concentration, the minimum tensile strength was recorded. On the other hand, when glycerol concentration increases smoothly at the starting point that means from lower level to the center point the maximum tensile strength (7.25Mpa) was recorded, this is due to the perfect ratio of mixing between the glycerol and the starch matrix with fixed fiber concentration, Since, the ratio reaches a saturation point (Lutfi et al., 2017).

The increase in dry oven temperature at constant initial glycerol concentration the tensile strength increase and become maximum at a center point(35% conc.). Dry oven temperature increase and glycerol concentration decrease the elongation and water absorption increase, on the other hand, dry oven temperature decreases and glycerol concentration raises both elongation and water absorption increase, this is because the higher mobility of the polymer due to increase glycerol, enables the film to absorb moisture over time which is likely due to the hydrophilic nature of glycerol (Gujar *et al.*, 2014).

The maximum EA was recorded at the minimum dry oven temperature and at the maximum glycerol conc. (50%). On the other hand, the minimum EA was recorded at the maximum temperature (65°C) and minimum glycerol concentration (20%).

WA was not significantly affected by the interaction of dry oven temperature and glycerol concentration. But as shown in the coefficient of a coded equation in appendix C, this interaction had a small negative coefficient, so it affected in a small manner. The above justification is clearly shown by the interaction 2D graph in the appendix part. Generally, as dry oven temperature and glycerol concentration increase, tensile strength increases up to a center point, and then it decreases.

And as dry oven temperature and glycerol concentration increase both EA and WA decrease. The interaction effect was shown below in Figure 4.13(A, and B).

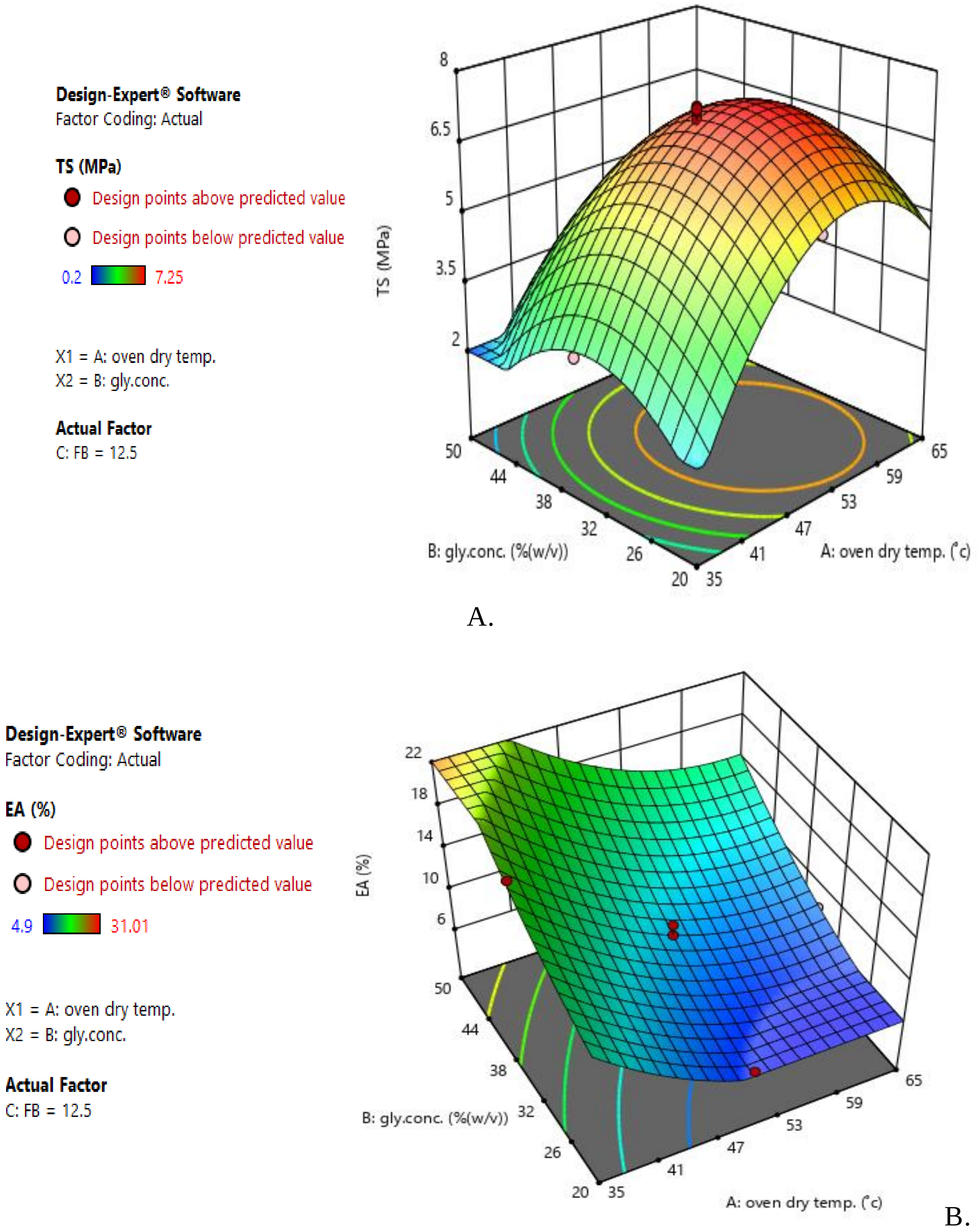
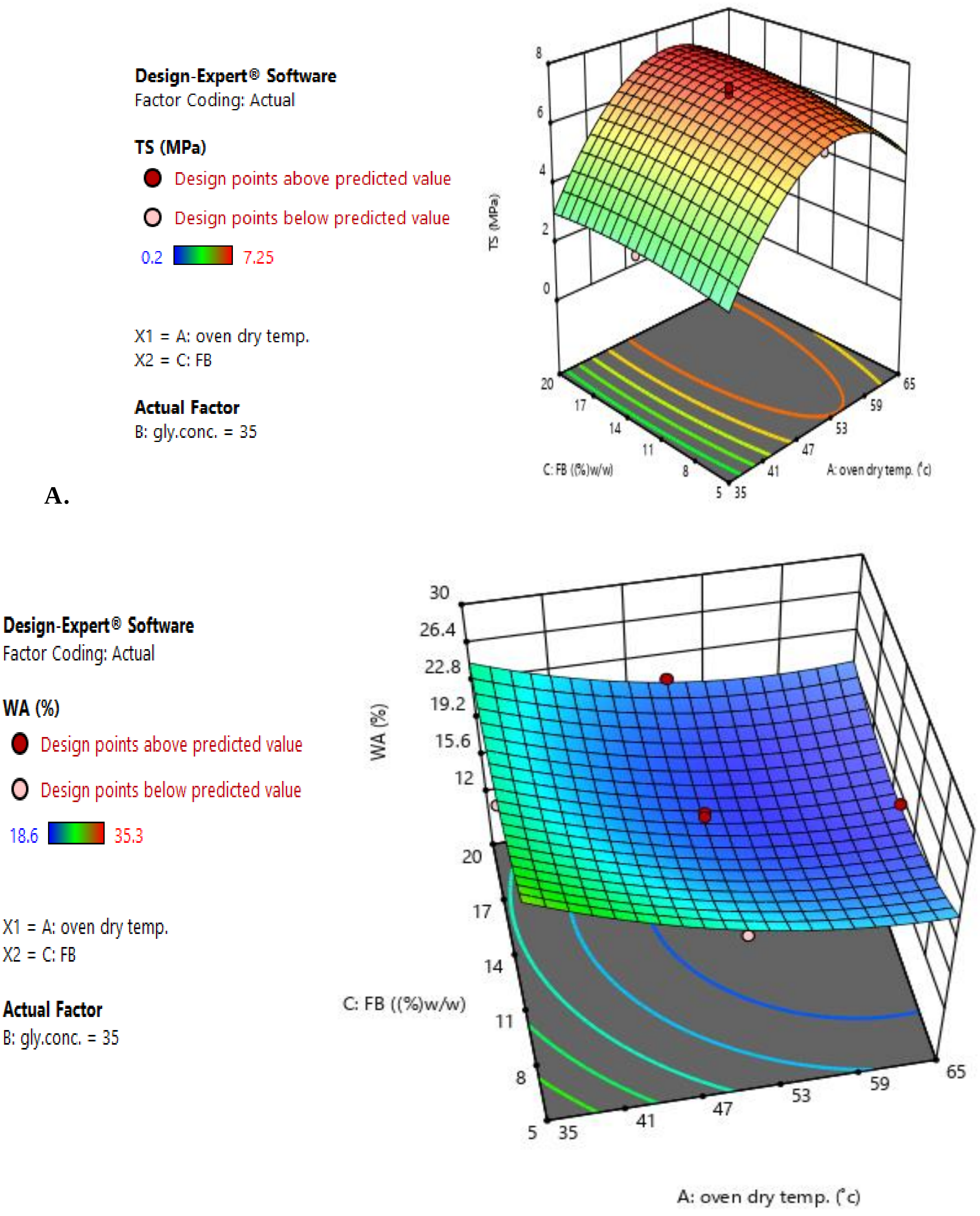


Figure: 4.13.Interaction effects of dry oven temperature and glycerol concentration on tensile strength (A) and elongation (B)

2. Interaction effects of dry oven temperature and fiber concentration on the tensile strength, elongation and water absorption

Figure 4.14 (A and B) of the 3D shows that the effects of dry oven temperature with fiber concentration and their mutual interaction on the tensile strength and water absorption at constant glycerol concentration (35%).

The increase in dry oven temperature and fiber concentration increases the tensile strength. The highest response value 7.25 Mpa was observed at 12.5% fiber concentration 35% glycerol concentration, further increased the temperature and fiber decreases the tensile strength due to the degradation of starches. Then both fiber concentration and dry oven temperature rises, tensile strength increases up to the center point, finally it decreases. The interaction graph tells this situation. From the analysis of variance, it indicates also AC/ interaction of dry oven temperature with fiber concentration affects the response tensile strength significantly (p-value less than 0.05). Then as both dry oven temperature and fiber concentration decrease, the tensile strength of the film also decreases. The coded equation of the tensile strength also shows AC with a large positive coefficient. On the other hand, both dry oven temperature and fiber concentration rise, EA and WA become decreases, and as both dry oven temperature and fiber concentration decrease, EA and WA become increases. The effects of interactions of dry oven temperature with fiber concentration (AC) had a significant effect on the TS (tensile strength) and water absorption (WA), but not for EA. As fiber concentration and dry oven temperature decrease, EA increased from 4.9 to 31.01% and WA rises from 18.6 to 35.3%. The above justification is easily shown by the interaction graph in the appendix (E) part. The coded equation also shows a large positive coefficient for WA and a very small negative coefficient for EA.



B.

Figure: 4.14. Interaction effects of dry oven temperature and fiber concentration at 35% gly conc on tensile strength (A) and water absorption (B).

3. Interaction effects of both fiber and glycerol concentration on the tensile strength, elongation and water absorption

Figure 4.15 of the 3D represents the effects of fiber concentration with glycerol concentration and their mutual interaction on the elongation of the film**.** Varying fiber concentration and glycerol concentration had a significant effect on the EA value. As both fibers concentration and glycerol concentration increase EA increase. Probably this effect is due to an interaction of the fiber with the glycerol, increasing the molecular mobility and resulting in less rigid and high flexible materials; the glycerol concentration dominates the fiber concentration.

Interaction effects of both fiber and glycerol concentrations significantly affect EA, but the effect is insignificant for both TS and WA. The coded equation also shows a large positive coefficient of AC for EA and very small negative coefficients of AC for both TS and WA. The above justification is easily shown by the interaction graph in appendix C.

The significant interaction effects of the independent variables on the three responses were clearly shown in the appendix K of the contour plot.

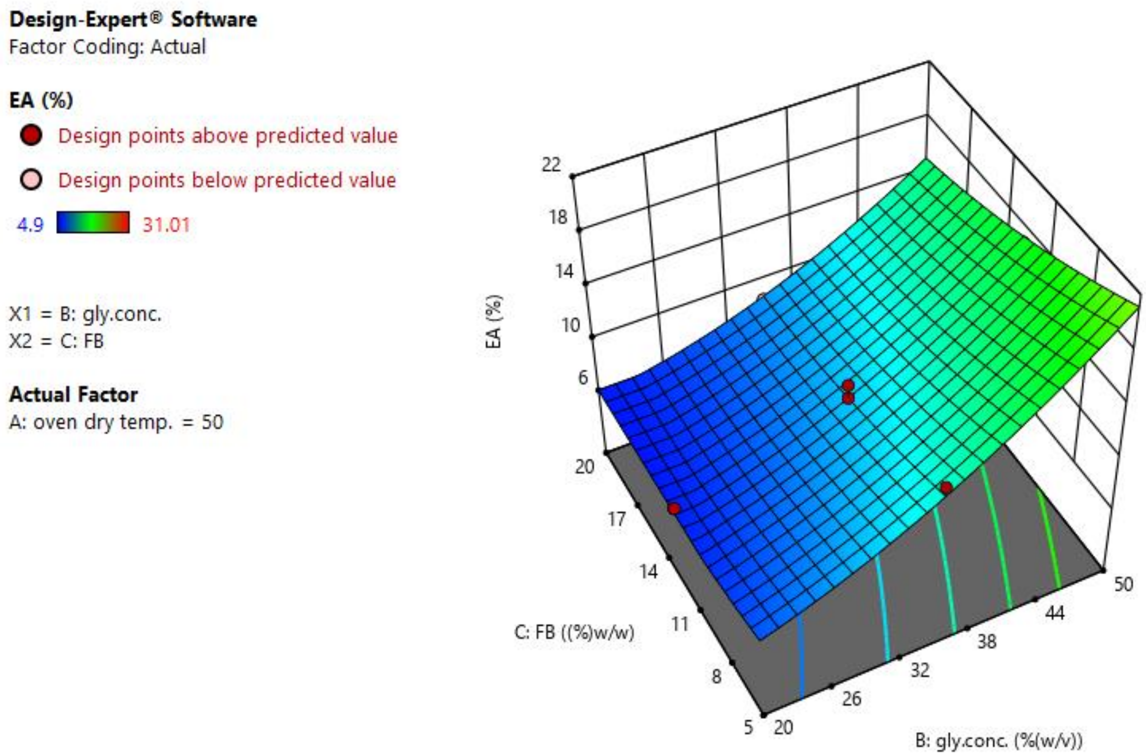


Figure: 4.15. Interaction effects of fiber and glycerol concentration on elongation when dry oven temperature fixed at 50^{o}C.

