Identification and Characterization of Natural Latex from Tapioca (Manihot Esculenta)

by

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LIST OF SYMBOLS, ABBREVIATION AND NOMENCLATURES

$CDCl_3$	Deuterated Chloroform
CR	Chloroprene Rubber
DRC	Dry Rubber Content
DTG	Derivative Thermogravimetric
EPS	Expanded Polystyrene
ER	Extractable Rubber
FTIR	Fourier Transform Infrared Spectroscopy
GPC	Gel Permutation Chromatography
^{1}H-NMR	Proton Nuclear Magnetic Resonance
HA	High Ammonia
Hevb1	Rubber Elongation Factor In Hevea Rubber
Hevb3	Small Rubber Particle Protein
KBr	Potassium Bromide
KOH	Pottasium Hydroxide
LEAP	Latex Eliza for Antigenic Protein
µm	Micrometer
MDa	Megadalton
Micro-BCA	Micro-Bicinchoninic Acid
mRNA	Messenger Ribonucleic Acid
MW	Molecular Weight

MWD	Molecular Weight Distribution
M_w/M_n	Molecular Weight Distribution
M_n	Number Average Molecular Weight
M100	Modulus at 100%
Na_2SO_3	Sodium Sulphite
NBR	Nitrile Butadiene Rubber
NH_4OH	Ammonia Hydroxide
ng/ml	Nanogram/Milliliter
NMR	Nuclear Magnetic Resonance
NRL	Natural Rubber Latex
NR	Natural Rubber
PPO	Polyphenoloxidase
RI	Refractive Index
SALB	South American Leaf Blight
SBR	Styrene Butadiene Rubber
SDS	Sodium Dodecyl Sulphate
SRP	Small Rubber Particles
SRPP	Small rubber particle protein
TGA	Thermal Gravimetric Analysis
THF	Tetrahydrofuran
Tid	The Initial Decomposition Temperature
TMS	Tetramethylsilane
Tmax	The Maximum Mass Loss Rate

TSC	Total Solid Content
USA	United States of America
USSR	Union of Soviet Socialist Republics
WWII	World War II

LIST OF EQUATION

PENGENALPASTIAN DAN PENCIRIAN LATEKS ASLI DARI UBI KAYU (MANIHOT ESCULENTA)

ABSTRAK

Penyelidikan ini tertumpu kepada pengenalpastian dan pencirian lateks asli dari ubi kayu. Pengekstrakan lateks asli daripada daun dan tangkainya dilakukan menggunakan kaedah pengisaran. Sifat kimia, sifat fizikal, sifat mekanikal dan kandungan protein bagi lateks daripada pokok getah dan ubi kayu dibandingkan untuk megkaji potensinya sebagai salah satu alternatif untuk getah asli. Berat molekul dan taburan berat molekul lateks ubi kayu yang diekstrak didapati lebih rendah daripada getah lateks asli dan ianya ditunjukkan oleh taburan unimod dan indeks kepoliserakan luas. Untuk saiz zarah getah, lateks ubi kayu yang diekstrak menunjukkan saiz zarah lebih besar berbanding dengan saiz zarah getah dari getah lateks asli. Dari segi spektroskopi bagi pengujianan NMR, tiga tanda utama cis-poliisoprena pada ujian ditemui untuk kedua-dua lateks. Manakala untuk spektrum FTIR, ianya menunjukkan pecahan-pecahan hidrokarbon dari getah lateks asli dan lateks ubi kayu yang diekstrak mempunyai puncak cis-CH_3 dalam struktur molekul mereka. Sebagai tambahan, kestabilan haba lateks ubi kayu yang diekstrak lebih tinggi daripada getah lateks asli. Ketumpatan sambung silang lateks ubi kayu rendah daripada ketumpatan sambung silang lateks hevea. Lateks hevea menunjukkan sifat-sifat tegangan lebih baik daripada lateks ubi kayu. Penentuan kandungan protein menunjukkan kandungan protein lateks ubi kayu yang diekstrak adalah lebih rendah berbanding getah lateks asli.

IDENTIFICATION AND CHARACTERIZATION OF NATURAL LATEX FROM TAPIOCA (MANIHOT ESCULENTA).

ABSTRACT

This research was focused on the identification and characterization of natural latex from tapioca. The extraction of natural latex from tapioca leaves and stalk was made up by using blender method. The chemical properties, physical properties, mechanical properties and protein content of natural rubber latex from rubber tree and tapioca was compared to investigate its potential as another alternative source for natural latex. The molecular weight and molecular weight distribution of extracted tapioca latex was found lower than natural rubber latex and it represent by unimodal distribution and broad polydispersity index. For a rubber particle size, the extracted tapioca latex show bigger particle size compared to rubber particle size from natural rubber latex. In terms spectroscopy from NMR testing, three major signals of cis-polyisoprene in was found in both lattices. While for FTIR spectra, it shows hydrocarbon fractions from both lattices exhibit peaks that indicate the presence of cis-CH_3 in their molecular structure. In addition, the thermal stability of extracted tapioca latex was higher than the natural rubber latex. The crosslink density of tapioca latex was lower than the crosslink density of hevea latex. Hevea latex shows better tensile properties than tapioca latex. Determination of protein content shows lower protein content in the extracted tapioca latex compared to natural rubber latex.

