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## **PREFACE**

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By the Grace and Blessings of Allah the Almighty, we would like to present, with great pleasure, the volume 01 number 02 of Food ScienTech Journal (FSJ). This journal is part of the Universitas Sultan Ageng Tirtaya series of journal.

This journal was envisioned and founded to represent the growing needs of food technology as an emerging and increasingly vital field, now widely recognized as an integral part of agriculture and human living. Its mission is to become a voice of the food technology and science community, addressing researchers and practitioners in areas ranging from chemistry to management, from microbiology to industry, presenting verifiable methods, findings, and solutions.

The journal is intended as a forum for practitioners and researchers to share their research, idea, and solutions in the area of food science and technology. We would like to request for the reader to participate on writing the articles in this journal.

Thank you for your kind attention and support, hopefully this journal will provide lots of benefits for you and society.

Serang, December 2019

Editorial Team

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# A SHORT REVIEW OF BONGKREKIC ACID IN FOOD SAFETY PERSPECTIVE

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## ABSTRACT

Food safety is one of the concern today for consumer and producer of food products. One of its aspect is the availability of dangerous toxin. Bongkreikic acid belongs to foodborne toxin commonly produced by bacteria *Burkholderia cocovenenans*. This toxin's name comes from Indonesian local food, tempe bongkreik, and made several outbreaks in Indonesia with casualties. Bongkreikic acid causes lethal food poisoning which is associated with hyperglycemia. Studies of the bacteria and toxin itself had developed the strategies to prevent the outbreaks. Supported by the hygiene and technologies in parts of the world, bongkreikic acid could be considered under control in the perspective of food safety today.

**Keywords:** Bongkreikic Acid, Food Safety, Toxin, Tempe

## INTRODUCTION

The quality of the foodstuff is measured by many aspects, one of it is food safety. Grunert (2005) distinguished two schools of thought about quality. The first one, the holistic approach, equates quality with all the desirable properties a product is perceived to have. The second, the excellence approach, suggests that products can have desirable properties that consumers, in their own language, may not view as part of quality. Food safety is part of food quality based on the holistic approach.

Microbial indicators are oftenly used to assess food safety and sanitation (Jay, 2000). Presence of metabolites or toxins from specific microorganisms are parts of the microbial indicators for the food safety. Uniquely, bongkreikic acid is a microbial toxin that caused outbreaks but only occurred in very narrow range of foodstuff.

This short review will cover the history, biochemistry, detection, epidemiology, contamination prevention, and regulatory standard of bongkreikic acid.

## HISTORY OF BONGKREKIC ACID

The name *bongkreik* comes from Indonesia's most famous (or infamous) types of solid fermented food called *tempe bongkreik*. This fermented food is mainly made of coconut presscake or the residue from homemade coconut milk inoculated with *Rhizopus oligosporum*, rather than common tempe which made of soybeans. This tempe could become toxic when contaminated and for as long as the local people can remember, food poisoning and death in Central Java, mainly Banyumas and surrounding areas, were periodically caused by contaminated tempe bongkreik (Shurtleff, 2007).

The first outbreak of bongkreik poisoning was recorded by Dutch authorities in 1895 and reported several types of tempe bongkreik in 1902 by Vorderman. Historically between 1931 and 1937 during Indonesia's economic depression, some villagers tried to make tempe bongkreik by themselves rather than buying it from experienced producers, the poisonings become very numerous, recorded up to 10 or 12 a year (Shurtleff, 2007).

A group of Dutch scientist named W.K Mertens and A.G. van Veen from Eijkman Institute Jakarta began to investigate the causes of bongkreki poisoning in the early 1930s. They found the cause of poisonings and discovered the producing bacterium is *Pseudomonas cocovenenans*. Furthermore they isolated and named the two poisonous substances as toxoflavin and bongkreki acid (van Veeg, 1967). Recent genetic sequencing studies have confirmed the bacteria producing bongkreki acid belongs to *Burkholderia cocovenenans* (Lynch, 2009).

**BIOCHEMISTRY OF BONGKREKIC ACID**

Bongkreki acid is a highly unsaturated and heat-stable tricarboxylic fatty acid with a molecular weight of 485 kDa (Fig. 1) (Moebius, 2012). The IUPAC name of this acid is (2E,4Z,8Z,10E,14E,18E,20Z)-20-(carboxymethyl)-6-methoxy-2,5,17-trimethyl docosa-2,4,8,10,14,18,20-heptaenedioic acid with molecular formula of C<sub>28</sub>H<sub>38</sub>O<sub>7</sub> (NCBI, 2019). It was before considered as biologically active secondary metabolites to impart a survival advantage such as inhibiting the growth of other microorganism, known as polyketides.

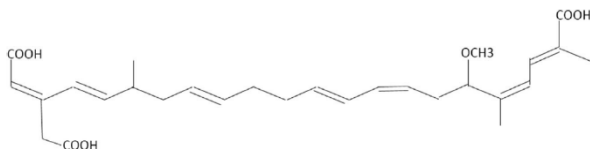


Figure 1. Bongkreki Acid Structure (Moebius, 2012)

This toxin is produced by the gram-negative, aerobic, rod shaped bacteria *Burkholderia cocovenenans*. Like other species of *Burkholderia* genus, the bacteria is

commonly found in the plants and soil. *B. cocovenenans* and the other *B. gladioli* pathovars also produce an electron carrier that generates hydrogen peroxide and subsequent toxicity related to free radicals formation, this toxin named toxoflavin. Its toxicity is relatively mild and secondary to that of bongkreki acid (Lynch, 2009).

Several studies revealed the lethal dose of this toxin. Deshpande (2002) reported that 1-1.5 mg of bongkreki acid can be fatal in humans. Another research suggests an oral LD<sub>50</sub> of 3.16 mg/kg (Liu, 2002). Studies on mice suggest an intravenous LD<sub>50</sub> of 1.41 mg/kg (Moebius, 2012) and an oral LD<sub>50</sub> of 0.68-6.84 mg/kg (Hu, 1984). Bongkreki acid causes lethal food poisoning which is associated with hyperglycemia, research (Kiranadi, 1991) showed that bongkreki acid is a potent inhibitor of the mitochondrial ATP/ADP translocase, inhibits glucose-induced electrical activity in the pancreatic beta-cell through the stimulation of ATP-sensitive potassium channel (K-ATP-channel) activity.

**DETECTION OF BONGKREKIC ACID**

The contaminated food of bongkreki acid can be either detected by the presence of *B. cocovenenans* or bongkreki acid itself. Molecular identification of 16S rDNA is the most commonly used method for the identification of *B. cocovenenans*. However it is reported that sometimes it identified falsely as other *Burkholderia* pathovars for *B. cocovenenans* (Lynch, 2009). Commercial test kits for example the Biologic GN2 System can be used also for the identification. Other methods such as capillary electrophoresis-single strand conformation polymorphisms (CE-SSCP), probe-based cell fishing, or microarray analysis have the ability to be used

Table 1. Bongkreki Acid Outbreaks in Indonesia

Outbreak location	Year	Number affected	Deaths
Java	1895	Unknown	Unknown
	1951-1975	7216	850
	1975	1036	125
	1977	400	70
	1983	450	42
	1988	200	14
Magelang regency	2007	30	10
Banjarnegara	2013	4	1

Source : summarized by Anwar (2017)

Table 2. Optimal conditions for proliferation of *B. cocovenenans* and bongkreki acid production

Factor	Growth of <i>B. cocovenenans</i>	Bongkreki acid production
Temperature	30-37 °C	22-30 °C
pH	>5.5	6.5-8.0
NaCl	<6%	<1.5-2%

Source : summarized by Anwar (2017)

for identification of *B. cocovenenans*. Above all, the most reliable method might be the multiplex PCR protocol (Lynch, 2009).

The presence of bongkreki acid in food samples can be tested using rapid thin-layer chromatographic procedure (Soedigdo, 1977), while the bongkreki acid in environmental samples can be quantified using chromatography-mass spectroscopy and high-pressure liquid chromatography (Hu, 1984).

### EPIDEMIOLOGY IN INDONESIA

Several outbreaks had been occurred in the country of origin of the toxin. Table 1 shows the places of bongkreki acid outbreak in Indonesia. All of it happened in the Island of Java. During the first time of bongkreki acid discovery to 1975, high number of affected people were recorded. Near thousand casualties because of the bongkreki acid outbreaks in this period. Outbreaks still happened several times after 1975 but the number was decreased significantly. Further studies of the toxin made it possible to reduce the casualties. Recent outbreaks of bongkreki acid in Indonesia occurred in 2007 and 2013, both of them were also in Java Island. The development of food technology and food safety in Indonesia had saved great amount of people from outbreak.

### CONTAMINATION PREVENTION

Research and studies of the toxin and its producer had created ways to prevent its lethal effect to the human. Summarized in table 2, strategies can be managed to minimize the contamination and production of bongkreki acid in food. Optimal growth of *B. cocovenenans* is in slightly above room temperature, 30-37°C. Storage of the food is recommended below this temperature to decrease the chance of contamination from the bacteria of bongkreki acid producer. Attention should be made too because the optimal temperature for *B. cocovenenans* to

produce bongkreki acid is slightly below room temperature, 22-30°C. Further decrease of the storage temperature for the food to prevent the production of this toxin is suggested.

Growth of the *B. cocovenenans* is optimum at pH of above 5.5 and it produces bongkreki acid optimally at pH around 6.5-8.0. This indicates the recommended acidity of the food to be 5.5 or less in order to reduce the production of bongkreki acid. While the salinity (NaCl concentration on the substrate) for the optimum growth of *B. cocovenenans* is 6% or less and it optimally produce bongkreki acid in salinity of 1.5-2% or less. These optimal conditions for the production of the bongkreki acid are similar condition for the production of common tempe (Deshpande, 2002).

