DEVELOPMENT AND CHARACTERIZATION OF CASSAVA BAGASSE (*Manihot esculenta* Crantz) AND BLACK SEED (*Nigella sativai* L.) FIBER FILLER CORN (*Zea mays* L.) STARCH BIOCOMPOSITES

By

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Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfilment of the Requirements for the Degree of Doctor of Philosophy

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Doctor of Philosophy

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The vast application of petroleum-based plastics had led to severe issues threatening the environment with numerous hazards such as accumulation of non-biodegradable plastic wastes and release of toxic gases through incineration. Researchers and scientists are in effort to develop eco-friendly materials that could rival the current petroleum-based plastic in terms of physical properties, mechanical strength, gas permeability and thermal stability. In this study, black seed (BS) fiber and cassava bagasse (CB) were used to reinforced corn starch-based polymer through the method of solution casting. Although studies have also been performed using corn starch as bioplastic, limited research has been reported in which BS and CB are reinforced with corn starch for solving the mechanical properties of bioplastics. A series of lab analysis were conducted to quantify the capabilities of the hybrid composites as packaging plastic. The physico-chemical properties of BS and CB were analyzed to understand the strengths and weaknesses in developing composites material. Both BS and CB indicated low cellulose content of 0.14% and 5.39%, respectively, which are beneficial to reduce the hydrophilicity of a composite. High number of lignin (65%) layers in BS is responsible to coat the cellulose hindering them to react with other molecules. The incorporation of 30% of fructose (F) and glycerol (G) into corn starch (CS) based polymer yield improved tensile strength, elongation at break, water absorption capacity and decomposition temperature. Fructose is good in improving mechanical and morphological properties meanwhile glycerol reduces the water sensitivity feature. CS/BS9% composite film exhibit optimum tensile strength and elastic modulus of 14.07 and 83.65 MPa, respectively. CS-BS/CB9% recorded a crystallinity value of 38.8± 2.1% compared to the control composite film (34.6 ± 1.6%). The tensile strength and elastic modulus were enhanced from 14.07 to 18.22 MPa and 83.65 to 118.32 MPa, respectively. Interestingly, a reduction in water absorption capacity but faster biodegradation rate during soil burial test were observed. The development of such fully biodegradable packaging films is important in the effort to address the ongoing environmental problems and gradually substitute the widely used conventional packaging materials.

Keywords: Black seed fiber; cassava bagasse fiber; corn starch; food packaging; hybrid composites

Abstrak tesis yang dikemukan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk Ijazah Doktor Falsafah

PEMBANGUNAN DAN PENCIRIAN BIOKOMPOSIT KANJI JAGUNG (*Zea mays* L) TERISI HAMPAS UBI KAYU (*Manihot esculenta* Crantz) DAN JINTAN HITAM (*Nigella sativa* L)

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Penggunaan plastik secara meluas telah membawa kepada beberapa isu kritikal yang mengancam alam sekitar dengan pelbagai risiko seperti pengumpulan sisa plastik tak terurai, peningkatan kawasan pelupusan sampah, dan pelepasan gas beracun melalui pembakaran. Para penyelidik dan saintis sedang berusaha untuk membangunkan bahan mesra alam yang mampu menandingi plastik buatan sedia ada dari segi sifat fizikal, kekuatan mekanikal, kebolehtelapan wap, dan kestabilan haba. Di dalam pengajian ini, gentian jintan hitam (BS) dan hampas ubi kayu (CB) telah digunakan bagi memperkuat polimer berasaskan kanji jagung melalui teknik tuangan larut. Satu siri analisa makmal telah dijalankan untuk mengukur keupayaan komposit hibrid sebagai plastik pembungkus. Sifat fiziko-kimia BS dan CB telah dianalisa untuk memahami kekuatan dan kelemahan kdua bahan dalam membangunkan bahan komposit. Kedua-dua BS dan CB menunjukkan kandungan selulosa yang rendah iaitu 0.14 % dan 5.39 %, masing-masing, bermanfaat untuk mengurangkan sifat hidrofilik sesebuah komposit. Kandungan lapisan lignin (65 %) yang tinggi pada CB bertanggungjawab menyaluti selulosa untuk menghindari ia daripada bertindakbalas dengan molekul yang lain. Penerapan 30 % fruktosa (F) dan gliserol (G) di dalam polimer berasaskan kanji jagung memperoleh peningkatan pada kekuatan tegangan, pemanjangan semasa putus, kapasiti penyerapan air, dan suhu penguraian. Fruktosa berguna bagi meningkatkan sifat mekanikal dan morfologi manakala gliserol mengurangkan sensitiviti kepada air. Filem komposit CS/BS9% menunjukkan kekuatan tegangan dan modulus keelastikan yang optima, masing-masing pada 14.07 MPa dan 83.65 MPa. CS-BS/CB9% mencatatkan nilai kehabluran sebanyak 38.8± 2.1% berbanding dengan filem komposit terkawal iaitu 34.6 ± 1.6 %. Kekuatan tegangan dan modulus elastik telah dipertingkatkan, masing-masing, daripada 14.07 kepada 18. 22 MPa dan 83.65 kepada 118.32 MPa. Menariknya, pengurangan pada kapasiti penyerapan air tetapi kadar biodegradasi yang lebih cepat ketika ujian di dalam tanah telah didapati.