4.5.6. Process factors and response variables optimization

Optimization is the process of finding conditions that give maximum or minimum values of a particular function. Optimization is the act of getting the best possible result under given circumstances. In design, construction, maintenance... engineers have to make decisions. The goal of all such decisions should be either to minimize effort or to maximize benefit. The effort or benefit can be usually expressed as a function of certain design variables. Hence, optimization is the process of finding the conditions that give the maximum or the minimum value of a function.

Optimization of bioplastic film production process factors and responses was carried out by a multiple response method called desirability (D) function to optimize different combinations of process parameters. The process factors oven-dry temperature, glycerol concentration, and fiber to starch ratio and the process responses are optimized based on the principles of optimization.

The principles of optimization tell us to maximize the economic benefit by minimizing process cost, the process variables need to set as much as possible at their minimum value and the three response variables tensile strength, elongation at the break and water absorption were set to maximum, minimum and minimum respectively. Numerical optimization was used to optimize the process.

Table: 4. 13.Summary of constraints and goals of optimizations

Constraints Name	Goal	Lower Limit	Upper Limit	Lower Weight	Upper Weight	Importance
A: oven-dry temp.	is in range	35	65	1	1	3
B: gly.conc.	is in range	20	50	1	1	3
C:FB	is in range	5	20	1	1	3
TS	Maximize	0.2	7.25	1	1	3
EA	Minimize	4.9	31.01	1	1	3
WA	Minimize	18.6	35.3	1	1	3

Based on desirability analysis a total of 4 optimum points via numerical optimization (appendix table 11) generated from this the more preferred one is selected based on the operating cost and the product quantity as well as quality. Desirability function was used to identify the optimum levels of factors and to get maximum desirable responses and the optimized combinations of process variables were selected among the solutions generated with maximum combined desirability value i.e. 0.993 from shown in Table 4.14.

Table: 4. 14.Optimum operating point

D. oven temp	Gly.conc.(%)	FB %	TS	EA	WA	Desirability
56.516	28.603	18.264	7.247	5.461	18.600	0.993

In order to verify this prediction, experiments were conducted and the results showed that 7.25Mpa tensile strength, 5.49% elongation at the break and 18.70 % water absorption was obtained as shown in Table 4.15. Therefore, the numerical optimization can be taken as an optimal value because the predicted value is close enough with experimental. The model capable of predicting the maximum tensile strength, and the minimum elongation and water absorption value showed from Table 4.15, that the optimum values of the process variables were 56.5^{0}C dry oven temperature, 28.6%glycerol concentration, and 18.23% fiber.

Table: 4. 15.Model validation

Number	Dry oven temp.	Gly. conc.	FB	TS Mpa	EA%	WA%
Pridicted	56.52	28.60	18.26	7.23	5.46	18.60
Experimental	56.5	28.6	18	7.25	5.49	18.70

The tensile strength of banana peel via banana pseudo-stem fiber-based bioplastic film was found to be 7.23Mpa, which was higher than the bioplastics produced from cassava (4.5Mpa), corn starch (3.59Mpa), potato starch (4.87Mpa) (Al et al., 2016) and banana peel (0.443-1.596Mpa) (Mukhopadhyay *et al.*, 2017). Tensile strength of (8.29 -24.87 N/mm^2) plastic from waste paper also reported (Ezeoha, 2013). The value obtained showed that tensile strength of banana peel bioplastics film was low when compared with other species such as wheat flour (20.30 N/mm^2) (Harunsyah *et al.*, 2017), antimicrobial film(19.52 Mpa) (Muche, 2018), while higher than chitosan(6.787 ± 0.274 Mpa) (Sariningsh *et al.,* 2018). The tensile strength of the film from corn starch as 26 Mpa (Mosissa, 2017). By using nano clay as a filler with cassava starch, the tensile strength was improved from 5.2 MPa to 6.3 Mpa (Wahyuningtiyas & Suryanto, 2018). Tensile strength, 8.29 -24.87 N/mm^2 tensile strength of plastic from waste paper also reported (Ezeoha & Ezenwanne, 2013). Tensile strength also determined as 6.49Mpa at 35% glycerol concentration from wheat flour

(Leblanc, 2012). Elongation at the break result obtained under the optimal condition was determined as 5.46%. Zerihun *et al., (*2016) reported as 8.69% elongation for antimicrobial plastic film. Mosissa, (2017) also, reported as 4.32% elongation for the bioplastic developed from corn starch via bagasse cellulosic fiber. The water uptake for the film at the optimal condition was determined as 18.6%. Water absorption also obtained as 18.54%. Water absorption of the film as 20.45% (Mosissa, 2017). Tensile strength was determined as 0.039Mpa, and 6.5 to 20% EA at 10 to 30% glycerol concentration (Lutfi *et al.*, 2017). Further comparison is found in the appendix part Appendix (I).

4.6. Physico-chemical characterization of the bioplastics film

Water solubility test: The film's solubility in water was determined according to the method reported (Kuorwel *et al.*, 2011). Starch is highly soluble in water when fiber reinforced the solubility decrease. Starch, being a hydrophilic polymer, shows a high affinity towards water. Hence, upon hydration, starch-films absorb water and swell. The film with fiber had a lower swelling capacity than the films with only containing starch. Therefore, fiber- starch films showed a lower swelling capacity than only starch-based film. The result shows as the solubility of the control film were 45% and the film blend with fiber shows as 40% solubility.

Density test: The density of the starch-based films was measured for both control and matrix. It was observed that the possibility to increase the density by blending fiber.

The density increment is due to the compatibility of the extracted fiber. The obtained result was 1.65g/ml for the control film and 1.72g/ml for the blended with fiber film.

Transparency of produced bioplastics: The transparencies of the films are determined using a spectrophotometer (UV 7804C) for both control and for the film obtained at the optimum conditions.

The percentage of transmittance of banana peel starch film and the matrix film at a wavelength of 600 nm was determined. The results showed that the films were off white in color. The film without fiber had a higher percentage of transmittance than the blended film. 116% transmittance for the control film and 103% transmittance were obtained for the blended film. The addition of fiber increased film cloudiness for starch-based films. Because the fiber is cloudier than the banana peels starch so the transparency decreases from 116% for starch to 103% for matrix.

This indicates that starch-fiber films being inherently cloudier than starch-based composite films. The decrease in transparency values was observed due to the fiber incorporation. Transparency of the films is of importance in some instances when used as packaging materials (Yehuala & Emire, 2013). The addition of fiber generally causes the films to reduce their transparency, but the small differences compared to control. A starch composite film without fiber was rather transparent.

Moisture: Since starch is highly hydrophilic it absorbs more moisture than the film obtained by blending fiber. The moisture percentage for the pure starch film is higher than the blended. The result obtained was 9.56% for the control and 9.21% for the blended plastic film.

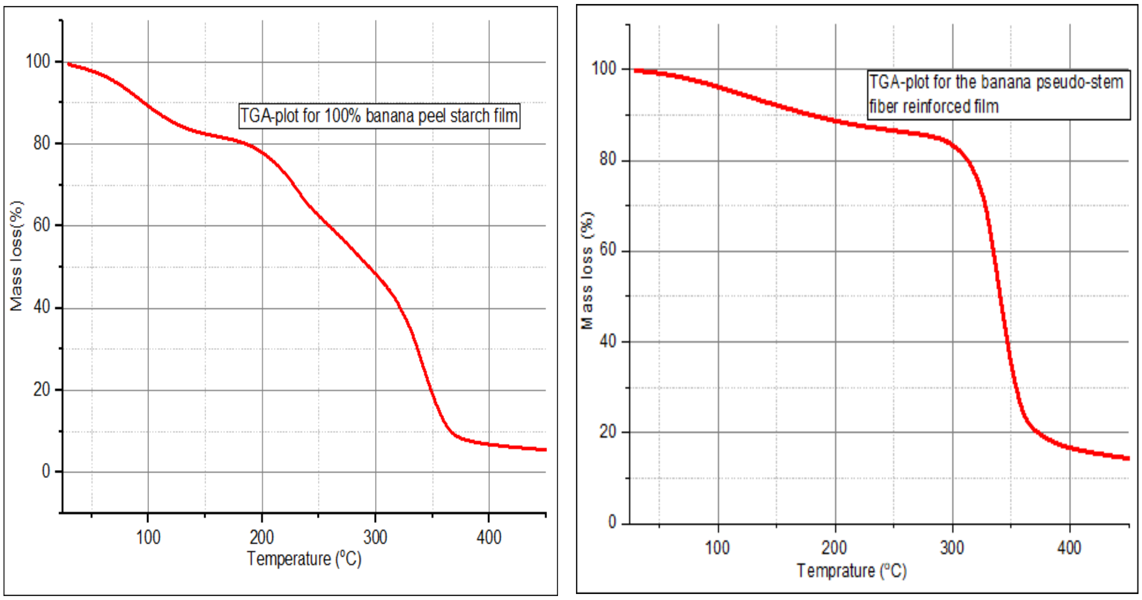
The thickness of films: A digital micrometer was used to measure the film thickness. Since 40 mL hot solution was put into a Petri dish for the control and blended, glycerol and to some amount of starch might vaporize during gelatinization and oven drying the film, comparing the two samples (control and blended) the film synthesized from only starch had small thickness difference than with fiber.

The reason is more starch and glycerol vaporizes during oven drying than for the film synthesized by blending. The obtained thickness was 0.41 mm for control and 0.42 mm for the blended film by taking the average of the triplicates.

Table: 4. 16. Results on some physicochemical properties for both reinforced and control films.

Product	Thickness(mm)	Moisture(%)	Transparency(%)	Solubility(%)	Density(g/ml)
Control	0.41	9.56	116	45	1.65
Reinforced	0.42	9.21	103	40	1.72

Thermal analysis of films using TGA is a technique measuring the variation in the mass of a sample as a function of time or temperature when it undergoes temperature scanning in a controlled, inert or oxidative atmosphere. As shown in the Figure 4.16, the weight loss for the controlled film increase more rapidly than the reinforced bioplastic film. This indicates that the incorporation of highly thermo resistant cellulosic fiber which is extracted from the pseudo-stem is best option to improve the thermal stability of the film.Since starch is more hydrophyilc than fiber initially, low voletile components are viporize in the controlled film rapidly than the reinforced film. This overheating generally implies variations to the molecular weight (and molecular weight distribution) of the polymer and typical property changes include minimize ductility and embrittlement, chalking, color deviations, cracking, a general reduction in most other desirable physical properties of the film. The largest mass loss was observed within a hgher temperature, this is mainly because of the fact that at high temperatures the components of the long-chain backbone of the film can begin to be broken (chain scission) and react with one another to change their properties.



A **B**

Figure: 4.16. Mass loss by using TGA for the controlled (A) and reinforced films (B)

Mass loss for both controlled and reinforced film at a specific temperature was taken as a comparasion (Appendix g). Half mass of the controlld film was lost at 291°C, and the reinforced film also lost at 340 °C. The final total mass residue or ash for controlled film was 4.93% and also for reinforced film the total ash was 13.4% at 450°C. The derivative mass loss also tells that the controlled film decompose slightly at 75.3 °C, highly decompose at 225.16°C, and critically decompose at 336°C. This film was sensitive to heat, and highly affected by temperature. The reinforced film obtained by the incorporation of 18% fiber on the otherhand is slightly affected by temperature, a small decomposition was observed at 100°C, and large decomposion was shown at 340°C.

DSC analysis of the reinforced film: DSC determines the variation in the heat flow given out or taken in by a sample when it undergoes temperature scanning in a controlled atmosphere. With heating or cooling any transformation taking place in a material is accompanied by an exchange of heat; DSC enables the temperature of this transformation to be determined and the heat from it to be quantified.

It enables us to determine the number of parameters such as melting temperature (T_m), crystallization temperature (T_c), glass transition temperature(T_g), degradation temperature(T_d), etc.(Bp, 2009).The Tg, Tc, and Tm were taken from the onset temperatures of endothermic step change, exothermic peak and endothermic peak, respectively. According to DSC analysis, it was noted that a first endothermic peak between the temperature range of 55°C and 58°C representing residual water, which was used during the pre-gelatinization process. The second endothermic peak which was between at a temperature range of 95 °C and 120°C representing residual water evaporation, which was used during the process of molding of gelatinized starch. The last and the third endothermic peak were between 170°C and 175°C representing the total loss of heat due to overheated.