CHAPTER 1

INTRODUCTION

1.0 Research Background.

Natural rubber is a unique polymeric material, in many of its most significant applications, cannot be replaced by synthetic alternatives. Natural rubber latex has been commercialized into either liquid latex concentrated or dry rubber in a bale form. Dry rubber has long been chosen as the raw material for heavy duty tires that required elasticity, flexibility and resilience properties (Soo et al., 1998). On the other hand, liquid latex concentrated is widely used for manufacturing medical and everyday product such as gloves, condoms, balloon, catheters, pacifier and many more. However, the manufacturer for latex product might face more challenge because of the risen of several issues. This study has been identified three reasons why new alternative of natural rubber latex supplies should be developed:

- Increasing evidence of allergic reaction to the proteins in natural rubber obtained from the rubber tree Hevea Brasiliensis;

- A disease risk to existing supplies of raw material, from Hevea Brasiliensis that could potentially decimate current production;
- Predicted shortage of supply of natural rubber;

Therefore, immediate action needs to develop natural rubber sources that do not cause such the above reasons.

The protein allergy issue becomes part of our everyday when the FDA issued on "Medical alert on latex allergy" in 1991 (Cacioli, 1997). Protein allergen can be life threatening because it can effect on respiratory system for severe cases. In addition, the protein allergy also can cause developing organ manifestation and local wheal and flare reaction (Baur et al., 1997; Yeang et al., 1997). To counter with the issue arises some modification on glove manufacture especially for medical examination has been done. Leaching process has been added into the glove processing. At first, leaching process prior to powdering has been applied to make it easy to strip the glove from the mould. The powder, later on are reported to be found in wound scars andintra-abdominal adhesions that leadto serious complications which suggest that the powdered glove is not suitable for medical purpose. Next, powdered-free gloves have been introduced at which chlorine has been applied in the leaching process. However, the chlorination process is delicate and, ifnot carefully controlled, can damage the glove (Numanoglu et al., 2007; Field, 1997;Truscott, 2002).

Hevea brasiliensis is growth in tropical or sub-tropical country. The tree is originated from Brazil and colonists brought the seed to Malaysia to establish plantation for constant rubber supplies. Malaysia is one of the largest country produce rubber with approximately 49% of the world output rubber (Duggal and Burrett,2010). However, recent spreading rubber tree disease brings concern to many parties. South American Leaf Blight (SALB) is a disease which caused by the ascomycota fungus, Microcyclus ulei that can bring harm to rubber tree. This disease has already become a threat in the South America long time before. The rubber produced during that time is economically lower and not much. If this disease will ever be introduced to either Asia or Africa, it will be a major threat because of the very high susceptibility of all the clone plant at that area. The East Asia including Malaysia on that time will faces devastating economic consequences especially for millions of smallholders whose main income based in rubber tree cultivation (Gronover et al., 2011; Guyot et al., 2008; Le Guen et al., 2008).

The choice of Hevea brasiliensis for its latex does not only because its ability to produce latex with higher amount. In fact, the abundance of the tree, latex quality and the ease of harvesting is another trait that led Hevea brasiliensis become the main source for latex (Soo et al., 1998). High demands of rubber over 40,000 consumer product including tires, footwear and medical devices strike the population of Hevea brasiliensis. Hevea brasiliensis in their natural environment are growing widely distributed within forest but for larger scale, it is planted in monoculture (Davis, 1997; Hagel et al., 2008). In Malaysia, rubbers are growth on both estate and small holders. The estate made up as plantation where 40 hectares of land is required. While for small holder, usually the rubber will be grow in small lot less than 40 hectares. The plantation of rubber covered on mostly in all state of Malaysia until the west side being called as belt of Malaysia (Duggal and Burrett, 2010). Rubberwood as by-product in rubber plantation is relatively cheap source of raw material (Puasa et al, 2010). In 1990, more then 70-80% of the wooden furniture from rubberwood has been produced in Malaysia. Rubber plantation continues to grow until palm oil plantation take place which causes the reduction on the acreage for rubber plantation. Year by year, the acreage of rubber plantation is reduced due to land acquisition for housing as well as the conversion of rubber plantation to palm oil plantation. Due to high rise on the prices of oils and fats, the palm oil is expected to give more profit to the country. In the meantime, oil palm trunk is discovered as one potential raw material to replace the rubber wood in the wood based industry (Abdullah et al., 2012; Basiron et al., 2007).