Kata kunci: Jintan hitam; hampas ubi kayu; kanji jagung; komposit hibrid; pembungkusan makanan

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Doctor of Philosophy. The members of the Supervisory Committee were as follows:

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Declaration by Members of the Supervisory Committee

This is to confirm that:

- The research conducted and the writing of this thesis was under our supervision;
- Supervision responsibilities as stated in the Universiti Putra Malaysia (Graduate Studies) Rules 2003 (Revision 2012-2013) are adhered to.

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LIST OF SYMBOLS AND ABBREVIATIONS

Aa	Amorphous area
Ac	Crystalline area
ASTM	American Society for Testing and Materials
ATR	Attenuated total reflective
BS	Black seed
C_I	Crystallinity index
Ca	Calcium
Ci	Relative crystallinity
Cu	Copper
CA	Crude ash
CB	Cassava bagasse
CBN	Cellulose cassava bagasse nanofibrils
CEO	Clove essential oil
CF	Cassava flour
CHP	Combined heat and power
CNF	Cassava bagasse nanofiber
CO_2	Carbon dioxide
CP	Cassava peel
CS	Corn starch
CSR	Cassava stillage residue
DM	Dry matter
DMA	Dynamic mechanical analysis
DSC	Differential scanning calorimetry

DTG	Derivative thermogravimetric
E	Elongation at break
EE	Ether extract
F	Fructose
Fe	Iron
FAO	Food and Agriculture Organization
FG	Fructose glycerol
FTIR	Fourier transform infrared spectroscopy
G	Glycerol
GMA	Glycidyl methacrylate
GS	Glucose syrup
H_2CO_3	Carbonic acid
HPLC	High-pressure liquid chromatography
K	Potassium
KBr	Potassium bromide
LCNF	Cassava bagasse lignocellulose nanofibers
Mg	Magnesium
$MnFe_2O_4$	Manganese ferrite
MC	Moisture content
MIC	Minimum inhibitory concentration
MMT	Million metric tons
Na	Sodium
NFRC	Natural fiber-reinforced composites
NSO	*Nigella sativa* oil

OEO	Oregano essential oil
OPEC	Organisation of Petroleum Exporting Countries
P	Phosphorus
PC	Protein concentrates
PBS	Poly butylene succinate
PCL	Polycaprolactone
PLA	Poly (lactic) acid
PSD	Particle size distribution
PVC	Polyvinylchloride
PVA	Polyvinyl alcohol
RH	Relative humidity
SA	Succinic acid
SEM	Scanning electron microscope
SiO_2	Silicon dioxide
T_g	Temperature of gelatinization
TGase	Transglutaminase
TGA	Thermogravimetric Analysis
TPS	Thermoplastic sugar palm starch
TS	Tensile strength
WA	Water absorption
WF	Wheat flour
WHC	Water-holding capacity
WS	Water solubility
WVP	Water vapour permeability