Another research conducted by Garcia *et al.* (1999) indicated that the concentration and type of lipid in the substrate is critical for bongkreki acid formation. This may explain why bongkreki acid intoxication is limited to certain foods. Thus the fat content of the food should be decreased to prevent the production of bongkreki acid.

### REGULATORY STANDARD

Standard for tempe bongkrek or bongkreki acid contamination is not found in Indonesian documentation. Indonesian National Standardization Body (BSN) have created SNI 3144:2009 for soya bean tempe standard. Table 3 shows the requirements and limits for the production of tempe in Indonesia. Microbial toxins are not mentioned specifically, only the numeration of coliforms and *Salmonella*.

Codex Alimentarius also have created Regional Standard For Tempe (CAC, 2017). In this document, tempe shall comply with the maximum levels (MLs) of the General Standard for Contaminants and Toxins in Food and Feed (CXS 193-1995). Tempe bongkrek or

bongkreki acid also not mentioned in Codex documents.

Table 3. Quality requirements for soya bean tempe production (BSN, 2009)

Criteria	Unit	Requirement
Odor	-	Normal, unique
Color	-	Normal
Taste	-	Normal
Water content (w/w)	%	Max. 65
Ash content (w/w)	%	Max. 1.5
Fat content (w/w)	%	Min. 10
Protein content (N x 6.25) (w/w)	%	Max. 2.5
Crude fiber content (w/w)	%	Max. 2.5
Cadmium (Cd)	mg/kg	Max. 0.2
Lead (Pb)	mg/kg	Max. 0.25
Tin (Sn)	mg/kg	Max. 40
Mercury (Hg)	mg/kg	Max. 0.25
Arsenic (as)	mg/kg	Max. 0.25
Coliform	MPN/g	Max. 10
<i>Salmonella</i> sp.	-	Negative/25 g

## CONCLUSION

Bongkreki acid was first discovered and named after the local Indonesian food, tempe bongkreki. This toxin, produced by bacterium *B. cocovenenans*, had already caused several outbreaks in Indonesia since early 1990s. Studies of the bacteria and toxin itself had developed the strategies to prevent the outbreaks. Supported by the hygiene and technologies in parts of the world, bongkreki acid could be considered under control in the perspective of food safety today.

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## INDONESIAN FOOD INDUSTRY ON HALAL SUPPLY CHAINS

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### ABSTRACT

Halal food industry is one of the potential economic sectors. This industry is experiencing a rapid global development. Indonesia is a country with the most Muslim population in the world. In addition, this country also has various potentials great in developing the halal food industry on the global scene. However, various existing problems and forms of regulation make Indonesia still unable to compete with other countries. Therefore, this review tries to explore various opportunities and the challenges of the halal food industry at Indonesian, especially in terms of the halal supply chain at industrial revolution 4.0. The method used is library research using a variety of secondary sources as research material.

**Keywords:** Food industry, halal supply chains

### INTRODUCTION

Guarantees and safety regarding halal food products are carried out in accordance with the principles of protection, justice, legal certainty, accountability and transparency, effectiveness and efficiency, and professionalism. Guaranteed implementation of halal food products aims to provide comfort, security, safety, and the certainty of the availability of halal products for the public in consuming and using halal products, as well as increasing added value for businesses in producing and selling halal products.

The Halal food product guarantee becomes important considering the progress of science and technology in the fast developing food sector. This significantly affects the shift in processing and utilization of raw materials for food, beverages and other products from what was originally simple and natural to the processing and utilization of raw materials resulting from scientific engineering. Product processing with the use of advances in science and technology allows a mixture of halal and unlawful whether intentional or unintentional.

Therefore, to find out the halal of a product, a special study is needed that requires multidisciplinary knowledge, such as

knowledge in the fields of food, chemistry, biochemistry, industrial engineering, biology, pharmacy and understanding of sharia.

As one of the countries, Indonesia is a country with the largest Muslim population in the world. Based on data from the Central Statistics Agency (BPS), the total population of Indonesia reaches 237.6 million, with a Muslim population reaching 207 people or around 87 percent. With the largest Muslim population in the world, Indonesia has a large market potential for the global halal industry. One of the efforts made by the state to realize the welfare state as a form of the rule of law is to pay attention to the interests of the community.

Especially those who have a Muslim population in this case is Indonesia which has the largest Muslim population in the world in carrying out state life, where the Republic of Indonesia has different characteristics from other countries. Living in Indonesia is heavily influenced by actualized dogma-Islam in public life, without prejudice to the interests of non-Muslim communities.

The achievement of safe food provision has the aim of avoiding people from material that is detrimental to health. Indonesia is as an

archipelagic country which has a large population with a variety of levels of development and food patterns, in this case the role of the government to ensure food security for the people must be very large and it cannot fully rely on free market mechanisms. In this regard, government policy is needed that is adjusted to the objective conditions, and if necessary direct intervention can be done to ensure the achievement of adequate food supply, safety and affordable purchasing power of the community. Products from the halal food industry are the main and most recognized components of the halal industry (Zulfakar et al., 2014).

On the other hand, in the halal industry, there are other challenges, especially in the halal food industry where the behavior of consuming halal food is not necessarily in line with the large Muslim population. Although the appeal of the halal market segment and its potential for growth is so rapid, but research into the consumption of halal food in the Muslim market segment has more or less been ignored (Muhammad et al., 2014).

Food safety is a problem that many developing countries face, including Indonesia. This is usually due to germ contamination and chemical contamination as well as various toxic substances in the food consumed. While it has high nutrition, delicious taste and attractive appearance, but if it's not healthy, the food has no benefit. In this case, the community needs to get adequate protection against the safety of food consumed by improving the quality and health of domestic food, in addition to being able to enhance a positive image for international trade.

Often, the interpretation of halal is largely in the food producers (Alhabshi, 2013). Food intake into a human being is passed down from one generation to another, which supplies a lot of nutrients for humans to survive (Mat Isa & Ismail, 2015). However, in recent years, halal food consumers have increasingly worried about the authenticity of the food they eat (Lubis et al., 2016). The state of the product *halalan thayyiban* can only be achieved if all the possible contamination of illicit and dangerous products can be avoided not only in production but also during the process of the supply chain took place.

Considering halalness is very important for Muslims for their type and food choices, a lot of research on halal has grown significantly in recent years (Yasid et al., 2015). Food is a basic need for human survival, so everyone needs to be guaranteed to get quality and safe food.

Food that is not produced properly and can be a source of microorganisms and chemical contaminants that can be dangerous and cause disease to humans. Cases of food poisoning should not need to occur if food products are processed with the correct processing procedures from upstream to downstream. The halal supply chain is now an emerging business that has attracted global attention (Noorsiah et al., 2015). The application of technology in the halal supply chain, especially in halal products is very important for halal control especially on location, tracking, item identification and data communication. Risk factors for traceability in food products contribute to the risk of contamination of food products during shipping (Yacoob, 2016).

Until now, many efforts have been made to improve food sanitation and hygiene, generally through improving the health quality of food processing facilities. The effort is not easy to do because essentially the food consumed by humans includes a very large amount and type and is produced by an increasing number of processing plants. This problem is an increasingly complex problem and is a challenge that must be faced in the future, because on the one hand the community will be more sensitive to demands for getting better quality food.

At a time when the development of science and technology related to food was not yet advanced, one could easily distinguish between halal food and haram food. In the conditions in the Industrial Revolution 4.0 era like today, differentiating halal or haram food is not an easy matter. This is related to the rapid development of science and technology in the field of food, where food no longer consists of raw materials only, but there are additional ingredients that are likely to come from prohibited foods and their derivatives.

Industrial Revolution 4.0, besides having an impact on the manufacturing sector, also greatly affected globalization, disguising

### *Hadi Peristiwa*

international boundaries and competition. So that halal food is easier to obtain for producers and consumers by using elastic machines, the halal food industry is more affordable both domestically and abroad with the help of some halal food detection devices or applications both from places, materials, and so on. useful and effective in finding halal food without having to struggle and no doubt about its halal status and can lead to more and more competitiveness.

This paper aims to explain food eligibility in the Indonesian halal food industry within condition that will guarantee that produced food according to stages that is fit for consumption. The main focus of this paper is the various opportunities and challenges of the halal supply chain that is growing and developing rapidly in the Indonesian food industry in the Industrial Revolution 4.0 era.

## **HALAL FOOD ISSUES AND DEVELOPMENT IN INDONESIA**

Industrial Revolution 4.0 is an opportunity to make life more prosperous and has introduced technology so easily (Kagermann, 2013). Given the speed of trade globalization, the halal industry is progressing and becoming a significant industry locally and internationally (Damit et al., 2017). The issue of food has long been a special topic in every religion in the world. Food is not only a marker of an ongoing tradition but also a tool in various religious worship rituals. Because it is part of the ritual of belief, religion is an important factor that determines whether something can be eaten or not, when it is permissible or not, as well as what types may or may not be used.

Halal has now become a universal concept. Halal is a term that is exclusively used in Islam which means it is permitted or halal. No party can claim that food is halal without complying with Islamic law. Halal and non-halal include all symbols in Muslim life, not limited to food and drink, but also for safety, animal welfare, social justice and a sustainable environment. Halal and Thayyiban, which means clean and healthy, symbolize intolerance towards cleanliness, safety and quality of food consumed by Muslims (Baharuddin et al., 2015).