The conversion to palm oil plantation and South American Leaf Blight (SALB) disease spreading are expected to cause shortage of rubber supply. Meanwhile, the high demand of natural rubber in the worldwide market leads to an idea on using alternative sources. A lot of study regarding on natural rubber alternative sources has been made in order to meet the demand. The study of alternative sources are focusing on euphorbiaceae family that is well known belongs to Hevea brasiliensis. In addition, about 2500 cis 1-4 polyisoprene-producing plant species has been found. Parthenium argentatum is among the alternative species that has garner attention because of its high quality, hypoallergenic latex (Mooibroek and Cornish, 2000). Parthenium argentatum or guayule was actually commercialized started in the early 20th century and then the production is gradually abandoned when Hevea brasiliensis production become more efficient. This extended period of guayule neglect have caused the loss experience and knowledge. However, by the recent study that has been made, they finally succeeds on discover guayule role as another resources for latex (Jan B. van Beilen and Yves Poirier,2007).

Tapioca or cassava (Manihot esculenta) is thought to have originated in tropical Brazil, from where it spread to other parts of South America and to countries bordering the Indian Ocean (India, Sri Lanka, Malaysia, Indonesia) during the 18th century. Products from tapioca roots/tuber such as starch and livestock feed are used in substantial and increasing quantities within and outside the tropical belt (Khelikuzzaman Meera Hussain, 1982). Nowadays, tapioca root has become great potential as a biodegradable material to expanded polystyrene (EPS) in food and industrial foam packaging due to its low density, thermal insulation, strength, low cost and environmental friendly (Glenn and Orts, 2001). Meanwhile, Polhamus (1962) has reported that botanical species such as tapioca also contain rubber. However, no extensive research regarding on developing natural latex from tapioca has been done until now because of less expectation on the capability of latex from tapioca on providing natural rubber latex as rubbery material. With trials to develop a new natural latex, this research study will not only resolve problems on protein allergy in NRL and shortage of NRL supply, but there would be opportunity to incorporate the production of tapioca's latex into integrated, zero waste biorefinery systems. This could provide new income opportunities to Malaysia and further support economic sustainability of biochemistry, biotechnology, agriculture sector and wider industry such as latex manufacturing company.

1.1 Problem Statement

Natural rubber latex (NRL) from Hevea brasiliensis tree is unique biopolymer in many of the applications cannot be replace by synthetic alternatives. However, NRL from the Hevea brasiliensis trees contains a number of proteins that cause minor to severe allergic reactions (Yassin et al., 1994; Kelly et al., 1994). Other problem that NRL encounter with are a disease risk to existing supplies of raw material, which could potentially decimate current production, and predicted shortage of supply of NR (Jan Van Beilen, 2006). Besides, the very little genetic of variability of Hevea Brasiliensis cultivator will increase the potential of being attacked by pathogens and also make the disease spreading easily (Davis, 1997).

The replacement of rubber tree to palm tree worsen the condition more. The demand for rubber is predicted to exceed supply by 25% by the year 2020. Approximately, over 40,000 products from rubber which include 400 medical devices prove that the consumption of rubber is high. About 70% of rubber consumption just only comes from tire industry and this does not count by the consumption of other products such as glove that covered up 10% of rubber consumption. It is concern since only a single feedstock that been used in order to meet all this demands (Mooibroek et al.,2000).

As for alternative, for many decades, chemist were actively searching for rubbery materials which could be manufactured artificially, which is mostly derived from petroleum oil based materials such as styrene butadiene rubber (SBR), nitrile butadiene rubber (NBR), chloroprene rubber (CR) (Ciesielski, 1999). Unlike NRL, these synthetic rubbers are derived from non-renewable sources. A decline in global oil production may begin within the next 10 to 20 years, leading to sharply increasing prices of synthetic rubbers (Hirsh et al., 2006). Therefore, the development of the new natural latex from new renewable resource is needed to tackle problem of protein allergy, reduce dependency of existing rubber supplies and promote utilization local material source as well as give contribution to Malaysia's industries.

1.2 Objective

The main objective of this study is to identify and characterize the natural latex from tapioca (Manihot esculenta) as the potential alternative source of rubber tree Hevea brasiliensis. The objectives are supported by three objective which are:

1.2.1 To extract the natural latex from tapioca leaves and stalk by using blender method.

1.2.2 To investigate and evaluate properties of natural latex from tapioca based on its chemical, physical, mechanical and protein content.

1.2.3 To compare chemical, physical, mechanical and protein propertiesof extracted tapioca latex with natural rubber latex.

1.3 Scope of Study

The main principal of this study is to identify and characterize natural latex from tapioca in term of chemical, physical and protein properties. The study was done in two stages. The first stage is involved with the extraction of natural latex from tapioca through blender method. The second stage is to identify and characterize the extracted natural latex from tapioca. There are several testing that was performed throughout this study for the identification and characterization purposes. The testing was selected in accordance to the protein, biological, chemical and physical properties of natural latex from tapioca. The properties of extracted natural latex from tapioca were compared with natural rubber latex from Hevea brasiliensis.