XRD	X-ray diffraction
Zn	Zinc
G'	Storage modulus
G"	Loss modulus
ρ	Density
θ	Diffraction angle

CHAPTER 1

INTRODUCTION

1.1 Background

The growing environmental devastation ascribed to the disposal of plastic packaging waste. Therefore, it's necessary to develop environmentally materials to save the ecosystem. Nowadays, most materials used in plastic industry are produced from synthetic polymers that obtained from fossil fuels. These conventional materials due to their non-degradable and non-renewable nature had created serious environmental problems upon disposal. Hence, biopolymer made from natural resources have attracted the attention of many researchers. The creation of materials that degrade faster in the environment has attracted considerable attention such as polylactic acid, thermoplastic starch (TPS) (Ilyas et al., 2020), poly (butylene succinate) (PBS) (Ayu et al., 2020), polycaprolactone (PCL), polylactide acid (PLA) (Nazrin et al., 2020), and polyvinyl alcohol (PVA) (Abral, Ariksa, et al., 2019). Among renewable natural biodegradable polymers, starch is probably the most promising material due to its availability and low cost (Ilyas, Sapuan, Ishak, et al., 2018b). Biopolymers from various natural resources have been considered as attractive alternatives for non-biodegradable petroleum-based polymer, since they are abundant, renewable, inexpensive, environmentally friendly, as well as biodegradable and biocompatible (Kanmani & Rhim, 2014). The development and application of biodegradable starch-based materials have attracted increasing attention since well-known issues of oil shortage and an increased interest in environmental burden reduction due to the extensive use of petrochemical-derived polymers. Currently, more and more countries are introducing regulations and refusing to ban disposable plastic materials. On the other hand, the study of the unique microscopic structures of different starches and their multi-stage transitions during heat treatment have increased basic knowledge of polymeric sciences, particularly for an understanding of the structure and treatment relationships and properties of polymers (Yu & Christie, 2005). Normally starch was extracted from sugar palm, tapioca, potato, corn, rice, and wheat, but recently discovered starch such as corn starch also possess comparable properties as biopolymer.

Due to its unique functional properties, lack of odor, low cost, and quality, corn starch is widely used in processed foods, pharmaceuticals, textiles, and paper products among others. Starch is used as a stabilizer and thickener of fluid foods in the food industry, due to its high viscosity, bland taste, and transparency of newly prepared starch pastes (Xu et al., 2012). The corn plant is a great source of commercial starch available, with maize granules containing approximately 70% of the starch in addition to protein, oil, sugar, and ash (McAloon et al., 2000). Corn starch is a semi-crystalline polymer consisting of a mixture of linear polysaccharide amylose, and a highly branched polysaccharide amylopectin (Maiti et al., 2010). Roughly more than 80% of world starch production is from corn, other important botanical sources of starch are, cassava, rice, wheat, and potato. Corn starch granules consisting of linear amylose ranged between 20-28% and the rest of amylopectin (Bertoft, 2017). Corn starch is widely used in grain processing industries, and it is a valuable ingredient used in applications in the food and non-food industries. Modified corn starch by genetic and physiochemical methods is widely used as a compound enhancement matrix. In addition to amylopectin and linear amylose, the chemical composition of corn starch granules contains secondary components (proteins, lipids, and minerals) and the amounts of these components vary due to the botanical source (Kane et al., 2016). Corn starch is used in textile finishing processes to increase the hardness of the fabric to improve its appearance. It plays a crucial role in the paper and cardboard industry with different types of qualities and qualities because it is mixed in different steps of the manufacturing process (Tabasum et al., 2019). It can act as a pattern for color inks when printing fabrics (Dokić et al., 2010). It is used as a tablet binder and sugar-free ingredient in various products such as toothpaste, emulsifiers, lotions, liquid medicines and creams (Kim et al., 2017). Cornstarch is often combined with other decomposers such as cellulose fibers, plasticizers, and decomposing polyester to make mostly environmentally friendly materials for temporary use such as hygiene, packaging and agriculture (Abbas et al., 2010).