In connection with the enactment of the JPH Law (Halal Product Guarantee Act) in Indonesia related to the knowledge of the halal positive list of materials, it is a matter that must be known by the industry in making a product. This inevitably will force the industry to clearly include the halal logo on their products, except for products that are intended as illicit material. Of course, to be able to obtain a halal certificate for industry or business, they must know the knowledge related to the materials (raw materials or supporting materials) used during the production process. Knowledge of material critical points in the food sector will influence or determine whether a product has a status as a halal or haram product

According to the Indonesian Ulama Council (MUI) halal products are products that are in accordance with Islamic sharia, including: First, animals slaughtered must be in accordance with Islamic teachings. Second, the ingredients may not contain pork. Third, not from prohibited materials such as materials made from blood. Fourth, product storage, selling and transporting goods that are not used that are other unclean unless sanctified by Islamic procedures. Fifth, the ingredients may not be from khamr be it food or drink. Sixth, human organs, disgusting impurities and so on. To get halal certification for all products, the supply chain from agriculture to food preparations must be halal (Abidin et al., 2019).

Obviously, the halal industry has tremendous potential but is currently overshadowed by many problems such as the lack of a specific regulatory framework governing halal products, especially in most countries in the world and the uneven level of halal awareness that exists among Muslim consumers (Shah & Yusof, 2014). It was explained that the halal guarantee system was developed based on the concept of three zeros (3.0), namely zero limit, zero defect, zero risk.

The implementation of the halal guarantee system is very important to ensure effective and efficient production of halal products (Majid et al., 2015). In 2014 Law No. 33 of 2014 which there are 68 articles related to the guarantee of halal products it is explained that products circulating and entering and traded in Indonesia are required to be halal-certified. It has been proven to be very

difficult to get the respective halal standards from various halal authorities, especially in the world (Tieman, 2014).

The above law will provide comfort when consuming and utilizing goods or services that are in various places. Because food can be contaminated with food that is not halal when distributed to retailers or when displayed in retail stores (Yusoff et al., 2015). Halal certificates are a guarantee that a product is halal. The Minister of Religion of the Republic of Indonesia decides that halal certificates as collateral for the halal status of a product are marked with halal stamps so that they are safe for consumption by Muslim communities. Consciousness is aware, remember, know, feel, wake up, and understand. Halal awareness can be known with halal materials, slaughtering according to sharia, the place is not occupied by unclean goods (Azizi, 2013).

The technological development of the halal world trade situation should be very encouraging news for a country with a Muslim majority population such as Indonesia. However, the reality on the ground shows that the facts are less encouraging. Many agribusiness actors in the industry based on agricultural products such as fast food and manufacturing in Muslim countries are not ready to face these challenges. With the development of electronic device technology is changing the minds of people who used to use human power replaced by machines (Yulia, 2000). A high standard tool for halal food, environmental sustainability, and halal safety, this tool also provides various halal material requirements for halal products as well as exports to international markets and domestic management (Santo, 2018).

## **OPPORTUNITIES AND CHALLENGES ON INDONESIAN HALAL SUPPLY CHAINS**

Although the potential of the Islamic economic sector is huge, synergistic opportunities for growth and investment are far greater and can even be a necessity in realizing the true potential of each sector. However, many challenges that must be faced in capturing these opportunities include issues around standardization and fulfillment, supply chain integrity, lack of qualified human

resources, consumer education, strategic location on the global scene, business financing, and operational excellence. If halal food producers want to develop in the long run and increase their competitiveness, then they need to make food vendors related factors that better meet the needs of Muslim consumers (Yang et al., 2017). The main consumers of halal food are Muslims, because halal food is designed to meet Islamic requirements (Son et al., 2017). The process must take place from upstream to downstream, so that consumers can easily distinguish the two. It is believed that consumer acceptance of food is often the key to success as far as food processing methods are concerned (Krishnan et al., 2017).

Indonesia is not only potential as the largest halal food market share, but also the largest halal food producer with the wealth of its natural resources. However, these opportunities have not been utilized to the fullest. The existence of food acculturation indicates the adoption of habits to the new diet and food culture by a group of people from other countries dominant cultures (Halawa et al., 2018). Indonesia, in this case, is still the destination market for halal products from abroad. The development of Indonesia's halal industry is considered stagnant. This is because businesses in Indonesia do not consider the halal industry as a big and important business opportunity.

Halal supply chain is a necessity to implement halal value creation into logistics and supply chains that have been recognized recently. Halal food supply chain starts with finding various permitted raw materials and preparing them as needed (Son et al., 2017). If related to the halal supply chain, business practitioners in Indonesia who trade food products should provide clear, honest information about the composition, halal nature of traded food products to protect the rights of Muslim consumers against non-halal food products. But there are still many food products that are circulating in the community that have not included the halal logo or the halal logo is still doubtful. Food products that do not have a halal logo are not necessarily haram, so also products that have a halal logo are not necessarily halal either, because it is likely that the product is not halal.

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Halal supply chain refers to the process of managing procurement, movement, storage and handling of materials, spare parts, livestock and semi-finished inventory, food and non-food, and related information along with the flow of documentation through organizations that adhere to general principles of sharia law (Bahrudin et al., 2011). In addition, the halal supply chain adopts conventional supply chains but with Islamic legal requirements in force. The imposition of Islamic law in supply chain management acts as a basic requirement for sharia-based halal management processes in the sense that all must be halal (permitted) and also thoyyib along the whole chain (Omar et al., 2012).

Logistics plays an important role in protecting product quality and conditions through proper transportation, storage and handling in the supply chain, to achieving its final destination. The success of the halal industry depends on the ability of logistics service management to ensure the integrity of halal products. All halal products must comply with sharia law which states that products must be safe, harmless and healthy from beginning to end (Tieman, 2008).

Thus, adopting halal in the logistical context, halal products must be ensured halal during and throughout all logistical activities which specifically include transportation, storage and warehousing and retail must also comply with sharia principles as a whole (Kamaruddin et al., 2012). Halal supply chain requires a process approach, where processes and procedures must be clearly documented as proof of the halal logistics system. Currently, the food chain has become a part of the style of life has undergone many changes (Nakyinsige et al., 2012).

Although a well-established and well-documented halal food logistics system must be able to prevent contamination, proactive corrective measures need to be defined to reduce or at least minimize the risk of contamination of halal products and business strategies to overcome the perceptions and sensitivity of Muslim consumers. The main purpose of the halal supply chain is to ensure the integrity of halal products for end consumers (Jaafar et al., 2015).

Protection and prevention measures must be taken to ensure that halal products still

remain halal even during the transportation process in the supply chain. As such, halal supply chain management ensures halal integrity is guaranteed from the source (origin) to the point of purchase of the consumer end. This means that halal logistics plays an important role in halal supply chain management in linking suppliers with end customers so as to create a halal supply network, but the integrity of halal in the halal supply chain has not been guaranteed in the industry and its logistics practices. But sometimes consumers worry about the possible adverse effects of eating modified foods (Ismail et al., 2018).

In this case Muslim consumers are willing to pay the costs associated with halal logistics to ensure that sharia compliance parameters in every aspect of the food supply chain are maintained. This shows that the halal logistics policy especially related to the halal supply chain must be obeyed so that Muslim access to halal products can be guaranteed (Rohana et al., 2012).

To protect halal integrity throughout the supply chain or value chain, separation and communication are needed to provide a higher level of assurance to Muslim consumers in terms of protection for brand owners. This separation can be achieved by physical separation systems in transportation, storage and terminals (sea / air / land), to ensure that the flow of halal and non-halal goods is not combined with cargo carriers and for (destinations) Muslim countries are not mixed in transportation and storage (Tieman, 2012). The supply chain is a party network that connects sources to the point of purchase of consumers. Horizontal supply chain structure refers to the number of levels throughout the supply chain.

The supply chain may be long with many levels (many fast moving consumer goods supply chains), or short with only a few levels (such as a mass supply chain). The vertical supply chain structure refers to the number of suppliers / customers represented at each level. A company can have a narrow vertical structure (multiple suppliers and / or customers) or wide (many suppliers and / or customers) (Lambert et al., 2000). Because managing the entire supply chain is a complex task, there is tremendous potential to improve

the performance of halal supply chains through increased coordination throughout the supply chain.

The series of halal supply chains helps in managing global halal supply chains in accordance with the destination market specifications and ensuring that integrity is maintained throughout the halal network. The series utilizes a public halal distribution center at the main gate, transportation consolidation, and the use of innovative logistics concepts (such as halal cargo boxes) (Tieman, 2012).

This role can be fulfilled by fourth-party logistics service providers, namely integrators who gather resources, planning capabilities, and technology from their own organizations and other organizations to design, build and run comprehensive supply chain solutions. The application of the concept of vertical and horizontal collaboration provides better control of the halal supply chain from sources to the point of purchase of consumers and increases consumer confidence in halal-certified products.

The concept of vertical and horizontal collaboration requires a more advanced halal certification system (beyond product certification), expanding halal regulations along the supply chain. Industrial pilot projects and case study research are needed to test various concepts of halal supply chain collaboration and provide practical solutions for the halal industry to optimize halal supply chains. This is to support the development of a customer-driven supply chain that is better able to meet the diverse needs of the Muslim market and to meet general halal standards throughout the supply chain from source to consumer purchase point.

Empowering consumers through the use of strong information technology in all areas of life will lead to the emergence of goods and services economically as well as superior knowledge (Khan et al., 2018). Food handling throughout the supply chain process is important, because the production of halal thayyiban food will be meaningless if the halal and cleanliness of food is not taken care of during the delivery process from the source of supply to the final consumer.

The status of halal thayyiban product can only be achieved if all possible contamination of haram and dangerous

products can be avoided not only in production but also during the supply chain process. The halal process must be seen from the perspective of the supply chain because halal products can only be produced when all activities along the supply chain process are based on the provisions in Islam, not only on production.

So in this case to encourage and facilitate the implementation of the halal supply chain in Indonesia, it needs a strong role of the Indonesian government, in this case including preparing facilities and infrastructure in the implementation of the halal supply chain which includes logistics centers, warehousing airports, goods terminals, ports, goods terminals, warehousing and so on. Business providers and halal supply chain services require deeper insights from the attitudes and behavior of halal consumers (Al-Ansi et al., 2018).