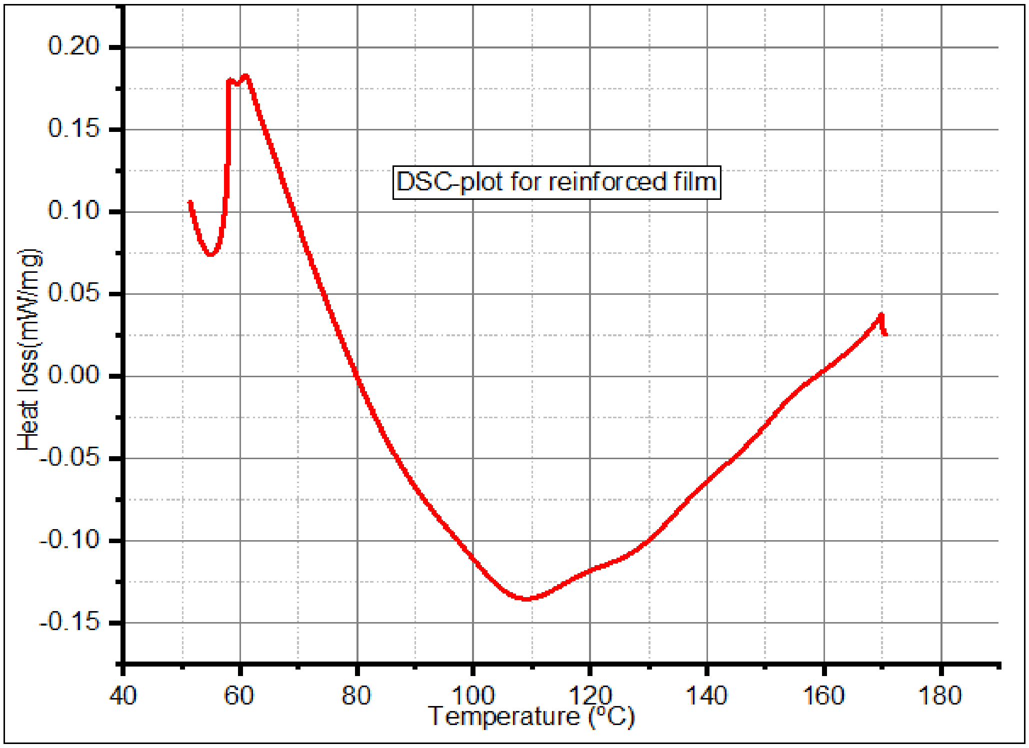


Figure: 4.17. Heat loss by using DSC for the reinforced films

The film containing 18% w/w of fiber concentration presented a higher melting temperature (Tm) which is 110°C and glass transition temperature(Tg) of 53-55°C. The Tm and Tg of the plastic film reported by (Petrovich, 2017) were (110-120 °C) and (50-95°C) respectively.

Degradation temperature(Td) also as shown the figure 4.17 gives 172 °C, the crestal temperature(Tc) also determined as 60^{o}C.

Fourier Transform Infrared Spectrometry (FTIR) Analysis of the film:

The FT-IR spectra of banana peel starch-glycerol based films with cellulosic fiber are shown in Figure 4.18. An absorption band at 3272.20 cm^{-1} for the film appears. An extremely broad peak appeared between 3000-3600 cm^{-1} is the region of hydrogen-bonded hydroxyl groups can be attributed to C-O-H bond stretching due to molecular bound hydroxyl groups (Salleh *et al.,* 2009), broad band at 3272.20 cm^{-1} due to the hydrogen-bonded hydroxyl groups, the sharp band at 2923.43 and 2852.88 cm^{-1} is characteristic of C-H stretches associated with the ring methane hydrogen atoms/ C-H stretching vibrations of aliphatic groups, the bands at 1650.746 and 1416.95 cm^{-1} are assigned to the (O-H) bending of water / adsorbed water signals and CH_2, respectively. C- C and C-O stretching at 1100 cm^{-1} and C-O-H bending vibration at 1007 cm^{-1}. At the peak, a1550.77 cm^{-1}, corresponding to the absorption caused by the bonding C = O (a type of aldehyde, ketone, carboxylic acid, esters) and C = C (a type of alkene compound). At the peak with wavenumbers of 1477.47 cm-1 to 459.06 cm^{-1}, corresponds to absorption caused by the C-H bond (alkane type compound), and C-O (a type of alcohol, carboxylic acid, ester). Therefore, the film developed from starch, glycerol, and fiber has a functional group which is a combination of specific functional groups contained in its constituent components such as C-H, O-H, C-O, C = O, and C = C. Generally from the band 1007-897 cm^{-1} C-O-H bending vibration, 1150-1005 cm^{-1} C-C stretching, 1235-1150 cm^{-1} C-O-C stretching of Easter, 1734-1631 cm^{-1} bounded water, 3000-2826 cm^{-1} C-H stretches and 3600-3000 cm^{-1} -OH stretching due to polymeric involvement of hydroxyl groups and bonded O-H stretching vibration present in starch was observed. A possible explanation for this may be hydrogen bonding between the CH_2-OH groups from the starch and cellulose fiber (Sarifuddin *et al.,* 2012). Having this the fiber, starch and glycerol are attached chemically, this implies that fiber is favored to the starch by forming /reinforced with the starch matrix.

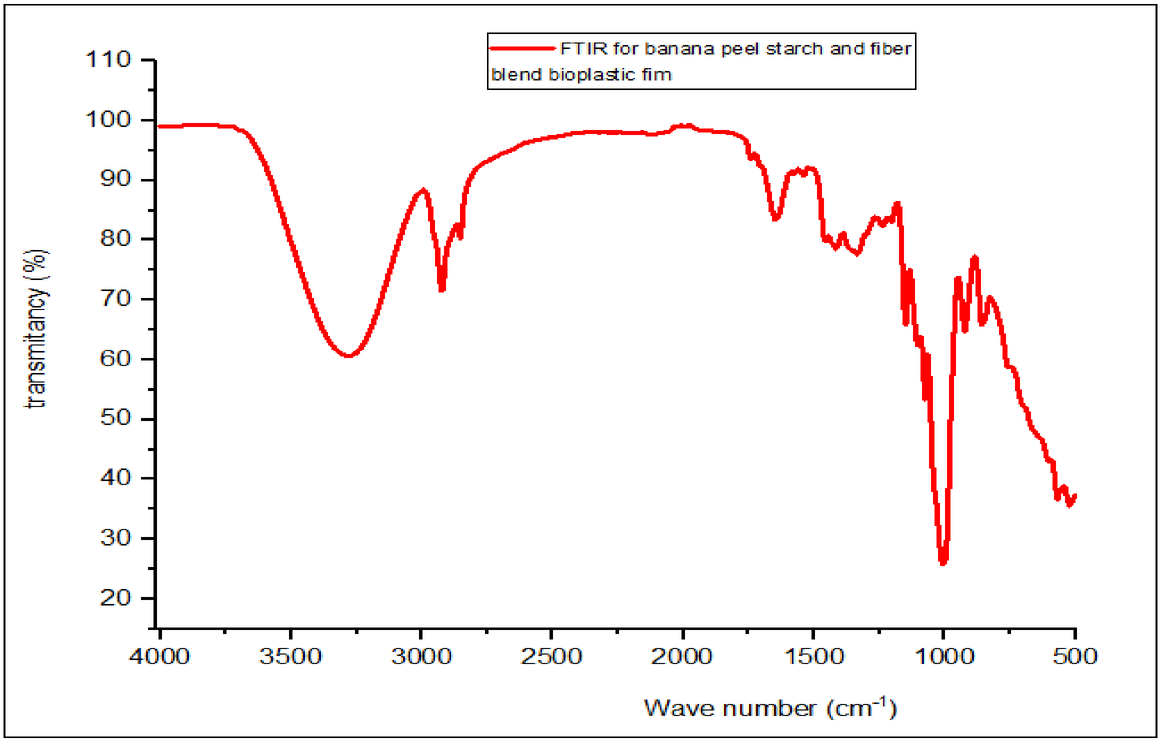


Figure: 4. 18. FTIR result of starch-fiber, bio-plastic film plot

Generally from the band 1007-897 cm^{-1} C-O-H bending vibration, 1150-1005 cm^{-1} C-C stretching, 1235-1150 cm^{-1} C-O-C stretching of easter, 1734-1631 cm^{-1} bounded water, 3000-2826 cm^{-1} C-H stretches and 3600-3000 cm^{-1} -OH stretching due to polymeric involvement of hydroxyl groups and bonded O-H stretching vibration present in starch was observed. A possible explanation for this may be hydrogen bonding between the CH_2-OH groups from the starch and cellulose fiber (Sarifuddin *et al.*, 2012). Having this the fiber, starch and glycerol are attached chemically, this implies that fiber is favored to the starch by forming /reinforced with the starch matrix.

5. CONCLUSION AND RECOMMENDATION

5.1. Conclusion

In this research, the development of bioplastic film from banana peel starch and banana pseudostem fiber was investigated. This work also intended to study the proximate composition of the banana peel, the influence of starch extraction parameters: extraction time and extraction temperature on the starch yield and characterization of starch properties. On the basis of these results, it can be concluded that banana peel should be considered suitable for bioplastic synthesis because the proximate composition shows that values of 10.5% moisture, 8.5% total ash, 6.66% crude lipid, 2.17% crude protein, 34.37% crude fiber and 37.8% nitrogen-free extract. This raw material is suitable for bioplastic synthesis, mainly because of its relatively higher carbohydrate, which is the main source of starch. Banana pseudostem fiber was extracted and the characterization shows high dense, low water uptake capacity, higher cellulose content, and higher thermal resistance capability when analyzed by TGA.

During the synthesis of bioplastics film, the effects of independent variables (dry oven temperature, glycerol, and fiber concentration) on tensile strength (TS), elongation at the break (EA) and water absorption (WA) using CCD response surface experimental design was studied. From the conducted experiment, the recorded result shows the minimum EA was obtained at higher drying temperature and fiber concentration but at a low level of glycerol concentrations. Minimum WA also founded at higher drying temperature and at medium fiber and glycerol concentrations.

This result also indicates that maximum (WA, EA), and minimum TS were obtained at the possible combination of both minimum dry oven temperature and fiber concentrations and at maximum glycerol concentrations. Higher TS strength was obtained at the combination of all medium-dry oven temperatures, glycerol and fiber concentrations.

The interaction of both drying temperature and glycerol had a significant effect on TS and EA. The significant effect has been observed on the TS and WA due to the interaction of drying temperature and fiber concentrations. Both fiber and glycerol interactions also significantly affect the EA of the film.

The optimized conditions that have been considered had high tensile strength, low elongation, low water absorption is chosen using numerical optimization as a combination of 56.5^{o}C dry oven temperature, 28.6% glycerol and 18.26% fiber concentration to obtain a good quality plastic film (7.23 Mpa TS, 5.46 % EA and 18.60% WA of the film). The addition of cellulosic fiber enhances both the physicochemical and thermal properties of the film. Cellulosic fiber enhances TS but lowers both EA and WA (Sarifuddin *et al.*, 2012) and (Prachayawarakorn *et al.*, 2011).

Using the optimized plastic film, the thickness, solubility, moisture, density, and transparency were determined for both the controlled and the reinforced with fiber films. The addition of 18% fiber indicates that a better bioplastic film with a better physicochemical property when compared to the control film. The TGA result shows that the mass loss of the reinforced film is less than the controlled film this is due to the incorporation of thermal resistant cellulosic fiber. The best plastic film synthesized at the optimum operating condition was also characterized by FTIR shows as the fiber and starch well interacts chemically. Finally, the overall results showed that the banana peel starch and banana pseudostem fiber has a promising potential to be used in combination with glycerol plasticizer in bioplastic film synthesis for dry good packaging applications.

5.2. Recommendations

- For more research works on the extraction of the banana pseudostem fiber, the further treatment process is better in order to get good quality as well as quantity of fibers used as a filler.
- In this research, only the effect of three independent variables was studied by holding the other variables constant for film synthesis. One can also do investigate the other factors effect like the particle size of banana peel starch and pseudostem fiber, stirring speed, gelatinization temperature, and try other plasticizer (sorbitol and polyvinyl alcohol).
- Banana peel and banana pseudostem that has been used for the study was collected from a specified part of our country, but the mineral content of the soil will have an impact on the composition, on the yield of starch and fiber. Therefore one can do investigate the effects by collecting the banana peel and pseudostem from different parts of the country.
- It is advisable to incorporate the antimicrobial inhibitor to enhance the shelf life of the good to be packed.

REFERENCES

Abubakar US, B. D., Yusuf KM. (2016). Proximate and mineral composition of corn cob, banana and plantain peels. International Journal of food science and nutrition,1,25–27.

Adeyefa, O. S. (2015).Biodegradable starch film from cassava, corn, potato, and yam. Chemistry and Materials Research,7(12), 15–24.

Al, R., Sujuthi, F. M., & Liew, K. C. (2016). Properties of bioplastic sheets made from different types of starch incorporated with recycled newspaper pulp. Journal of Food Technology, 3, 257–264.

Alcázar-alay, S. C., Angela, M., & Meireles, A. (2015).Physicochemical properties, modifications and applications of starches from different sources. Journal of Food Science and Technology, 35(2), 215–236.

Alemu, D. (2016). Banana markets in Ethiopia.Journal of biology, agriculture, and healthcare, vol.4, no.23.