Starch will be act as thermoplastic material in presence of a plasticizer at high condition of temperature. The combination of several plasticizers also beneficial to induce plasticizer-to-plasticizer molecular bonds preventing plasticizer migration. Furthermore, addition of natural fiber as reinforcement is a promising approach to develop a fully biocomposite material with enhanced properties. In most of the applications, the properties of polymers are modified using fillers and fibers to suit the high strength/high modulus requirements. The addition of natural cellulose fibers during the preparation of starch composite films is an effective strategy for improving the functional properties of packaging films, as documented by various researchers (Sahari & Sapuan, 2012). In past decades, there has been an increasing trend towards usage of agro-industrial waste such as corn husk/cob, arrowroot bagasse, apple pomace, sugar palm, wheat straw, sugar beet pulp, sugar cane bagasse, wheat bran, coffee husk/pulp, rice bran/straw, and cassava bagasse (Bodirlau et al., 2013). Utilization of such wastes may contribute to solving the waste problem and at the same time, enhances economic development through the waste to health transformation. Cassava bagasse and black seed (*Nigella sativa*) fiber are some of unwanted wastes generated through production of cassava starch and black seed oil. The higher cellulose content in cassava bagasse is a good reinforcement in starch matrix for better physical and mechanical properties. Even so, abundant hydroxyl groups in the composites still hinders in food packaging application. The hybridization of composites could complement drawbacks of using one type of fiber. The residual oil in the black seed fibers could act as moisture repellent for composite films. Furthermore, the antibacterial property of black seed composites was reported to be extensively utilized in water purification application (Siddiqui & Chaudhry, 2018).

This study focuses mainly on the extraction of biopolymers and biofibers with the aim of manufacturing bio composite materials from cassava bagasse/black seed fiber and then a hybrid composite by adding cassava bagasse fiber. A series of experiments will be carried out for characterization of corn starch as polymer, composites and hybrid composites. Utilization of corn wastes and black seed fibers may contribute to solving the waste problem and at the same time, enhances economic development through the waste to health transformation. Moreover, this research would help to evaluate a new hybrid composite based on natural resources and turn them into the new valuable green product.

1.2 Problem Statements

Despite plastics have been considered as a very convenient and durable material, they generate numerous environmental problems. Plastics are produced yearly to be used in many applications, consequently leading to non-degradable plastics to end up as waste. The complex composition of most conventional plastics made them sturdy and resistant to degradation in natural environment and therefore remain physically on earth for centuries long. This phenomenon is hazardous towards the nature, animal and man. Limited landfill spaces cannot allocate rapid accumulation of plastic wastes and dozen had made their ways into water bodies such as sea, river and lake (Nazrin et al., 2021). There are also alternatives of recycling and incineration to mitigate this issue. However, recycling is inefficient as it requires high cost for capital investment to cover both facilities and work labor. The recycling process itself is complex, which involves high energy consumption process with low return investment. Meanwhile, incineration approach led to the increase amount of greenhouse gases such carbon dioxide, methane, nitrous oxide and ozone (Verma et al., 2016). These gases are responsible for air pollution and global warming deteriorating human health.

Depletion of petroleum resources, landfill problem and pollutions add to the list of plastics drawbacks. This has led to an increasing interest in the production of bio-based materials which are environmentally degradable and can help save the environment. Ironically, the utilization of by-products from agricultural sector such as bagasse and fiber into composite materials as a reinforcement in the matrix potentially improved the attributes of final product and reducing unwanted wastes. Natural fiber composites derived from bioresource could be exploited to develop safe and biodegradable plastic material to rival or even substitute the current petroleum-based plastic. Extensive efforts had been conducted to tailor composites material befitting current available products. A systematic selection of matrix, fiber and additives with practical conceptual design helps to reduce the environmental impact of the product life cycle.

1.3 Research Objectives

The overall aim of this study is to develop and characterize of sustainable and biodegradable plastic material from cassava (*Manihot esculenta*) and bagasse/black seed (*Nigella sativa*) reinforced corn (*Zea mays*) starch biocomposites for packaging application, which are comparable in terms of mechanical strength, thermal stability and water barrier properties with current synthetic plastic. The specific objectives of this research are:

1. To determine the chemical composition, physical, structural and thermal properties of cassava bagasse and black seed fibers.