The development of halal supply chains in Indonesia needs to be encouraged and directed for integrated supply chain management. The blueprint halal supply chain development in Indonesia needs to be prepared, as the master plan for development of the halal industry in Indonesia. The blueprint will be needed for the supply chain concept of halal industry in Indonesia. Halal supply chain is the application of halal thayyiban halal principles throughout halal logistics activities. All activities starting from the source of supply, storage, transportation, manufacturing, handling, and distribution must comply with the concept of halal thayyiban.

The purpose of the halal supply chain is to guarantee the halal product along the flow in halal logistics. This halal supply chain develops due to the increasingly high level of consumer awareness, in addition to halal products as well as halal logistical processes. One of the benefits of the halal supply chain is the ability of logistics service management in ensuring the integrity of halal products.

The industrial revolution 4.0 presents huge challenges and opportunities for the advancement of the halal industry in Indonesia. Thus, the readiness of halal industry stakeholders is needed, especially in the field of halal supply chain in solving complex problems in the process of digitizing the halal supply chain. Halal industry players are

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required to continue to innovate in digital product research and is a mandate to advance the Indonesian halal industry.

### **CONCLUSION**

The opportunities and challenges of the halal food industry in Indonesia include ensuring success implementation of halal product guarantees, increase the capability of guarantee of halal products with utilizing technological developments, ensure logistics and supply chain halal food, Sharia funding for the development of the halal food industry. The government must be able to provide facilities both in terms of access, cost and procedural requirements for MSME entrepreneurs in conducting halal certification. Like, the granting of halal certification fee waivers, ease of document requirements submitted and so on. So it is expected, the number of MSMEs, especially food halal-certified drinks can increase rapidly, and can increase revenue in the halal food industry sector.

The government is expected to provide various soft skills training for industry players halal food especially on halal supply chain. The soft skill training can include: product branding, product marketing, Islamic business ethics and so on. So that halal food producers in Indonesia can gain more trust from the public and be able to increase its power their product competitiveness both nationally and internationally.

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## COLOR AND HARDNESS COMPARISON BETWEEN PARBOILED AND NORMAL BLACK RICE

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### ABSTRACT

Parboiling process had a potency to change the color and rice grain hardness of black rice. Although, this process could reduce the cooking time and improve the texture of black rice. The aim of this research was to compare the color difference and grain hardness from normal black rice and two different parboiled black rices. Cempo Ireng black rice variety as a sample was taken from local farmer in Ciampea, Bogor, West Java, Indonesia. There was normal (without parboiling process) black rice and two kind of parboiled black rice used in this research. First, parboiled black rice X (0% sodium citrate concentration, 5 minute steaming time, and 1 time freezing-thawing cycle), parboiled black rice Y (5% sodium citrate concentration, 15 minutes steaming time, and 4 times freezing-thawing cycles). One way Anova, Least Square Difference (LSD) and Independent sample t-test was used to determine the significant differences between mean values. There was significant difference on color parameter between normal and parboiled black rice grain. In cooked rice, there was significant difference on lightness parameter only. The grain of parboiled black rice X was harder than Y.

**Keywords:** black rice, color, grain, hardness, parboiled

### INTRODUCTION

Color is an important parameter that represent the quality of food materials. The color of food could describe the functional components in it. By example, carotenoids had orange, red or yellow color and anthocyanin had purple, black color in food substances (Guine *et al.*, 2009). Some rice become special because of their anthocyanin content. For instance, red and black rice had high anthocyanin content that represent by their color.

The hydrothermal process that purposed at inducing milling, nutritional, and organoleptics improvements is called parboiling (Arendt and Zannini, 2013). This process consists of dehulling, soaking, steaming, drying, and milling (Buggenhout *et al.*, 2013). Some modification such as sodium citrate addition or freezing-thawing cycle could be added to obtain the specific purposes.

Parboiling process on black rice had purpose to reduce the cooking time and improve the texture quality (Widyasaputra *et al.*, 2019).

During parboiling, the use of heat had a potency to destruct the color. Black color in the bran layer of black rice is the expressions of anthocyanins pigments (Oikawa *et al.*, 2015). The anthocyanin content is located in the pericarp and incorporated with fiber, minerals and some amino acids (Kushwaha, 2016). Cyanidin-3-glucoside, cyanidin-3-rutinoside, and peonidin-3-glucoside were some anthocyanin could be found in the black rice pericarp (Loypimai *et al.*, 2015).

Black rice cooking increase the thermal degradation of cyanidin-3-glucoside (Hiemori, *et al.*, 2009). Also, the parboiling process could change the rice grain hardness. Parboiling process made rice become more resistant to breakage during milling (Mir *et al.*, 2013). Hardness become the important parameter for

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storage and transportation. Consumer prefer the whole milled grain rice rather than the breakage one.

The purpose of this research was to compare the color difference and grain hardness from normal black rice and two different parboiled black rice. This comparison result was used as maximum and minimum factor in process optimization of parboiled black rice.

## **MATERIALS AND METHODS**

### **Materials**

Cempo Ireng black rice variety as a sample was taken from local farmer in Ciampea, Bogor, West Java, Indonesia. The other materials such as sodium citrate and distilled water were taken from Setia Guna chemicals store, Bogor, West Java, Indonesia.

### **Parboiling Process of Black Rice**

The process consisted of soaking, steaming under pressure, freezing, thawing, and drying. Soaking was prepared by using sodium citrate solution and distilled water at rice/water ratio of 1:2 (w/v) for 30 minute. Steaming was done by using Hirayama Hiclave HVE-50 (Hirayama Manufacturing Corp., Saitama, JP) in 1.1 bar pressure. One cycles of freezing was carried out with by using LG GR-M712YLA freezer (LG Corp., Seoul, KR) in  $-20 \pm 2$  °C for 22 hours and thawing was performed with LG GR-M712YLA refrigerator (LG Corp., Seoul, KR) in  $4 \pm 2$  °C for 40 minute then left under running water in room temperature for 20 minute. Drying process was done by using Memmert UF-110 universal oven (Mettler GmbH, Schabach, DE).

### **Experimental Design**

Two kind of black rice were analyzed. That were parboiled black rice X (0% sodium citrate concentration, 5 minute steaming time, and 1 time freezing-thawing cycle), parboiled black rice Y (5% sodium citrate concentration, 15 minutes steaming time, and 4 times freezing-thawing cycles). Independent sample t-test was used to determine the significant differences between mean values using the SPSS V.22 Statistical Software Program (SPSS Inc. Chicago, IL, USA). The normal black rice (without parboiling process) was also analyzed to be a reference. Anova one way analysis and Least Square Difference (LSD)

were used to determine the significant differences between normal black rice, parboiled black rice X and Y.

### **Total Color Difference for Rice Grain and Cooked Rice**

Color measurement was conducted by using Chromameter CR300 Minolta (Konica Minolta Sensing Singapore Pte Ltd., Pandan Gardens, SG). Rice grain or cooked rice sample was placed on the transparent dish then measured with Chromameter. The measurement resulted  $L^*$ ,  $a^*$ , and  $b^*$ .  $L^*$  value represent the lightness from dark to light (0-100).  $a^*$  value showed the red to green chromatic color,  $a^+$  for red color and  $a^-$  for green color.  $b^*$  value showed blue to yellow chromatic color,  $b^+$  for yellow color and  $b^-$  for blue color. The normal black rice color value was used as a reference. Total color difference was counted by using the equation:

$$\Delta E^*_{ab} = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$

### **Rice Grain Hardness**

The hardness of grain after parboiling without re-cooking was determined as the rice grain hardness. This analysis was performed by using Kiya Grain Hardness Tester Instrument (Kiya Seisakusho Co. Ltd., Kawagoe, JP). Ten grains of black rice sample were tested. The needle was set to 0. The grain was pressed by using tester spindle until it sounds "crack". The needle would show the hardness value in kg.

## **RESULTS AND DISCUSSION**

Before compared two kind of parboiled black rice, the normal black rice was analyzed. The result of analysis was showed in Table 1. Cooking process lower the lightness ( $L^*$ ) and blue-yellow chromatic color ( $b^*$ ) value of black rice. But, the red-green chromatic color ( $a^*$ ) of cooked black rice was higher than rice grain. The anthocyanin dissolution from rice to the water during cooking process made the cooked black rice had darker color than rice grain. Anthocyanin was the water soluble pigment that could be extracted during cooking process of colored rice (Handayani *et al.*, 2014). The water dispersed anthocyanin into the all surface of cooked rice.

The individual rice grain color difference between normal black rice,

parboiled black rice X, and Y could be found in Table 2. There was significant difference ( $\alpha=0.05$ ) between normal black rice and parboiled black rice in Lightness ( $L^*$ ), red-green chromatic color ( $a^*$ ) and blue-yellow

chromatic color parameter ( $b^*$ ). But, the difference between parboiled black rice X and Y in all ( $L^*$ ,  $a^*$ ,  $b^*$ ) color parameter was not significant.

Table 1. Physical characteristic of cempo ireng black rice variety

Parameters	Values $\pm$ Standard Deviation
<b>Rice Grain Color**</b>	
$L^*$	$27.78 \pm 0.13$
$a^*$	$3.89 \pm 0.11$
$b^*$	$2.66 \pm 0.01$
<b>Cooked Rice Color**</b>	
$L^*$	$18.10 \pm 0.18$
$a^*$	$7.02 \pm 0.16$
$b^*$	$1.85 \pm 0.04$
Rice Grain Hardness (kg)	$5.60 \pm 0.20$

\*\*Source: (Widyasaputra, Syamsir, and Budijanto 2019);  $L^*$  = Lightness;  $a^*$  = red-green chromatic color;  $b^*$  = blue-yellow chromatic color

Parboiling process could change the lightness of black rice became darker than before. This could happen because the soaking process in parboiling process helped water soluble component such as anthocyanin

pigment to leach out from the rice bran (Tang *et al.*, 2016). After leached out, the pigment was well dispersed into the surface area of black rice.