Alemu, K. (2014). Importance and pathogen spectrum of crown rot of banana Journal of Biology, Agriculture and Healthcare, 4(23), 106–112.

Alobi, N. O., Sunday, E. A., Magu, T. O., Oloko, G. O., & Nyong, B. E. (2017). Analysis of starch from non- edible root and tubers. Food Science and Technologies, 2, 27–32.

Altemimi, A. B. (2018). Extraction and optimization of potato starch and its application as a stabilizer in yogurt manufacturing. Journal of Food Science,1-11.

Amri, A., Ekawati, L., Agustin, Y. E., Padmawijaya, K. S., & Kartika, T. (2018). Mechanical properties of bioplastics cassava starch film with Zinc Oxide nanofiller as reinforcement.IOP Conf. Series: Materials Science and Engineering 210 (2018) 012015

Anhwange, B., & Ugye, J. T. (2014). Chemical composition of Musa sepientum peels. Journal of Environmental, Agricultural and Food Chemistry, 8 (6), 437-442.

Arikan, E. B., & Ozsoy, H. D. (2015). A Review: an investigation of bioplastics. Journal of Civil Engineering and Architecture, 9, 188–192.

Been, B., Chiang, H., & Chu, W. C. (1987). A Pilot-scale study for banana starch production, 8(1984), 6–9.

Bp, M. (2009). Chapter 2 2 Production of biodegradable plastic film, 14–38.

Bickford, R. (2018). Ethiopia fresh fruits market update report.Global agricultural information network volunteer-public report

Catálise, L. De, Universidade, A., & Avenida, F. (2017). Bioplastics production from starch and chitosan blends.Journal of polymers, 9, 46–53.

Ebisike, K., Attahdaniel, E., Babatope, B., & Olusunle, S. O. O. (2013).Studies on the extraction of naturally-occurring banana fibers.International Journal of Engineering Science, Volume 2, 9 (95-99).

Ebnesajjad, S. (2017). Handbook of biopolymers and biodegradable plastics properties, processing.

Ezeoha, S. L., & Ezenwanne, J. N. (2013). Production of Biodegradable Plastic Packaging Film from Cassava Starch, *3*(10), 14–20.

Fatimah, N., Sultan, K., Lutfi, W., & Johari, W. (2017).The development of banana peel/corn starch bioplastic film : a preliminary study. Bio-remediation science and technology research, 5(1), 12–17.

Gadhave, R. V, Das, A., Mahanwar, P. A., & Gadekar, P. T. (2018). Starch-based bioplastics : the future of sustainable packaging. Journal of Polymer Chemistry, (8),21–33.

Ghuttora, N. (2016). Increase the usage of biopolymers and biodegradable polymers for a sustainable environment, 7(4), 6-40.

Gopinathan, P., Subramanian, K. S., Paliyath, G., & Subramanian, J. (2017) Genotypic variations in characteristics of nano- fibrillated cellulose derived from banana pseudostem. Journal of bioresource,12, 6984–7001.

Gozan, M., & Noviasari, C. (2018). The effect of glycerol addition as a plasticizer in bioplastics. 67,11–14.

Guadalupe, M., Ramírez, L., Satyanarayana, K. G., Iwakiri, S., Bolzon, G., Muniz, D., Flores-sahagun, T. S. (2011). Study of the properties of biocomposite. Carbohydrate Polymers, 86, 1712–1722.

Gujar, S., Pandel, B., & Jethoo, A. S. (2014). Effect of plasticizer on mechanical and moisture absorption properties. An International Quarterly Scientific Journal.13(2), 425-428.

Hadisoewignyo, L., Foe, K. & Tjandrawinata, R.R. 1. (2017).Isolation and characterization of Agung banana peel starch from east java Indonesia. International Food Research Journal 2, 24(June), 1324–1330.

Halley, P., Coombs, S., Kettels, J., Christie, G., Jenkins, M., Beh, H., Polymers, S. (2001).Developing biodegradable mulch films from starch-based polymers. Research paper, 362–367.

John, M. J., & Thomas, S. (2008).Bio fibers and biocomposites. Carbohydrate Polymers 71, 343–364.

Joshi, G., & Sarangi, M. K. (2014).Review article a review on banana starch. Inventi Rapid: planta activa,1-8.

Kalia, S., Kaith, B. S., & Kaur, I. (2009). Pretreatments of natural fibers and their application as reinforcing material in polymer composites a review. Article in Polymer Engineering and Science, 1-34.

Kassaw (2018). Addis Ababa institute of technology school of chemical and bioengineering production and characterization of pulp from banana pseudostem via soda anthraquinone pulping process for papermaking

Kattas, L., Gastrock, F., Levin, I., & Cacciatore, A. (2018). Book of Plastic additives chapter 4.

Keziah, V. S., Gayathri, R., & Priya, V. V. (2018). Biodegradable plastic production from corn starch. Research articles, 10(7), 1315–1317.

Khazaei, N., Esmaiili, M., Emam, Z., & Ghasemlou, M. (2014). Characterization of the new biodegradable edible film made from basil seed (Ocimum basilicum.) gum. carbohydrate polymers, 102, 199–206.

Kumar, A., Singh, B. P., Jain, R. K., & Sharma, A. K. (2013). Banana fiber (Musa sapientum): A suitable raw material for handmade paper industry via enzymatic refining. International Journal of Engineering Research & Technology (IJERT), 2(10), 1338–1350.

Kuorwel, K. (2011). Incorporation of Natural Antimicrobial Agents into Starch-Based Material for Food Packaging. School of Engineering and Science Faculty of Health, Engineering and Science Victoria University

Leblanc, N. (2012). Effect of glycerol on the properties of 100 % biodegradable thermoplastic based on wheat flour effect of glycerol on the properties.Journal of polymer and environment, 2 (21), 388-394.

Li, Z., Guo, K., Lin, L., He, W., & Zhang, L. (2018). Comparison of physicochemical properties of starches from flesh and peel of green banana fruit. Articles on molecules, 1–15.

Lubis, M., & Harahap, M. B. (2018). Production of bioplastic from avocado seed starch reinforced with microcrystalline cellulose from sugar palm fibers. Journal of Engineering Science and Technology, 13(2), 381–393.

Luengo, M. Ã., & Ã, A. S. (2003). Bioplastics from microorganisms. Article in Current Opinion in Microbiology, 6, 251-260.

Lutfi, M., Sumarlan, S. H., Susilo, B., Zenata, R., & Perdana, L. P. R. (2017).The Glycerol effect on the mechanical behavior of biodegradable plastics.Environment and Pollution Technology on an International Quarterly Scientific Journal, Vol. 16 No. 4 pp. 1121-1124.

Ma, H., & Deng, M. (2017). An optimized procedure for determining the amylase/amylopectin ratio in common wheat grains based on the dual-wavelength iodine-binding method. Journal of Polymer & Composites

1(1), 23–30.

Mardhiah, A., Amin, M., Sauid, S. M., Halim, K., & Hamid, K. (2017). The effect of glycerol content on mechanical properties, surface morphology and water absorption of thermoplastic films. Jurnal of technology,79, 5–3

Maekawa, M. M. (2016). Fabrication and characterization of mesoporous silica nanochannels inside the channels of the anodic alumina membrane. Arabian journal of chemistry, 9(2), 269–273.

Mistry, A. H., Schmidt, S. J., & Eckhoff, S. R. (1992). Alkali extraction of starch from corn flour, 44(8), 284–288.

Mohiuddin, A. K. M., Saha, M. K., Hossian, S., & Ferdoushi, A. (2014). The usefulness of banana (Musa paradisiaca) wastes in the manufacturing of bioproducts : A Scientific Journal of Krishi Foundation, 12, 148–158.

Mohsen Esmaeili, Gholamreza Pircheraghi, R. B. (2018.).Optimizing mechanical and physical properties of thermoplastic starch through co-plasticization by sorbitol and glycerol, 33(6), 123-133.

Mosissa .G. (2018). Addis Ababa institute of technology school of chemical and bioengineering, Production and characterization of bioplastics from corn starch reinforced with sugar cane bagasse.

Muche, M. (2018) Production and Characterization of Starch-Based Antimicrobial Packaging Film thesis done from Addis Ababa university

Mukhopadhyay, R., Sree, K. D., Saneeha, R., Kale, P., & Iram, U. (2017). Preparation and characterization of biodegradable plastics. International Journal for Research in Applied Science & Engineering Technology (IJRASET), 5(8), 134–142.

Naranjo, J. M., Cardona, C. A., & Higuita, J. C. (2014). Use of residual banana for polyhydroxybutyrate (PHB) production : the case of study in an integrated biorefinery. Journal of waste management, 34(12), 2634–2640.

Offiong, E. U., & Ayodele, S. L. (2016). Preparation and characterization of thermoplastic starch from sweet potato. International Journal of Scientific & Engineering Research, 7(5), 438–443.

Petrovich, J. (2015). FTIR and DSC of polymer films used for packaging , 1–13.

Prachayawarakorn, J., & Ruttanabus, P. (2011). Effect of cotton fiber contents and lengths on properties of thermoplastic starch composites prepared from rice starches. Journal of polymer environment, 274–282.

Priedniece, V., Spalvins, K., Ivanovs, K., & Pubule, J. (2017). a review of bioproducts from potatoes. Environmental and Climate Technologies, 21, 18–27. https://doi.org/10.1515/rtuect-2017-0013

Puppala, S., M, S. R., & B, R. A. (2012). Seaweeds can be a new source for bioplastics. Journal of Pharmacy Research, 5(3), 1476–1479.

Pyar, H., & Peh, K. K. (2018). Chemical compositions of banana peels (Musa sapientum) fruit cultivated in Malaysia. Research Journal of Chemistry and Environment, 22(2), 108–113.

Saad, M. (2016). Data in brief nano-cellulose derived bioplastic biomaterial data for vehicle bio-bumper from banana peel waste biomass. Journal Data in Brief, 8, 286–294. https://doi.org/10.1016/j.dib.2016.05.029

Sagnelli, D., Kirkensgaard, J. J. K., Valeria, C., & Giosafatto, L. (2017). all-natural bio-plastics using starch-beta-glucan composites. Journal of carbohydrate polymers, 172, 237–245.

Salleh, E., Muhamad, I. I., & Khairuddin, N. (2009). Structural characterization and physical properties of antimicrobial starch-based films. International Journal of Biomedical and Biological Engineering, 3(7), 352–360.

Sanyang, M. L., Sapuan, S. M., Jawaid, M., Ishak, M. R., & Sahari, J. (2015). Effect of plasticizer type and concentration on tensile, thermal and barrier properties of biofilm.Journal of polymers,*7*, 1106-1124.

Sarifuddin, N., Ismail, H., & Ahmad, Z. (2012). Thermoplastic sago starch/kenaf core fiber.Article in Bioresources, 7(3), 4294-4306.

Schon, M., & Schwartz, P. (2018.). Production of bioplastic abstract what is bioplastics, potato starch 1–5.

S. Das, M. Rahman and M. (2018).physicomechanical properties of pineapple leaf and banana fiber reinforced hybrid polypropylene composites. IOP Conference Series: 438 (2018) 012027.

Sariningsih, N., Putra, Y. P., Pamungkas, W. P., Agustin, Y. E., & Padmawijaya, K. S. (2018). Bioplastic from chitosan and yellow pumpkin starch with castor oil. IOP Conf. Series: Materials Science and Engineering, 333 (2018) 012087.

Shuying Xu, Chunrong X. Wei T. & Yucang Z. (2015) Microstructural, thermal, and tensile characterization of banana pseudo-stem fibers. Journal of bioresource. 10(2), 3724–3735.

Talja, R. (2007). Preparation and characterization of potato starch films plasticized with polyols.

Tegangan, S., & Air, P. (2011). Tensile and water absorption properties of biodegradable composites derived from cassava skin / polyvinyl alcohol with glycerol as a plasticizer, journal of biotechnology. 40(7), 713–718.

Teli, M. D., & Terega, J. M. (2017). Chemical, physical and thermal characterization of Ensete ventricosum plant fiber. International Research Journal of Engineering and Technology, 67–75.

Uçkun, E., Trzcinski, A. P., Jern, W., & Liu, Y. (2014). Bioconversion of food waste to energy: a review. Journal of fuel,134, 389–399.

Usman, M., Ishfaq, M. T., Malik, S. R., Ishfaq, B., & Iqbal, M. (2017).Effects of temperature, ph and steeping time on the extraction of starch from Pakistani rice.Journal of international agro physics, 31, 377-382.

Vega-castro, O., Contreras-calderon, J., León, E., & Sobral, P. J. A. (2016). Characterization of a polyhydroxyalkanoate obtained from pineapple peel waste. Journal of biotechnology,231, 232–238.

Victo, M., Grossmann, E., Alves, V. D., & Bele, A. (2007). Effect of glycerol and amylose enrichment on cassava starch film properties, 78, 941–946.

Vigneswaran, C., Pavithra, V., Gayathri, V., & Mythili, K. (2015). C. Vi Department of fashion technology, chemical properties which can be used as a very good raw material for the textile and packaging industry. Journal of textile and apparel technology and management, 9(2), 1–7.

Wahyuningtiyas, N. E., & Suryanto, H. (2017). Analysis of biodegradation of bioplastics made of cassava starch.Bioremediation science and technology research, Vol 5, No 1, 12-17

Wahyuningtiyas, N. E., & Suryanto, H.(2018). Properties of cassava starch-based bioplastics reinforced by nanoclay. Iranian Journal of Science and Technology, Vol. 1 No. 1, 2580-081.