2. To investigate the tensile, thermal, and morphological properties of corn starch-based biopolymer using different plasticizers.

3. To determine the effect of black seed fiber loading on the thermal stability, morphological properties and tensile strength of corn starch-based biocomposite.

4. To identify the influence of cassava bagasse fiber loading on the performance of a hybrid composite made of cassava bagasse/black seed fiber reinforced corn starch.

1.4 Significance of the Study

1. The findings from the current study are expected to provide significant data in developing high performance biodegradable plastics derived from corn starch, cassava bagasse and black seed fiber for food packaging

2. The replacement of petroleum-based polymer with natural-based composites may reduce the production of conventional plastics thus preventing the health effects from it and reducing dependence on petroleum resource.

3. The production of food packaging plastics with enhanced properties are expected to aid in solving current plastics contamination in the environment

4. Utilization of cassava bagasse and black seed fiber help to minimize the waste products while provide extra income to the farmers by converting these agricultural by-products into useful filler in producing biocomposite materials.

5. Moreover, the employment of hybrid composites provides a cognitive contribution to product design specifications, material selection analysis, conceptual design development, and conceptual design selection.

1.5 Scope of Study

This research focuses on the development of the hybrid composites from bioresources particularly corn starch, cassava bagasse and black seed fiber. Cassava bagasse and black seed fiber waste are the by-products generated from agricultural sector. The obtained biomasses were characterized in terms of physical, thermal, morphological and chemical properties. The variation of plasticizer type (glycerol and fructose) and loading were investigated based on the barrier physical, thermal, tensile and structural properties of corn starch-based film. The composition of 30% both glycerol and fructose exhibited excellent functional properties was selected to proceed for natural fiber reinforcement. The optimal loading of black seed fiber was obtain based on the evaluation of biodegradability behavior, morphological, thermal and tensile properties of biocomposite films. The hybridization of black seed fiber with cassava bagasse were evaluated in terms of biodegradability behavior, morphological, tensile and thermal properties. All film samples were fabricated *via* solution casting method in aqueous condition of distilled water.

1.6 Structure of the Thesis

The thesis was structured in accordance with Universiti Putra Malaysia based on alternate publication thesis format consists of research chapter that represent a separate study that has its own: 'Introduction', 'Materials and methods', 'Results and discussion' and 'Conclusions'. The details of the thesis structure are presented below.

Chapter 1

The problem statements and research objectives regarding this study were mentioned in this chapter. The significant contribution and scope of the study were also pointed out within the chapter

Chapter 2

This chapter consists a comprehensive review of the literature on the vital areas related to the subject of this thesis.

Chapter 3

This chapter of methodology describes every single activity related in this research; from the beginning of materials procuring to samples preparation, testing and results analysis are detailed in this section.

Chapter 4

This chapter presents the first article with the title of "Extraction, characterization, and comparison of properties of cassava bagasse and black seed fibers". In this article, the extraction and characterization of black seed and cassava bagasse fibers will be conducted to explore their possibility in the development of biocomposites.

Chapter 5

This chapter presents the second article with the title of "Development and characterization of cornstarch-based bioplastics packaging film using a combination of different plasticizers". In this article, the tensile, thermal, and barrier properties of corn starch-based biopolymer developed by adding different plasticizers at different concentrations were evaluated.

Chapter 6

This chapter presents the third article with the title of "Effect of black seed fiber, on the physical, thermal, mechanical, morphological, and biodegradation properties of cornstarch-based biocomposites". In this article, the effect of black seed fiber on the physical, thermal, mechanical, morphological, and biodegradation properties of cornstarch-based biocomposites was evaluated.

Chapter 7

This chapter presents the fourth title with the title of “Preparation and Characterization of Black Seed /Cassava Bagasse Fiber Reinforced Cornstarch-based Hybrid Composites”. In this article, the tensile, thermal, physical, morphological and biodegradation properties of black seed/cassava bagasse fiber reinforced cornstarch-based hybrid biocomposites was evaluated.

Chapter 8

This chapter summarizes all findings for the whole research and proposes recommendations for the improvement of the related study fields.

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