Table 2. Rice grain color (CIE  $L^*$ ,  $a^*$ ,  $b^*$ ) comparison between normal black rice, parboiled black rice X and parboiled black rice Y

Parameters	Normal Black Rice**	Parboiled Black Rice X	Parboiled Black Rice Y
$L^*$	$27.78 \pm 0.13^a$	$19.70 \pm 0.82^b$	$20.03 \pm 0.75^b$
$a^*$	$3.89 \pm 0.11^a$	$2.29 \pm 0.59^b$	$1.48 \pm 0.49^b$
$b^*$	$2.66 \pm 0.01^a$	$0.65 \pm 0.18^b$	$0.52 \pm 0.21^b$

<sup>a</sup> the numbers in the same row with same letter did not have significant difference ( $\alpha=0.05$ ); \*\*Source: (Widyasaputra *et al.*, 2019);  $L^*$  = Lightness;  $a^*$  = red-green chromatic color;  $b^*$  = blue-yellow chromatic color

Table 3 showed the comparison of cooked rice color parameter between normal, parboiled X and Y black rice. The one way Anova test showed that there was significant difference ( $\alpha=0.05$ ) between lightness value ( $L^*$ ) of normal black rice and parboiled black rice. But it did not have significant difference in red-green chromatic color parameter ( $a^*$ ) and blue-yellow chromatic color parameter ( $b^*$ ) between normal and parboiled black rice. Also, there was no significant difference

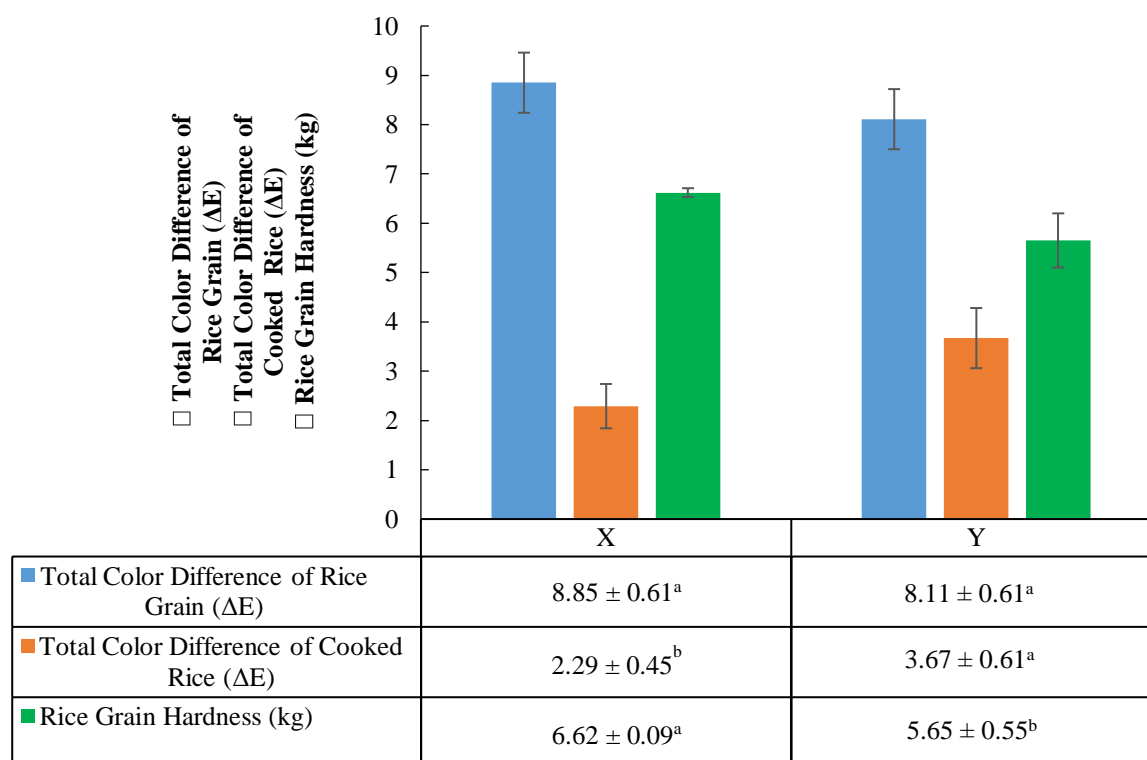
between parboiled black rice X and Y in all individual color parameter (CIE  $L^*$ ,  $a^*$ ,  $b^*$ ).

The cooked rice of parboiled black rice tended to have brighter color than normal black rice. Cooked parboiled black rice had been passed two time cooking process. Firstly, when it was parboiled. Secondly, when it was re-cooked. It made higher anthocyanin lost than normal black rice. Anthocyanin pigment in colored rice dissolve to the water during cooking process (Handayani *et al.*, 2014).

Table 3. Cooked rice color (CIE  $L^*$ ,  $a^*$ ,  $b^*$ ) comparison between normal black rice, parboiled black rice X and parboiled black rice Y

Parameters	Normal Black Rice**	Parboiled Black Rice X	Parboiled Black Rice Y
$L^*$	$18.1 \pm 0.18^b$	$20.27 \pm 0.57^a$	$21.50 \pm 0.77^a$
$a^*$	$7.02 \pm 0.16^a$	$7.17 \pm 0.73^a$	$6.10 \pm 0.78^a$
$b^*$	$1.85 \pm 0.04^a$	$1.95 \pm 0.27^a$	$2.27 \pm 0.70^a$

<sup>a</sup> the numbers in the same row with same letter did not have significant difference ( $\alpha=0.05$ )



<sup>a</sup> the numbers in the same row with same letter did not have significant difference ( $\alpha=0.05$ )

Figure 1. Total Color Difference of Rice Grain, Cooked Rice ( $\Delta E$ ), and Rice Grain Hardness (kg) Comparison between Parboiled Black Rice X and Parboiled Black Rice Y

The independent sample t-test showed that there was significant difference ( $\alpha=0.05$ ) between parboiled black rice X and Y on total color difference of cooked rice and rice grain hardness parameter. But, the total color difference of rice grain was not significant. After re-cooking, parboiled black rice Y had higher total color difference than X. Sodium citrate addition on parboiled black rice Y was higher than X. The addition of sodium citrate solution helped rice to resist the Anthocyanin. The anthocyanin degradation could be prevented by reducing the pH value with the addition of citric acid (Patras *et al.*, 2010).

Generally, parboiled black rice had higher grain hardness than unparboiled. This result was in line with parboiled white rice. Parboiling process could increase the rupture force of white rice (Taghinezhad *et al.*, 2015). Parboiled black rice Y had lower grain hardness than X. The sodium citrate addition in parboiled black rice Y was higher than X. This condition made rice had more porous structure (Husain *et al.*, 2007). Rice became more brittle and easily broken. Soaking in parboiling process also reduced the broken grain percentage (Ayamdoo *et al.*, 2013).

## CONCLUSION

The difference of normal black rice and parboiled black rice lightness, red-green chromatic colorand blue-yellow chromatic color was significant. There was significant difference between lightness value of normal black rice and parboiled black rice. Parboiled black rice X had significant difference with parboiled black rice Y on total color difference of cooked rice and rice grain hardness parameter. Although, the parboiled black rice color reduction X was bigger than Y, the difference was not significant. In cooked rice, parboiled black rice Y had bigger color reduction than X. The grain of parboiled black rice X was harder than Y. The total color difference of cooked rice and rice grain hardness result could be used as a maximum and minimum factor in process optimization.

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# INCREASE OF STABILITY SHELF LIFE AND KINETICS STUDY OF TYPE 1 BROWN RICE MILK THROUGH ADDITION OF ALGINATE EXTRACT

## FROM *Sargassum Binderi*

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### ABSTRACT

Brown rice milk is milk from brown rice. It is used lactose intolerance and low fat milk. This research aimed to use the sodium alginate extracted from *Sargassum binderi* as an stabiliztion and degradation of brown rice milk. *Sargassum binderi* seaweed taken from Sayang Heulang beach, Pameungpeuk Garut, West Java. The purpose of this research is increase the stability of milk and extend shelf life of brown rice milk. The method used was extraction alginate from *Sargassum binderi* and analysis of brown rice milk. The analysis were dye degradation, deposition rate and shelf life of brown rice milk. Result of this research was constant pH value of the composition of brown rice milk from various variations of the addition alginate until the 15 days to various compositions of adding alginate to milk. Then, the sedimentation rate of the effect addition alginate was in order 1. The addition of 1.2% alginate had the lowest deposition rate constant at 0,0171. The constant rate of deposition brown rice milk samples with the addition of 1.2% alginate which is 0.0022 was the best milk sample because it successfully suppressed the degradation rate of dyes up to 95.79% compared to those without the addition alginate. The number of microbes that appear in milk samples did not exceeded the maximum limit of microbial contamination.

**Keywords:** Brown rice milk, *Sargassum binderi*, alginate, stability

### INTRODUCTION

Milk is something we all know has a very important place in our daily diet (Gomez et al., 2009). This gives us the essential calcium needed for growth and maintenance of the body. Many people need nutrition in their body to supply energy and nutrition (Battaglini et al., 2009). Nutritional benefit in combination with acceptability in food products is a significant opportunity for food industry (Molkentin et al., 2013). People have started to drink vegetable milk instead of cow's milk.