Wittaya, T.(2012). Rice starch-based biodegradable films: properties enhancement.Chapter 5, http://dx.doi.org/10.5772/47751.

Wojciechowska, P. (2014). The effect of concentration and type of plasticizer on the mechanical properties of cellulose acetate butyrate organic-inorganic hybrids, 8, 235-344.

Yehuala, G. A., & Emire, S. A. (2013). Antimicrobial activity, physicochemical and mechanical properties of aloe (aloe debrand) based packaging films**.** British Journal of Applied Science & Technology, 3(4), 1257–1275.

Yue, H., Cui, Y., Yin, G., & Jia, Z. (2014). Preparation and environment-friendly cottonseed protein bioplastics. Porous Organic Polymers, 311-313.1851

Yue, H., Shuttleworth, P. S., National, S., & Clark, J. H. (2012). Green chemistry preparation and characterization of bioplastics made from cottonseed. Journal of Royal Society of Chemistry, 14-12-36.

Zerihun, M., Worku, T., & Sakkalkar, S. R. (2016). Development and characterization of antimicrobial packaging films. Journal of ready to eat food,03(02), 13–24.

APPENDIXES

Appendix A: Supporting pictures during the study

Figure A$_1$: glycerol and sodium hydroxide


Soaking the peel


Blended peel


Heading and stirring

a


Filtration

b


Starch sedimentation

c

Figure A_2: starch extraction/sample pictures

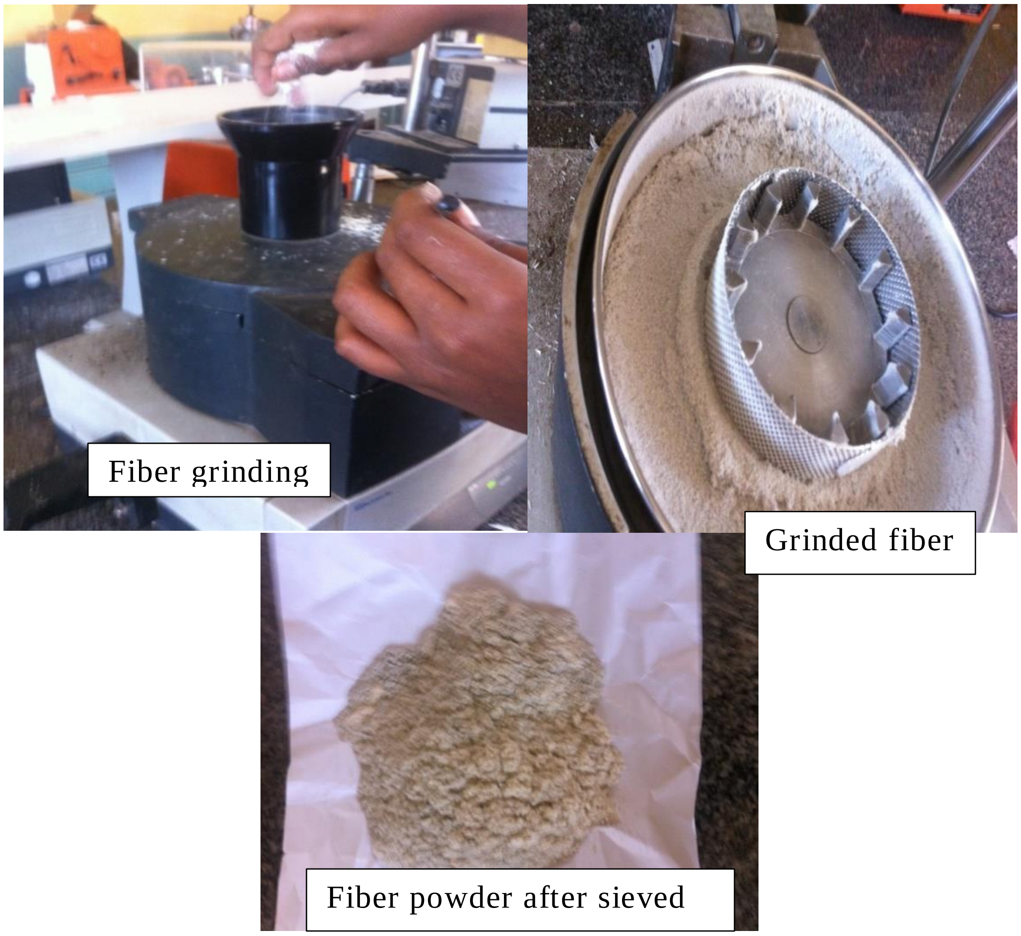


Figure A$_3$: Fiber preparation

a

b

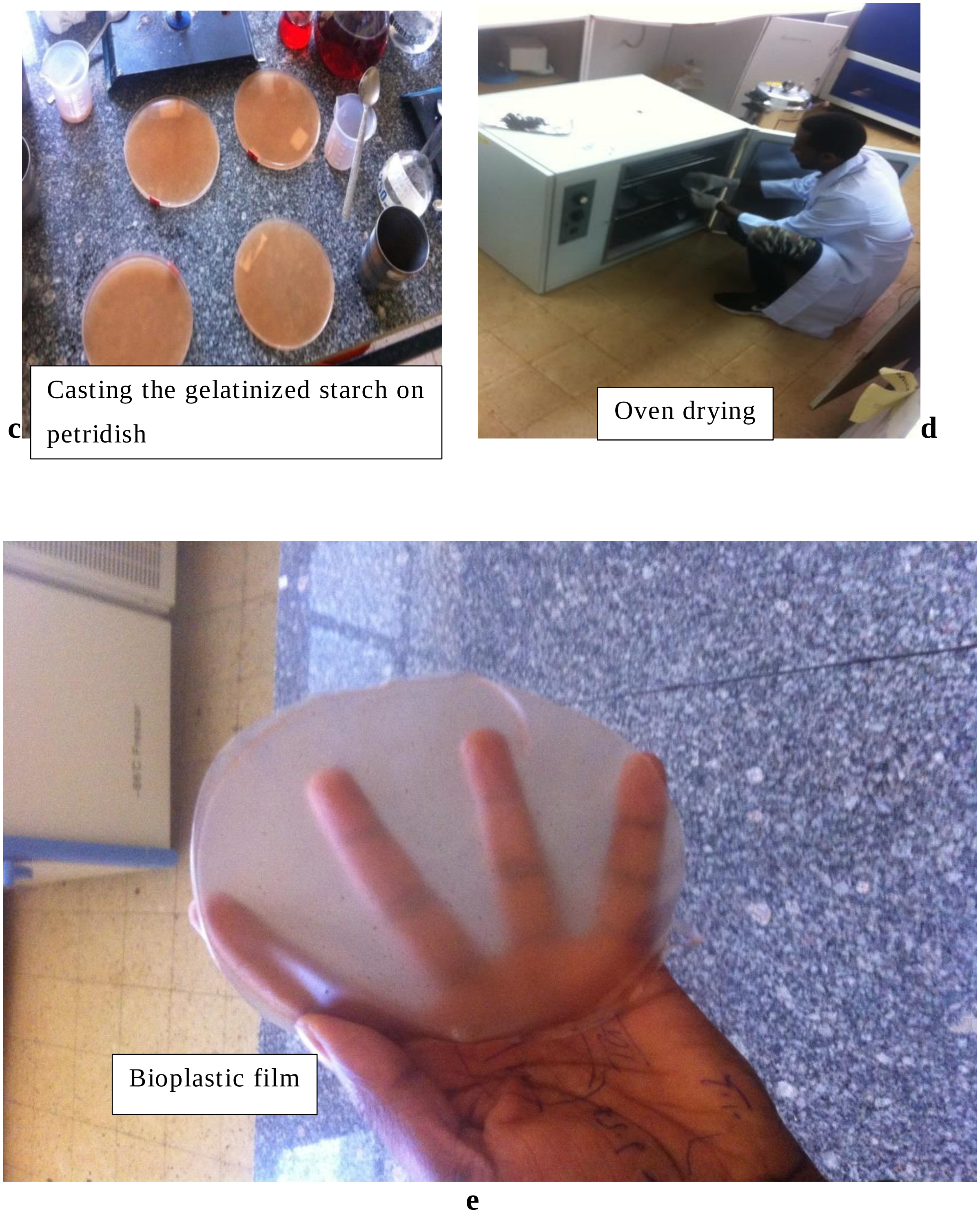


Figure A_4: Bioplastic film synthesis / sample pictures (a-e)

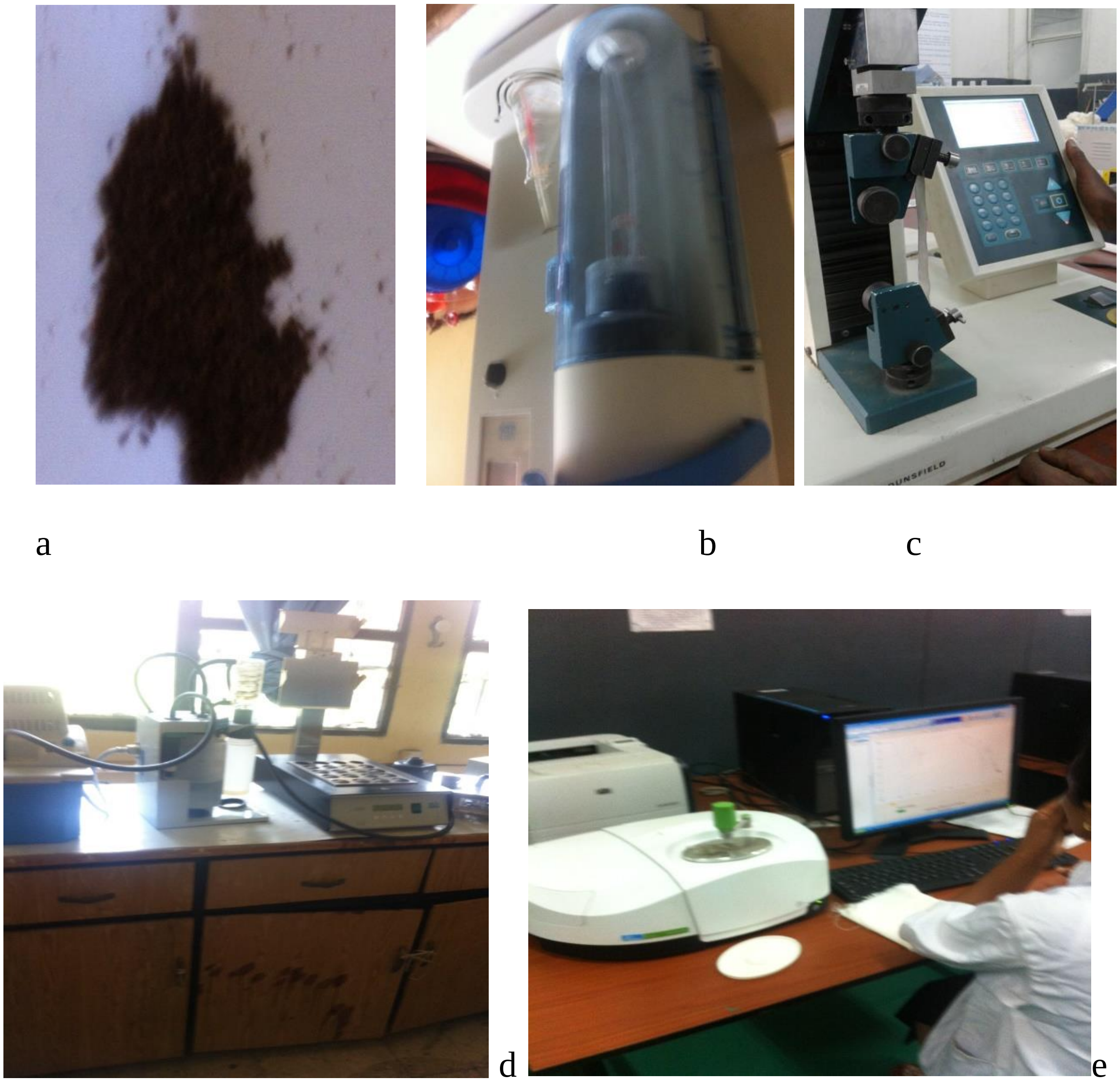


Figure A$_5$: sample taken for the banana peel for proximate analysis (a), condenser (b) and ASTM for tensile strength/tensile strength taster machine (c), Kjeldah (d), FTIR (e)

Appendix B: design expert data

Table B$_1$: Analysis of variance [Partial sum of squares], for elongation at the break

Source	Sum of Squares	Mean Square	F-Value	Prob > F	
Model	863.82	95.98	182.10	< 0.0001	Significant
A-oven dry temp.	319.34	319.34	605.89	< 0.0001	
B-gly.conc.	347.04	347.04	658.45	< 0.0001	
C-FB	36.90	36.90	70.02	< 0.0001	
AB	8.84	8.84	16.77	0.0022	
AC	0.1770	0.1770	0.3359	0.5751	
BC	10.15	10.15	19.25	0.0014	
A^2	38.59	38.59	73.21	< 0.0001	
B^2	6.57	6.57	12.47	0.0054	
C^2	1.33	1.33	2.53	0.1430	
Residual	5.27	0.5271			
Lack of Fit	3.48	0.6951	1.94	0.2429	not significant
Pure Error	1.80	0.3590			
Cor Total	869.09				