The reasons also variety, from trying to be a vegetarian, because they have lactose intolerance, to looking for lower fat milk choices. One of the vegetable substitutes for

cow's milk which is now widely consumed is brown rice milk (Moongngarm et al., 2010).

Self-made brown rice milk contains almost no trans fat and cholesterol. In some finished products, fat and cholesterol levels may be present as a by product of the production process as well as flavorings and or added sugar. That is, the content of trans fat from rice milk is the least compared to all vegetable milk substitutes for this cow's milk. Therefore, good brown rice milk is consumed for people who want a diet low in cholesterol and low in fat (Durand et al., 2003). Because it is low in fat and cholesterol, drinking vegetable milk can also nourish the heart. Moreover, the magnesium content in rice milk

is also efficacious for controlling blood pressure.

Brown rice milk is a substitute for cow milk which is very good and healthy (Alvarez et al., 2005). The benefits of brown rice for the body include rice milk extracted directly from rice grains, it is lactose-free milk as opposed to cow's milk. It really helps people who are lactose intolerant. Secondly, brown rice milk contains higher levels of carbohydrate than cow's milk. Third, the level of cholesterol in rice milk is zero. This makes it very healthy, especially for people with heart conditions. Fourth, brown rice milk is also very low in fat, so everyone who maintains weight can drink it without worry (Prado et al., 2008).

In this study brown rice milk combined with alginate from chocolate algae (Murdinah et al., 2009). The brown algae used is *Sargassum binderi*. One of the potential biological resources from Indonesian marine waters is seaweed with various types (Shirosaki et al., 2011). Seaweed is part of aquatic plants, which are included in the macro class of algae (Aseer et al., 2009). Alginate is a linear copolymer consisting of two monomeric units, namely D-mannuronic acid and L-guluronic acid (Vauchel et al., 2008).

The ratio of the monomers of alginate is important in relation to its bioactive properties and the structural properties of the gel (Siew-Ling et al., 2011). This research aimed to use the sodium alginate extracted from *Sargassum binderi* as a stabilization and degradation of brown rice milk. In this research, a kinetic analysis was carried out to determine the level of milk stability.

## MATERIALS AND METHODS

### Tools and Materials

The research materials is used brown rice from supermarkets. The brown rice was included in the cigeulis variety which resistant to WCK biotype 2,3 HDB strain IV. Brown rice is planted in lowland rice fields (less than 500 m above sea level). *Sargassum binderi* seaweed taken from Sayang Heulang beach, Pameungpeuk Garut, West Java. In this research, reagents was  $\text{CaCl}_2$ , aquabides, formaldehyde, EDTA,  $\text{Na}_2\text{CO}_3$  and chloroform.

## Method

### Extraction Alginate from *Sargassum binderi*

Two grams of samples was immersed in a 2%  $\text{CaCl}_2$  (w/v) solution for 2 hours, then washed with aquabides. The sample was soaked in a formaldehyde solution for 2 hours and washed again with aquabides 3 times. The extraction process was carried out by adding 3% (w/v) 1 M  $\text{Na}_2\text{CO}_3$  and 0.5 gram EDTA at pH 11. Then it was filtered a muslin cloth and was precipitated ethanol as sodium salt. The precipitate was separated by centrifugation and dried in an oven at 60°C (Latifi et al., 2009).

### Analysis of Brown Rice Milk Dye Degradation

One mL of sample is inserted into 2 mL microtube. Then it is added 1 mL chloroform and is stirred by vortex. The sample is centrifuged at a speed of 12,000 rpm for 30 minutes so that it will separate into two phases. The top phase is decanted and absorbance measurements are made at  $\lambda = 241$  nm based on the maximum wavelength scan in the range of 200-400 nm.

### Analysis of Brown Rice Milk Deposition Rate

One mL of sample is put into a measuring flask. Then it is demarcated to 10 mL by aquades. Then tranmitan measurements are carried out at a wavelength of 656 nm. The transmittal results are converted into turbidity values with the equation below.

$$S = -\log \left( \frac{T}{100} \right) \quad (1)$$

Information : S = Turbidans

T = Transmittans

### Self Life of Brown Rice Milk

The pH value is measured by a pH meter. Samples have been pasteurized and packaged in vial bottles of 10 mL, each pH value is measured on day 0 to day 15 using a pH meter. Before being used the pH meter was calibrated using a buffer solution of pH 4, 7 and 10.

## RESULTS AND DISCUSSION

### Production of Brown Rice Milk

Brown rice milk is milk obtained from rice. This is obtained by processing the rice through processing. For this process, the most



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preferred rice is brown rice. Brown rice goes through several long processes, after the final product of rice milk and husk is separated. In order to make rice milk, brown rice is crushed and milk is extracted. Brown rice milk in this study is SBM Type I. Brown rice milk comes from direct brown rice which is processed into milk comparison of the composition used in making brown rice milk between brown rice/brown rice with water is 1:15 (w/v). The following is the result of processed brown rice milk, SBM Type I.



Figure 1. Type of I brown rice milk

Physically SBM type I milk has a physical appearance red color. The red color produced from the milk comes from aleurons that contain genes that produce anthocyanins (red pigment pigments that act as antioxidants).

### ***Sargassum binderi* Seaweed Samples**

Seaweed is the main ingredient that used in this study. Seaweed was obtained from Sayang Heulang beach, Pameungpeuk District, Garut, West Java. The beach is one of the coral beaches in the southern region of Java. Seaweed in this research is *Sargassum binderi* which grows naturally, without cultivation process. Seaweed samples were taken in February 2016 during low tide, where the depth of the sea water was only 10-30 cm. The coordinates of the sampling point are 7° 40' 12 "LS and 107° 41' 37 "BT which are located around the main gate of Sayang Heulang Beach.

*Sargassum binderi* samples are found at a distance of 30-70m from the shoreline. *Sargassum binderi* seaweed is chosen still alive, fresh and brightly colored. The sample is separated from impurities such as sand, stone or pieces of coral, then packaged in a plastic filled with water. The following is a sample of *Sargassum binderi* seaweed which is the result of sampling at Pantai Sayang Heulang.

From the Figure 2, it can be seen that *Sargassum binderi* has the characteristics of a flat thallus ( $\pm 1.5$  cm), smooth/slippery, reaching a height of about 60 cm. The alternate branching is regular, opposite (left right), the main branches close together, arising on the short main stem ( $\pm 1-2$  cm) above holdfast. Oval leaf, serrated edge, length=5 cm, width=1 cm, sharp tip. Round bladder, rounded or pointed tip, winged 1 cm long, 0.4 cm in diameter.



Figure 2. Photo sample of *Sargassum binderi* from Sayangheulang Beach, Garut, West Java

Reproductive organs form special stalks, slashing, flattened, jagged. The leaves and bladder are shrink. It's habitat grows on rock sytrates, generally in flat reef areas, near the outer edges that are affected by relatively strong and constant water movements (DG of Fisheries and Aquaculture, 2009).

### **Alginate from *Sargassum binderi***

Sodium alginate in this study is yellowish brown. Brownish color is the result of reactions from the presence of phenolic compounds that are still contained in alginate. Density and viscosity of sodium alginate with a concentration of 0.1% (w/v) are 1.01 g/mL and  $7.65 \times 10^{-3} \text{ kg.m}^{-1}\text{s}^{-1}$ . Sodium alginate water content is 12.2% (w/w).



Figure 3. Na-alginate from *Sargassum binderi* (100 mesh)

Sodium alginate water content is in the range of 5-20%, still in the range of water content obtained in sodium alginate (Mushollaeni, 2011). While the amount of

sodium alginate water content determined by the Food Chemical Codex (1981) is a maximum of 15% and the maximum sodium alginate water content for food ingredients is 13% (Cottrell and Kovacs, 1977). Next is the structure of sodium alginate.

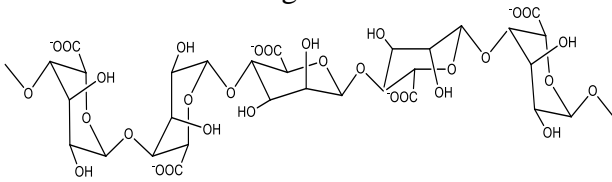


Figure 4. Structure of sodium alginate (Draget, 2000)

In the spectrum of figure can be seen the absorption peaks of alginates that have been characterized by FTIR. The presence of peaks in the area of about 3,500-3,200  $\text{cm}^{-1}$  indicates the presence of hydroxyl groups (O-H) that bind to hydrogen. Wave numbers 1,680-1,600  $\text{cm}^{-1}$  indicate the presence of a carbonyl group (C=O) as an aromatic group, 1,500-1,100  $\text{cm}^{-1}$  indicates the presence of a carboxyl group (COOH).

Sodium in the alginate isomer is located at the peak of absorption of 1,523  $\text{cm}^{-1}$ . Absorption peaks of 900-890  $\text{cm}^{-1}$  indicate typical areas of guluronate fingerprints, while 850-810  $\text{cm}^{-1}$  indicate typical areas of fingerprints of mannuronates. The presence of a typical fingerprint area of guluronate and mannuronate is a marker that the sample under study is an alginate compound.

Table 1. FTIR Sodium Alginate

Wave number ( $\text{cm}^{-1}$ )	Interpretation of functional groups
3.310	Hydroxyl group (OH)
1.625	Carbonyl group (C=O)
1.216	Carboxyl group (COOH)
1.523	Na in alginate

### **Effect of Addition of Alginate to the pH Brown Rice Milk**

Milk has nutritional content and contains various benefits that are needed by the body to be carried out storage that can maintain the conditions and nutrients present in milk. Milk storage capacity or known as shelf life can be defined as the time of production and packaging of a product with the point of time at which the product becomes fit for consumption. According to Sumaprastowo

(2000) storability is always expressed by the environmental conditions that are used to store a material, whether food, drink, or other objects. A good storage is a system that can be regulated in conditions such as room temperature so that it can inhibit the growth of microbes in food and drinks.

The effect of adding alginate to brown rice milk was also seen on the pH value in the composition of brown rice. The following is the pH value of brown rice milk with various variations in the composition of the addition of alginate in milk with a storage temperature of 4°C. An analysis of the effect addition of alginate on the pH was carried out at room temperature. The following is the pH data of the brown rice milk.

Based on figure 6, it appears the pH value of the composition of brown rice milk from various variations of the addition alginate is constant until the 15 days to various compositions of adding alginate to milk. In the variation of the addition of 1.0% and 1.2% alginate has the best pH and managed to maintain the stability of milk at a neutral pH that is at pH 7.00. The highest pH is the addition of 0.4% alginate in the composition of brown rice milk which is at a pH of 7.04. The addition of 0.8% alginate in the composition of brown rice milk also has a pH value that manages to maintain the quality of milk at pH 7.01.

The decrease in pH of brown rice milk during storage is caused by the acid production that is increased of producing bacteria from day to day. During storage is profitable which will improve product quality to opportunities for microorganisms to move mainly bacteria. The stability of the pH value is influenced by the presence of edible film factors that found in brown rice milk.

Edible film functions is a barrier against mass transfer (oxygen, lipids and other solutes). The fatty acids is contained in the sample fully oxidized because which are blocked by edible film so the acidification process is not optimal.

### **Effect of Addition Alginate to the Stability Brown Rice Milk**

The effect of alginate enhancers into brown rice milk was carried out by measuring the turbidan value of brown rice milk by using

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a UV-vis spectrophotometer at  $\lambda = 241 \text{ nm}$ . Sedimentation rate of brown rice milk with various variations of alginate concentration was tested by kinetics using the integral method in order 0, 1 and 2. Based on the turbid

value obtained then graphed the value of the turbidan on the y axis with respect to time (t) on the x axis. Of the three types of graphs obtained the highest  $R^2$  value is in order 1.

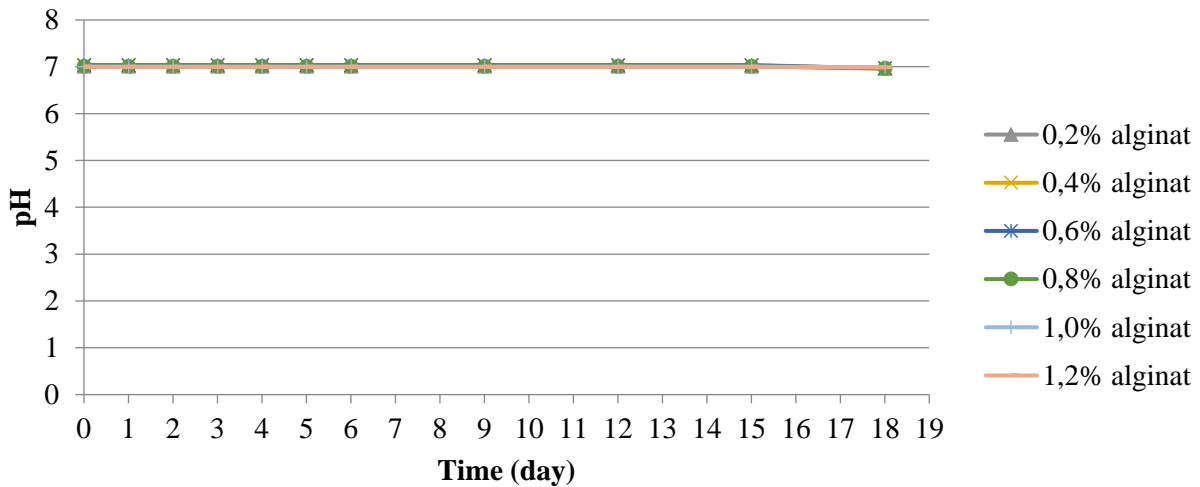


Figure 5. Effect of variations in the addition alginate on the pH with a storage temperature of 4°C

The sedimentation rate of the effect addition alginate is in order 1. This shows that alginate is able to increase the stability of brown rice milk so that the brown rice milk produced does not quickly settle or separate into 2 phases.

Alginate interacts with brown rice which is dispersed in the liquid phase so that the brown rice milk is maintained its stability. Based on the plot of the graph Ln S to get the constant rate of deposition of each milk sample.

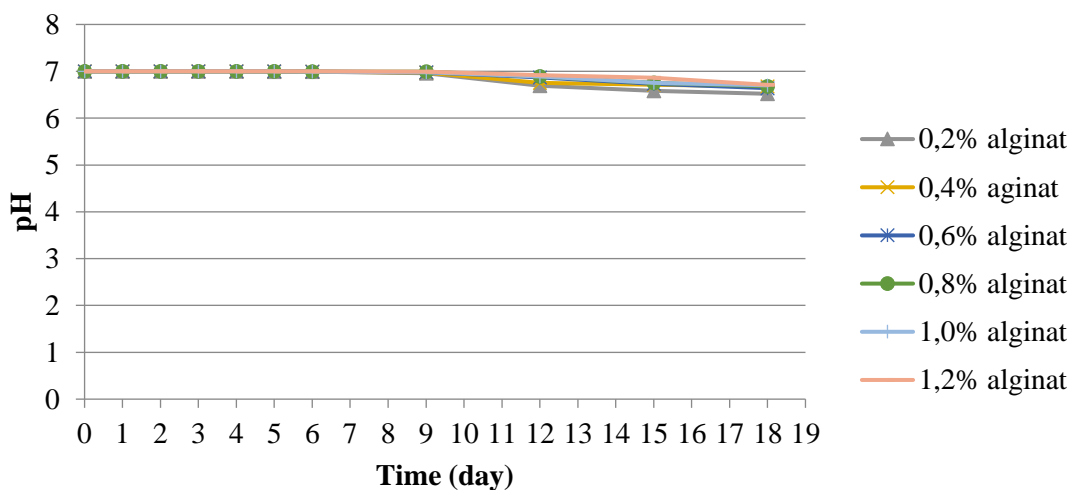


Figure 6. Effect of variations in the addition alginate at room temperature storage

Based on the Table 2, it can be seen that the highest rate of deposition constant is in the addition alginate with a concentration of 0.05%. Whereas the addition of 1.2% alginate has the lowest deposition rate constant. This shows that the higher the concentration of alginate added to milk, the stability of brown rice milk is maintained. From the data half-life of milk shows that brown rice milk has the

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highest stability that is 40 days 12 hours. The stability of brown rice milk with the addition alginate produces more stable milk quality than without the addition of alginate.

In the emulsion system, there is a difference in the boundary field stresses between the dispersed phase and the continuous phase in unmixed soymilk. The

voltage occurs between two phases is called the boundary plane voltage. The higher voltage differences that occur in the boundary plane cause the two phases to be more difficult to mix. The addition alginate can reduce the surface tension that occurs in the boundary

plane so that the two phases will be easily mixed (Kurniasari and Fithri, 2010). The higher surface tension in an area will cause two different phases to be difficult to mix (stable) due to the formation of new surfaces.

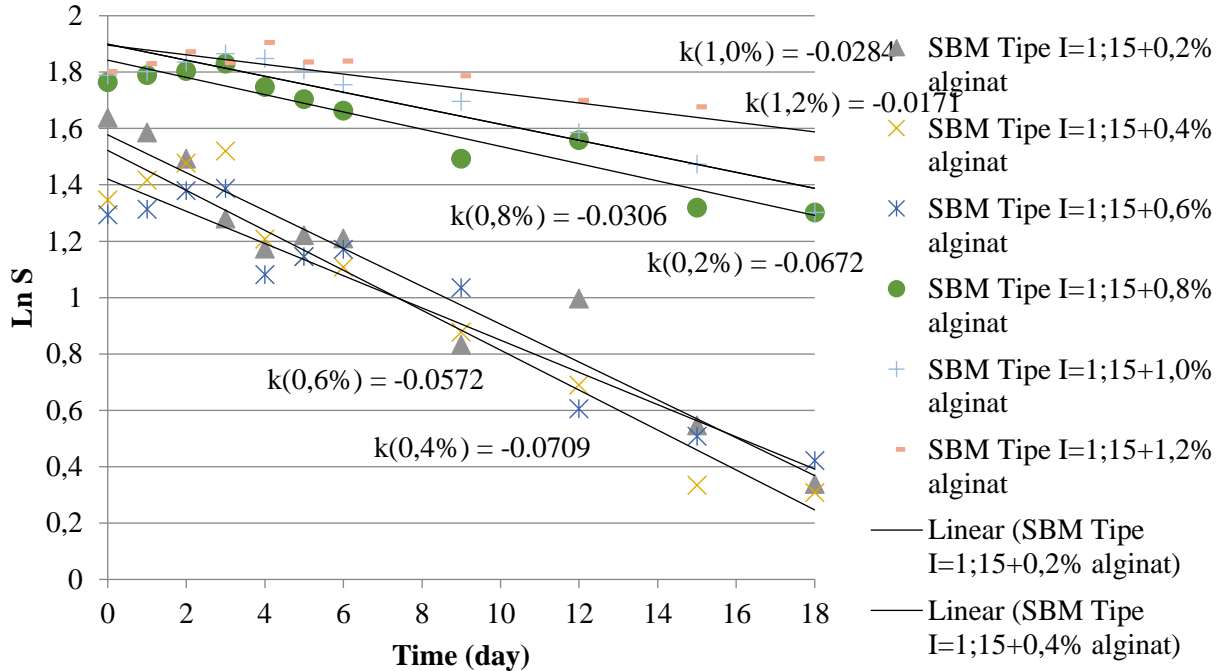


Figure 7. Plot graphic variations of the addition alginate to the stability of brown rice milk in order-1

Table 2. Rate and the half-life of the effect of addition alginate to the rate of precipitation of milk

Types of Milk	k (days <sup>-1</sup> )	Half time
SBM tipe I = 1:15+0,2% alginat	0,0672	10 days 7 hours
SBM tipe I = 1:15+0,4% alginat	0,0709	9 days 16 hours
SBM tipe I = 1:15+0,6% alginat	0,0572	12 days 2 hours
SBM tipe I = 1:15+0,8% alginat	0,0306	22 days 14 hours
SBM tipe I = 1:15+1,0% alginat	0,0284	24 days 19 hours
SBM tipe I = 1:15+1,2% alginat	0,0171	40 days 12 hours

**Degradation Analysis of Brown Rice Milk Dyes**

Determination of the maximum wavelength of brown rice milk was carried out at a wavelength scan ( $\lambda$ ) at 400-700 nm. The

following is a wavelength scan of a sample brown rice milk.