Table B_2: Analysis of variance [Partial sum of squares], for water absorption

Source	Sum of square	Mean square	F – value	Prob>F	
Model	403.76	44.86	159.97	< 0.0001	Significant
A-oven dry temp.	91.26	91.26	325.43	< 0.0001	
B-gly.conc.	66.00	66.00	235.34	< 0.0001	
C-FB	23.13	23.13	82.49	< 0.0001	
AB	0.0120	0.0120	0.0428	0.8402	
AC	4.49	4.49	15.99	0.0025	
BC	0.0091	0.0091	0.0325	0.8605	
A^2	10.64	10.64	37.93	0.0001	
B^2	27.58	27.58	98.34	< 0.0001	
C^2	18.12	18.12	64.61	< 0.0001	
Residual	2.80	0.2804			
Lack of Fit	0.8284	0.1657	0.4192	0.8190	not significant
Pure Error	1.98	0.3952			
Cor Total	406.57				

Table B_3: Model adequacy measures for elongation at the break

Std. Dev.	0.73	R^2	0.9939
Mean	12.99	Adjusted R^2	0.9885
C.V.	5.59	Predicted R^2	0.9529
PRESS	40.90	Adeq Precision	52.4515

Table B_4: Model adequacy measures for water absorption

Std. Dev.	0.5296	R^2	0.9931
Mean	23.30	Adjusted R^2	0.9869
C.V. %	2.27	Predicted R^2	0.9789
PRESS	8.57	Adeq Precision	44.5626

Table B_5: Diagnostics Case Statistics for TS

Standard Order	Actual value	Predicted value	Residual	Leverage	Student Residual	Cook's distance	Outlier T	Run order
1	5.70	5.80	-0.10	0.491	-0.834	0.067	-0.820	15
2	7.21	6.98	0.23	0.118	1.400	0.026	1.482	2
3	3.90	4.08	-0.18	0.491	-1.430	0.197	-1.522	10
4	5.01	4.99	0.019	0.793	0.239	0.022	0.227	3
5	1.05	1.04	0.013	0.793	0.163	0.010	0.155	16
6	2.86	2.82	0.045	0.793	0.567	0.123	0.547	17
7	3.02	3.15	-0.13	0.491	-1.027	0.102	-1.031	7
8	3.80	3.80	1.864	0.793	0.024	0.000	0.022	6
9	7.01	6.98	0.029	0.118	0.175	0.000	0.167	14
10	5.55	5.60	-0.053	0.491	-0.431	0.018	-0.413	13
11	0.20	0.16	0.039	0.793	0.491	0.093	0.472	19
12	7.18	6.98	0.20	0.118	1.217	0.020	1.250	1
13	6.90	6.98	-0.081	0.118	-0.498	0.003	-0.479	9
14	2.02	1.98	0.038	0.793	0.479	0.088	0.460	12
15	6.90	7.04	-0.14	0.491	-1.124	0.122	-1.141	8
16	6.30	6.39	-0.091	0.491	-0.737	0.052	-0.719	18
17	6.80	6.98	-0.18	0.118	-1.111	0.017	-1.125	11
18	0.32	0.26	0.056	0.793	0.706	0.191	0.688	4
19	1.52	1.50	0.020	0.793	0.251	0.024	0.239	5
20	7.25	6.98	0.27	0.118	1.645	0.036	1.828	20

Table B_6: Diagnostics Case Statistics for EA

Standard Order	Actual value	Predicted value	Residual	Leverage	Student residual	Cook's distance	Outlier T	Run order
1	7.80	8.09	-0.29	0.491	-0.553	0.030	-0.533	15
2	9.60	9.99	-0.39	0.118	-0.574	0.004	-0.554	2
3	17.00	17.43	-0.43	0.491	-0.827	0.066	-0.813	10
4	4.90	4.55	0.35	0.793	1.075	0.443	1.084	3
5	15.20	15.33	-0.13	0.793	-0.406	0.063	-0.389	16
6	12.00	11.97	0.028	0.793	0.084	0.003	0.080	17
7	20.10	19.39	0.71	0.491	1.373	0.182	1.447	7
8	5.90	6.43	-0.53	0.793	-1.612	0.996	-1.777	6
9	9.40	9.99	-0.59	0.118	-0.868	0.010	-0.856	14
10	6.50	5.65	0.85	0.491	1.648	0.262	1.831	13
11	31.01	31.47	-0.46	0.793	-1.397	0.748	-1.477	19
12	9.50	9.99	-0.49	0.118	-0.721	0.007	-0.703	1
13	9.60	9.99	-0.39	0.118	-0.574	0.004	-0.554	9
14	18.80	18.36	0.44	0.793	1.320	0.668	1.378	12
15	8.50	8.77	-0.27	0.491	-0.515	0.026	-0.495	8
16	13.30	12.61	0.69	0.491	1.335	0.172	1.397	18
17	11.00	9.99	1.01	0.118	1.479	0.029	1.588	11
18	26.10	25.67	0.43	0.793	1.290	0.638	1.340	4
19	13.50	14.04	-0.54	0.793	-1.642	1.034	-1.823	5
20	10.00	9.99	8.364E-003	0.118	0.012	0.000	0.012	20

Table B_7: Diagnostics Case Statistics for WA

Standard Order	Actual Value	Predicted value	Residual	Leverage	Student residual	Cook's distance	Outlier T	Run order
1	18.60	18.39	0.21	0.491	0.553	0.029	0.533	15
2	20.10	19.45	0.65	0.118	1.317	0.023	1.374	2
3	25.00	25.18	-0.18	0.491	-0.479	0.022	-0.460	10
4	20.69	20.86	-0.17	0.793	-0.689	0.182	-0.670	3
5	30.00	29.79	0.21	0.793	0.851	0.278	0.839	16
6	25.70	25.85	-0.15	0.793	-0.619	0.147	-0.598	17
7	24.00	24.43	-0.43	0.491	-1.146	0.127	-1.167	7
8	22.36	22.33	0.027	0.793	0.112	0.005	0.107	6
9	18.90	19.45	-0.55	0.118	-1.096	0.016	-1.109	14
10	20.00	20.04	-0.043	0.491	-0.114	0.001	-0.108	13
11	35.30	35.08	0.22	0.793	0.922	0.326	0.914	19
12	19.20	19.45	-0.25	0.118	-0.493	0.003	-0.474	1
13	19.22	19.45	-0.23	0.118	-0.453	0.003	-0.434	9
14	27.54	27.46	0.079	0.793	0.328	0.041	0.313	12
15	20.80	20.49	0.31	0.491	0.818	0.064	0.803	8
16	23.00	23.53	-0.53	0.491	-1.411	0.192	-1.496	18
17	20.50	19.45	1.05	0.118	2.121	0.060	2.713	11
18	30.50	30.47	0.029	0.793	0.121	0.006	0.114	4
19	25.30	25.32	-0.023	0.793	-0.095	0.003	-0.091	5
20	19.20	19.45	-0.25	0.118	-0.493	0.003	-0.474	20

Table B_8: Fit summary for TS (1), EA (2) and WA (3)

1. Response 1: TS

Source	Sequential p-value	Lack of Fit p-value	Adjusted R^2	Predicted R^2	
Linear	0.2875	< 0.0001	0.0553	-0.4049	
2FI	0.9906	< 0.0001	-0.1533	-4.6599	
Quadratic	**< 0.0001**	**0.5977**	**0.9952**	**0.9932**	**Suggested**
Cubic	0.9918	0.1154	0.9923	-0.2544	Aliased

2. Response 2: EA

Source	Sequential p-value	Lack of Fit p-value	Adjusted R^2	Predicted R^2	
Linear	< 0.0001	0.0003	0.7734	0.6539	
2FI	0.6468	0.0002	0.7534	-0.1742	
Quadratic	**< 0.0001**	**0.2429**	**0.9885**	**0.9529**	**Suggested**
Cubic	0.2019	0.3321	0.9920	0.4124	Aliased

3. Response 3: WA

Source	Sequential p-value	Lack of Fit p-value	Adjusted R^2	Predicted R^2	
Linear	0.0217	0.0002	0.3394	-0.0126	
2FI	0.9653	0.0001	0.2032	-3.2032	
Quadratic	**< 0.0001**	**0.8190**	**0.9869**	**0.9789**	**Suggested**
Cubic	0.7310	0.6125	0.9837	0.6457	Aliased

Table B_9: Sample Sequential Model Sum of Squares for TS (1)

Source	Sum of Squares	Df	Mean Square	F-value	p-value	
Mean vs Total	409.51	1	409.51			
Linear vs Mean	24.51	3	8.17	1.37	0.2875	
2FI vs Linear	0.7730	3	0.2577	0.0354	0.9906	
Quadratic vs 2FI	**94.27**	**3**	**31.42**	**1039.39**	**< 0.0001**	**Suggested**
Cubic vs Quadratic	0.0115	4	0.0029	0.0591	0.9918	Aliased
Residual	0.2909	6	0.0485			
Total	529.37	20	26.47			

Select the highest order polynomial where the additional terms are significant and the model is not aliased.

Table B_{10}: Lack of Fit Tests for TS

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Linear	95.18	11	8.65	256.48	< 0.0001	
2FI	94.41	8	11.80	349.79	< 0.0001	
Quadratic	**0.1336**	**5**	**0.0267**	**0.7923**	**0.5977**	**Suggested**
Cubic	0.1222	1	0.1222	3.62	0.1154	Aliased
Pure Error	0.1687	5	0.0337			

The selected model should have insignificant lack-of-fit.

Table B_{11}: Solutions found for numerical optimization during plastic synthesis.

Number	**Dry oven temp.**	**gly.conc.**	**FB**	**TS**	**EA**	**WA**	**Desirability**	
1	**56.524**	**28.613**	**18.266**	**7.248**	**5.462**	**18.600**	**0.993**	**Selected**
2	56.556	28.510	18.266	7.240	5.436	18.600	0.993	
3	56.636	28.619	18.280	7.243	5.448	18.600	0.993	
4	56.649	28.683	18.283	7.250	5.476	18.600	0.993	

Appendix C: Regression model equations

A. Final equations in terms of the actual factor for tensile strength (TS) elongation at the break(EA) and water absorption(WA).

TS=-37.60699+1.21855* d.oven temp+0.67742* gly.conc+0.10827* fiber conc.-0.011137* d.oven temp^{2}-9.51515E-003* gly.conc.2-4.72727E^{-003}* fiber conc.^{2}-1.04444E^{-003}* d.oven temp * gly.conc.+1.62222E-003* d.oven temp * fiber conc.

EA=+56.87257-1.86153 * d.oven temp+0.27053* gly.conc.-0.14893 * fiber conc. +0.016648* d.oven temp^{2}+6.87071E-003* gly.conc.^{2}+0.012372 * fiber conc.^{2}-4.67222E-003 * d.oven temp * gly.conc.-0.010011* gly.conc. * fiber conc.

WA =+76.00817-1.15271* d.oven temp-0.80160* gly.conc.-1.66589* fiber conc.+8.74141E-003* d.oven temp^{2}+0.014075 * gly.conc.^{2}+0.045632* fiber conc.^{2}+6.65556E-003*d.oventemp*fiber.conc.

Where D.oven temp= dry oven temperature, Gly.coc= glycerol concentration, FB= fiber conc.,TS= tensile strength, EA= elongation at the break, and WA= water absorption

The equation in terms of actual factors can be used to make predictions about the response for given levels of each factor. Here, the levels should be specified in the original units for each factor. This equation should not be used to determine the relative impact of each factor because the coefficients are scaled to accommodate the units of each factor and the intercept is not at the center of the design space.

B. Final equations in terms of the coded factor for elongation at the break and water absorption

$TS=+6.98+1.33*A-0.76*B+0.32*C-2.51*A^{2}-2.14*B^{2}-0.27*C^{2}-0.24 *A*B+0.18 *A* C$

$EA =+9.99-5.65 * A+5.89 * B-1.92 * C+3.75 * A^{2}+1.55* B^{2}+0.70 * C^{2}-1.05* A * B -1.13* B * C$

$WA=+19.45-3.02 *A+2.57*B-1.52 * C+1.97 * A^{2}+3.17* B^{2}+2.57* C^{2} +0.75*A* C$

Where A= dry oven temperature, B= glycerol concentration, C= fiber concentration, TS= tensile strength, EA= elongation at the break, and WA= water absorption.

The equation in terms of coded factors can be used to make predictions about the response for given levels of each factor. By default, the high levels of the factors are coded as +1 and the low levels are coded as -1. The coded equation is useful for identifying the relative impact of the factors by comparing the factor coefficients.

Appendix E: 2D interaction graphs of each factor on the three responses

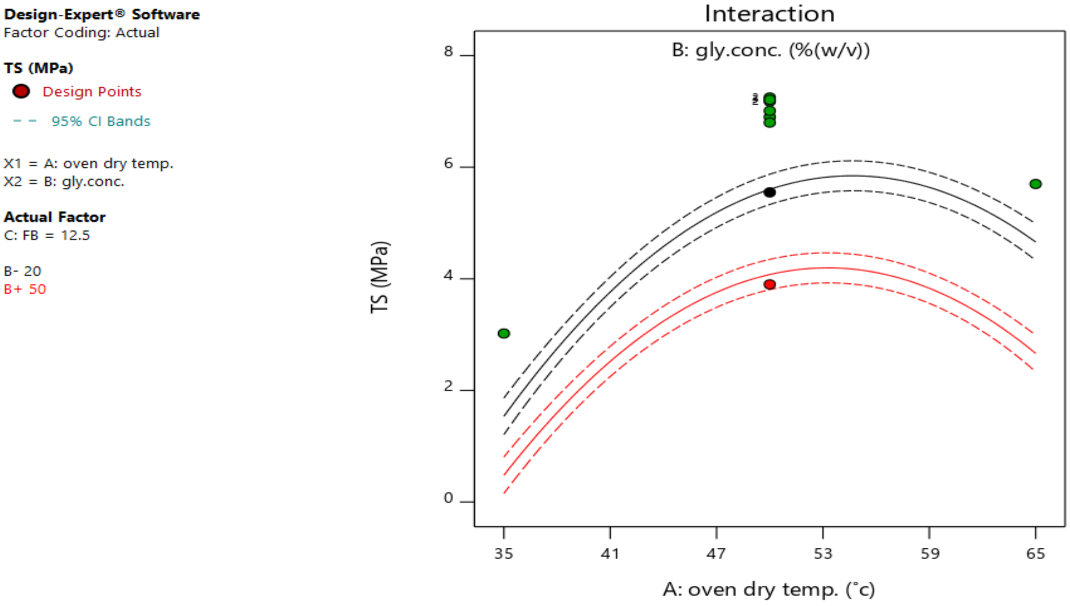


Figure E$_1$: The effect of dry oven temperature and glycerol concentration on the tensile strength of the film

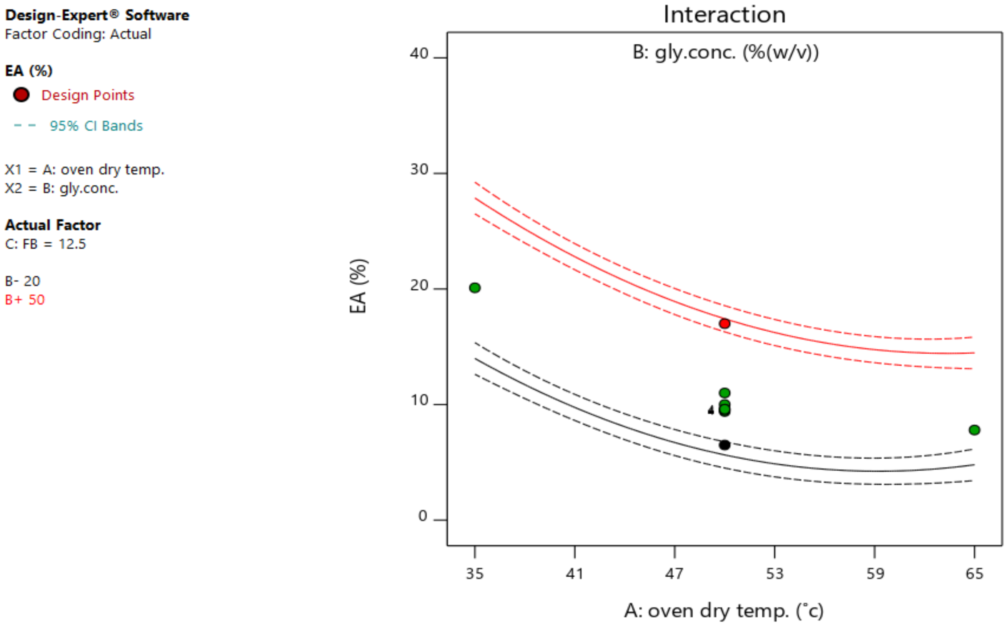


Figure E$_2$: The effect of dry oven temperature and glycerol concentration on elongation at the break of the film.

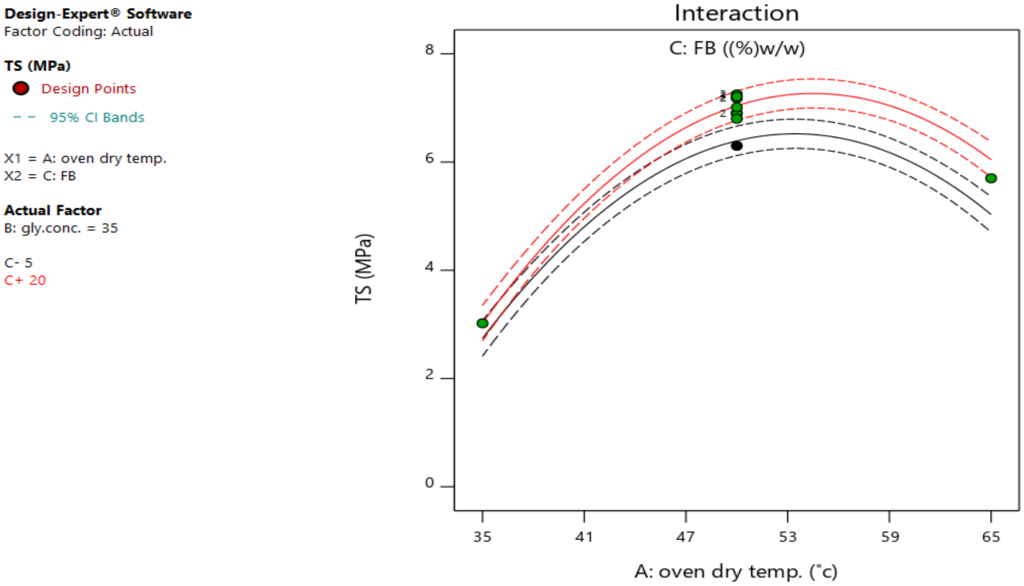


Figure E_3: The effect of dry oven temperature and fiber concentration on the tensile strength of the film

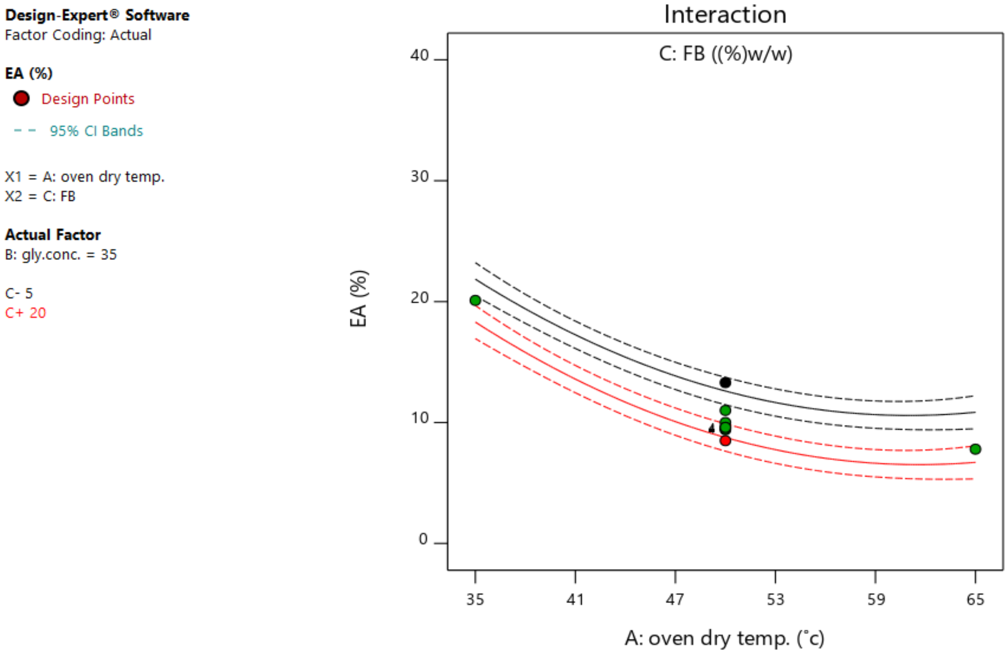


Figure E_4: The effect of dry oven temperature and fiber concentration on elongation at the break of the film.

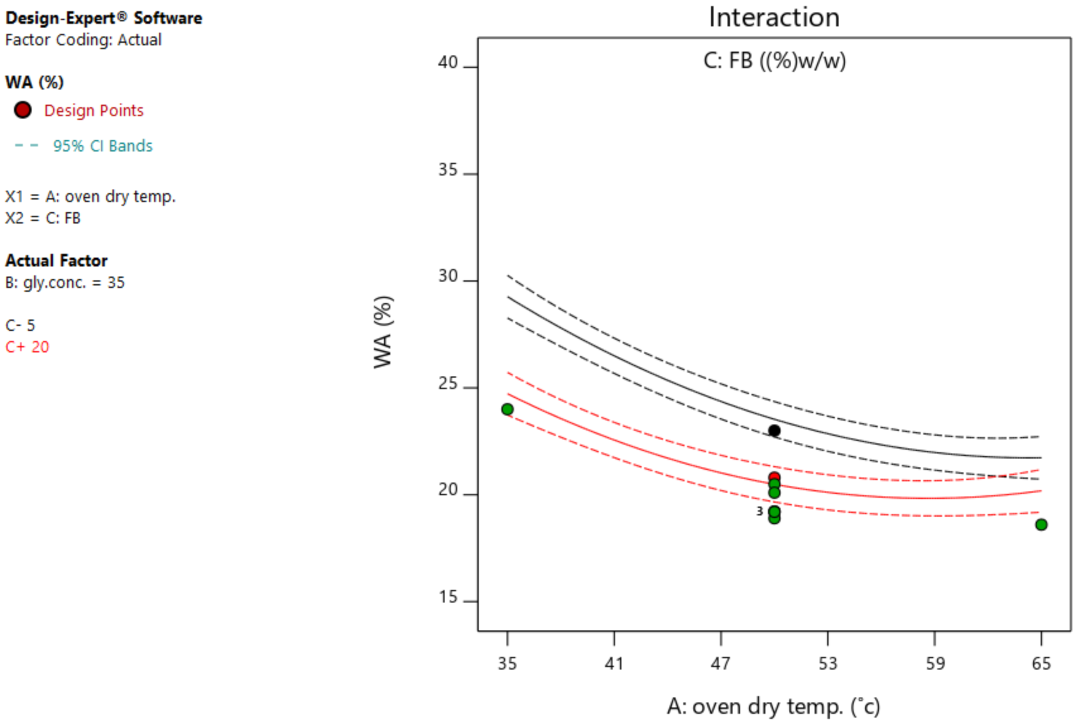


Figure E$_5$: The effect of dry oven temperature and fiber concentration on water absorption of the film.

Appendix F: Ramps numerical optimization.

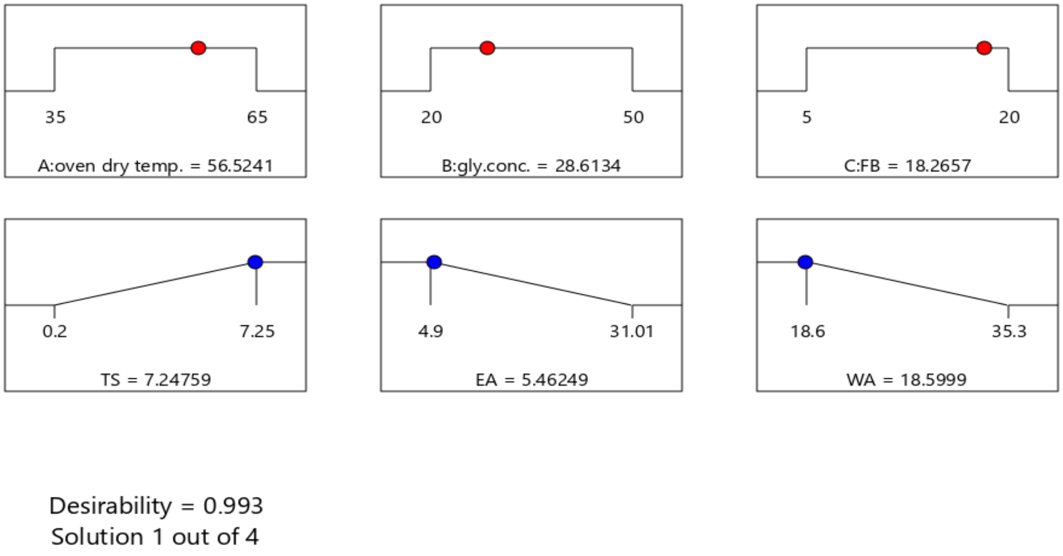


Figure F$_1$: Ramps solution during numerical optimization

Appendix g: sample values of mass loss (%) obtained by TGA

Table g1: Mass loss determined by TGA for both controlled and reinforced film.

Temperature (^oC)	Controlled film.Mass loss(%)	Reinforced film. Mass loss(%)
29.63	0.4	0.19
30	0.65	0.25
40	1.34	0.5
70	4.8	1.79
100	10.8	3.81
150	17.5	7.86
200	22.05	11.3
250	37.5	13.5

Appendix h: A standard amylose calibration curve

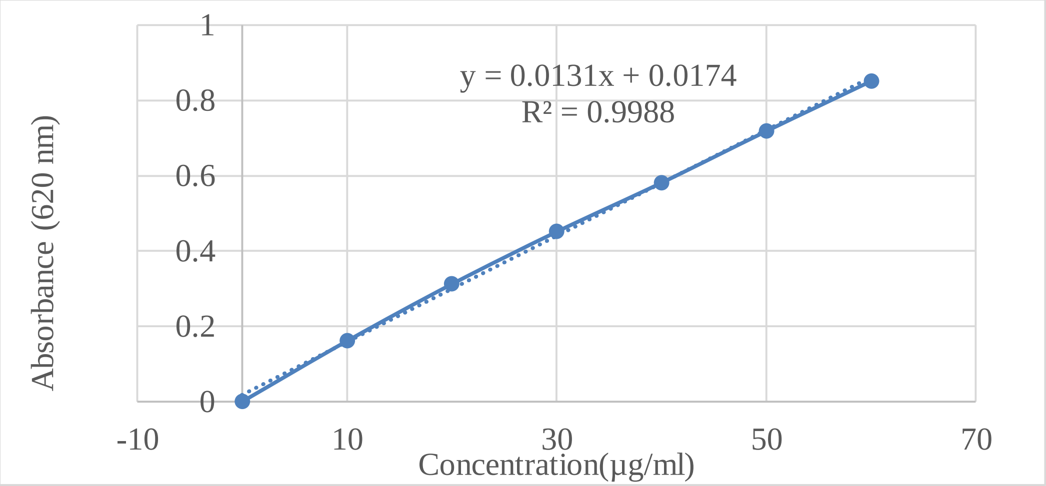


Figure h1: Standard amylose calibration curve

Appendix I; Comparisons of present and previous studies of bioplastics properties

Table I_1: Comparisons of present and previous studies of bioplastics properties

Chemical properties of film	Present Study	Previous studies	Name of authors
Moisture content (%)	9.22	10.22	(Zerihun *et al.*, 2016)
Water Absorption (%)	18.54	18.34	(Mossisa, 2017)
Transparency (%T)	103	113	(Mossisa, 2017)
Thickness (mm)	0.42	0.37	(Mukhopadhy *et al.*, 2017)
Tensile strength (Mpa)	7.23	1.596	(Mukhopadhy *et al.*, 2017)
Density g/ml	1.72	1.07	(Mossisa, 2017)

Appendix J; Perturbation graphs

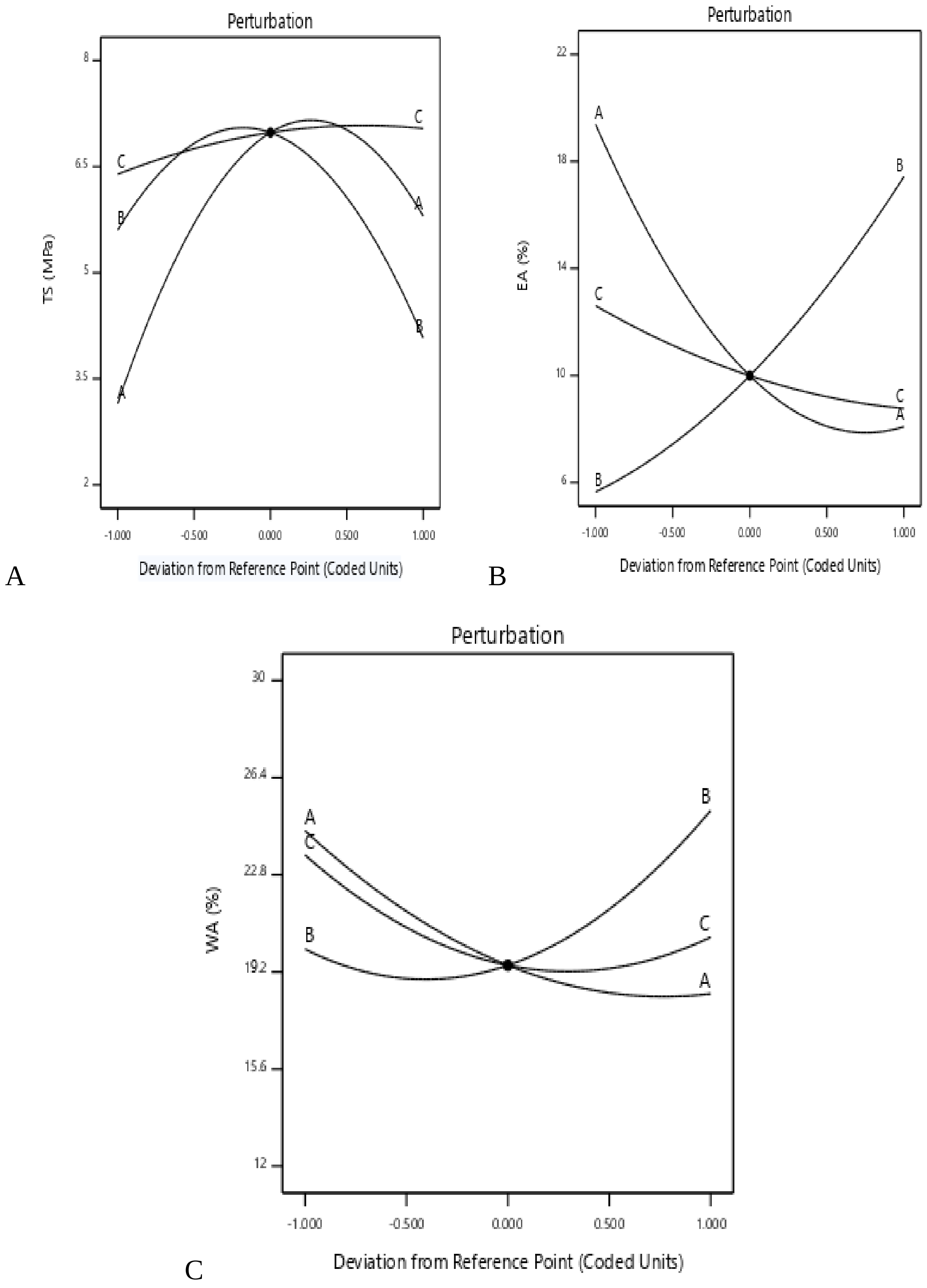


Figure J1; Perturbation graphs for TS(A), EA (B) and WA(C)

Where: A=drying temprature, B=glycerol and C=fiber concentration.

Appendix K; Sample contour plots for significant factors

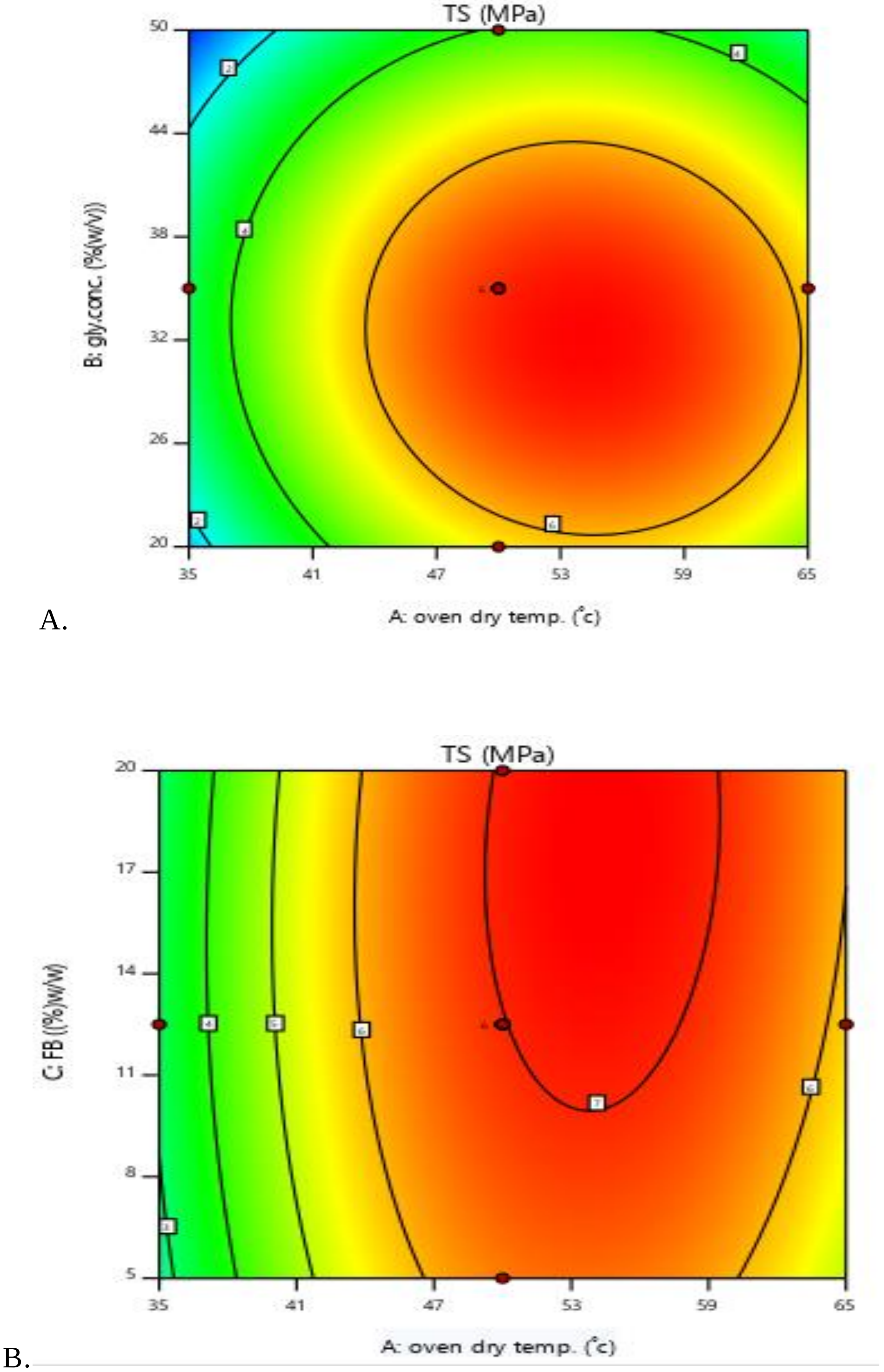


Figure K1; Interaction effect of drying temperature and gly conc.on TS(A)

And the effect of drying temperature and fiber concentration on TS(B)

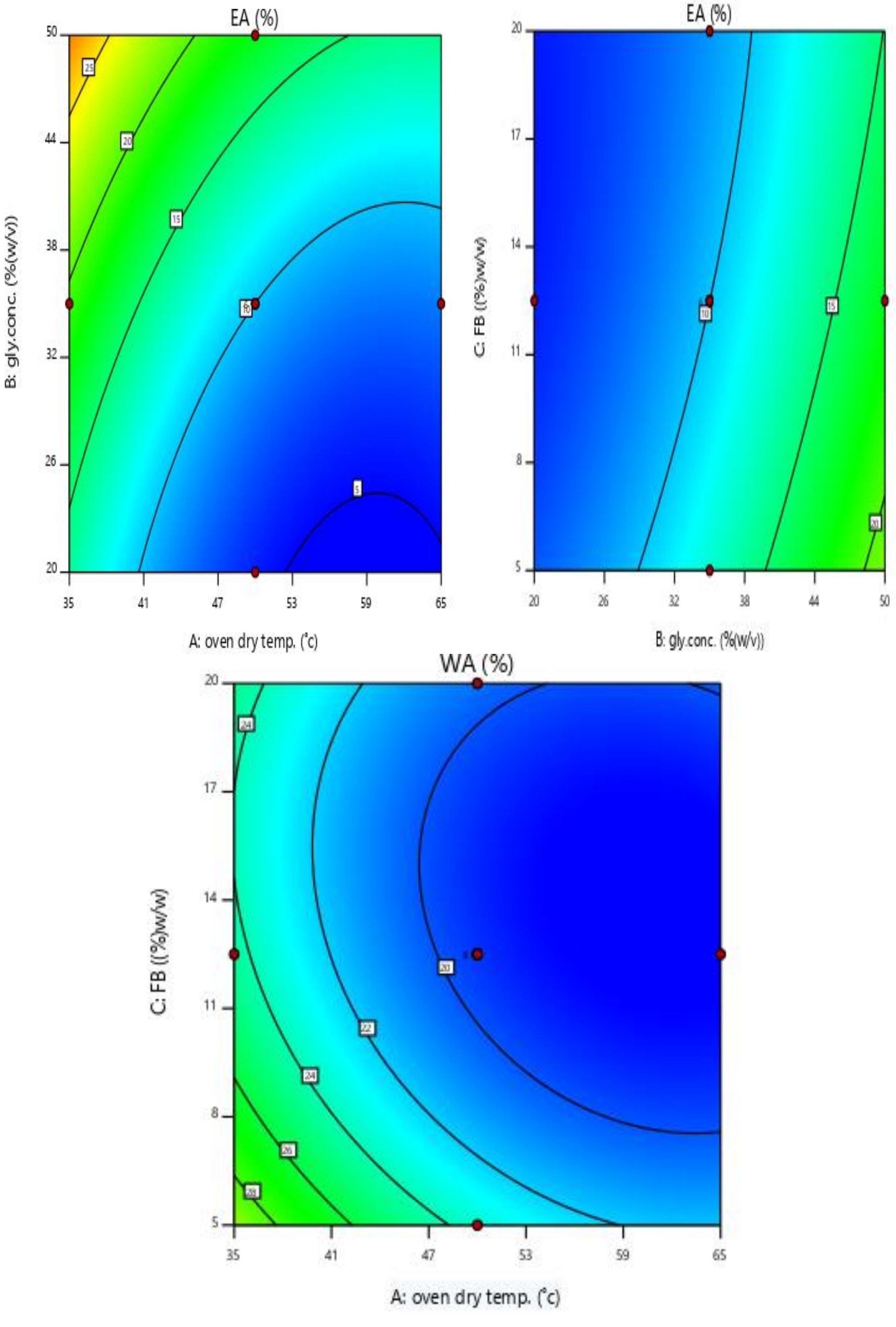


Figure: K2. Effect of the dry oven with glycerol and glycerol with fiber on EA and dry oven with fiber on WA.