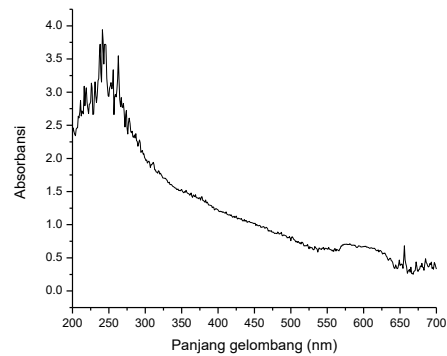


Figure 8. Wavelength of brown rice milk

From the figure 8, it appears that the maximum wavelength obtained from brown rice milk at wavelength 656 nm with an absorbance of 0.48.  $\lambda$  at 656 nm shows the anthocyanin pigment content contained in brown rice milk. The anthocyanin pigment is in the range of wave length 510-700 nm. Anthocyanins are a class of flavonoid compounds, which are the largest group of natural pigments in red, water-soluble browns

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which are responsible for giving red rice a red color.

Dyestuff degradation was analyzed by measuring the absorbance value of the milk by UV-vis spectrophotometer at 656 nm. Kinetic analysis to determine the rate of degradation dyes from brown rice milk by using the kinetic analysis of the integral method based on the law of the rate equations in order 0.1 and 2. The rate equations are used:

Orde 0:  $A - A_0 = -kt$  (2)

Orde 1:  $\ln A - \ln A_0 = -kt$  (3)

Orde 2:  $\frac{1}{A} - \frac{1}{A_0} = kt$  (4)

Information : A = sample absorbance  
 k = rate constant  
 t = time

Kinetic analysis of the dyes degradation rate on brown rice milk samples with the addition alginate. Based on the results of the reaction kinetics, the degradation rate of brown rice milk dyes is in order 0. This shows that alginate can maintains the stability and resistance of brown rice milk. This means that the addition alginate does not affect the degradation of milk sample dyes or inhibit the rate precipitation of brown rice milk as shown below.

Based on the constant rate of deposition brown rice milk samples with the addition of 1.2% alginate which is 0.0022 is the best milk sample because it successfully suppresses the degradation rate of dyes up to 95.79% compared to those without the addition alginate.

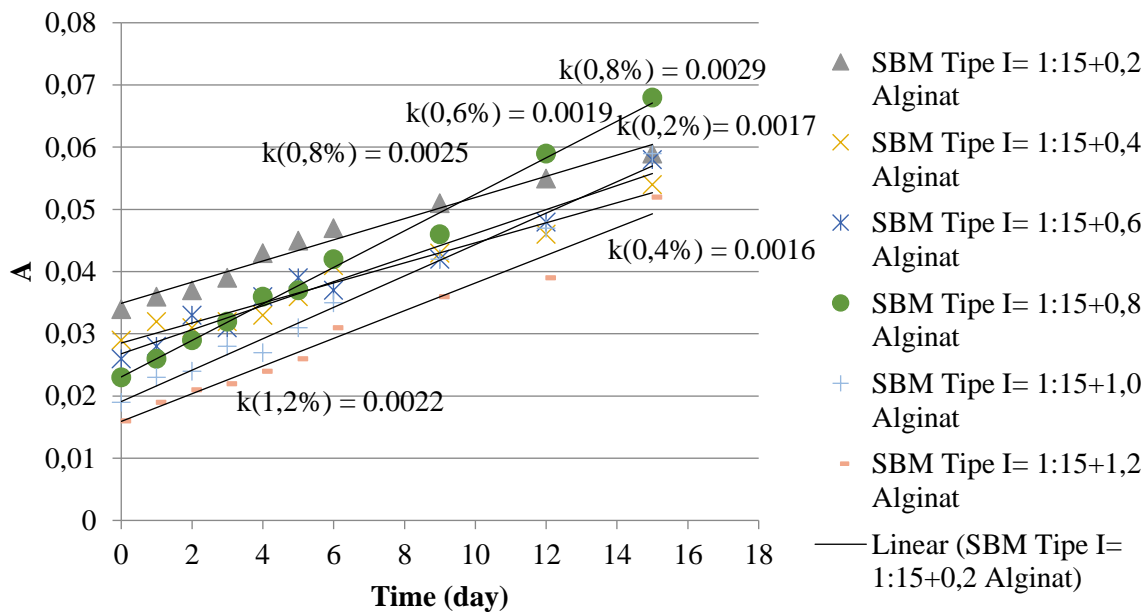


Figure 9. Plot graph degradation rate kinetic of brown rice milk dyes with the addition of alginate in order-0

Table 3. The constanta of the degradation rate of milk dyes and the half-life of the effect of addition alginate to the precipitation rate of milk

Types of Milk	k (Abs/days <sup>-1</sup> )	Half time
SBM tipe I = 1:15+0,2% alginat	0,0017	7 days
SBM tipe I = 1:15+0,4% alginat	0,0016	9 days 1 hours
SBM tipe I = 1:15+0,6% alginat	0,0019	6 days 19 hours
SBM tipe I = 1:15+0,8% alginat	0,0029	3 days 21 hours
SBM tipe I = 1:15+1,0% alginat	0,0025	3 days 19 hours
SBM tipe I = 1:15+1,2% alginat	0,0022	3 days 14 hours

Based on showing that the degradation rate of milk dyes has decreased to order 0. This shows that alginate is able to maintain the stability of the dyes of brown rice milk. Then determined the constant rate of deposition of milk and half-life of brown rice milk.

From the table 3 shows that the deposition rate of milk is in order 0, where this shows that alginate can inhibit the degradation of brown rice milk dyes. The half-life data shows that the most stable brown rice milk is the addition of 1.2% (w/v) alginate with a half-life of 3 days 14 hours.

**Analysis of Microbial Number of Brown Rice Milk by Addition Alginate**

Microbial analysis of brown rice milk was carried out by a dilution method. The medium used is LB medium with incubation temperature of 4°C and room temperature. Brown rice milk in the image incubated at 4°C did not show any microbes that appeared. Then the milk was observations from day to day on the number of microbes that appear in milk. From observations up to the 15<sup>th</sup> day milk is still in a safe condition and free from microbial contamination. But milk incubated at room temperature appears as microbes appear.

The table 4 in below shows the presence of microbial activity during storage at room temperature. The number of bacteria produced increases from day to day. But from the data obtained the number of microbes that appear in milk samples has not exceeded the maximum limit of microbial contamination as required by SNI 7388 (2009). Where the maximum amount of microbial contamination in milk samples is 5x10<sup>4</sup> cfu/mL. This shows that milk with storage at room temperature has a resistance of up to 3 days.

Table 4. Analysis of the number of incubation microbes at room temperature

Sample type	The number of microbes (cfu/mL)		Average number of microbes (cfu/mL)
SBM Tipe I = 1:15	10,2 x 10 <sup>2</sup>	10,3 x 10 <sup>2</sup>	10,25 x 10 <sup>2</sup>
SBM Tipe I = 1:15 + 0,2% alginate	9,4 x 10 <sup>2</sup>	9,4 x 10 <sup>2</sup>	9,40 x 10 <sup>2</sup>
SBM Tipe I = 1:15 + 0,4% alginate	9,6 x 10 <sup>2</sup>	9,5 x 10 <sup>2</sup>	9,55 x 10 <sup>2</sup>
SBM Tipe I = 1:15 + 0,6% alginate	7,2 x 10 <sup>2</sup>	7,3 x 10 <sup>2</sup>	7,25 x 10 <sup>2</sup>
SBM Tipe I = 1:15 + 0,8% alginate	6,8 x 10 <sup>2</sup>	6,4 x 10 <sup>2</sup>	8,6 x 10 <sup>2</sup>
SBM Tipe I = 1:15 + 1,0% alginate	6,1 x 10 <sup>2</sup>	6,2 x 10 <sup>2</sup>	6,15 x 10 <sup>2</sup>
SBM Tipe I = 1:15 + 1,2% alginate	4,3 x 10 <sup>2</sup>	4,4 x 10 <sup>2</sup>	4,35 x 10 <sup>2</sup>

**CONCLUSION**

*Sargassum binderi* as alginate can increase the stability of brown rice milk. Besides being able to extend milk endurance and prevent acidification at the milk pH. The number of microbes that appear in milk samples has not exceeded the maximum limit of microbial contamination as required by SNI 7388.

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## CHEMICAL AND SENSORY CHARACTERISTIC OF SORGHUM (*Sorghum bicolor*) TAPAI WITH TRADITIONAL PACKAGING

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### ABSTRACT

Tapai is one of the traditional food from Java island. Nowadays, traditional food is being ignored by people. Conserving traditional foods are needed. Processing into a variety of foods that has good nutrition can make traditional food exist. Sorghum (*Sorghum bicolor*) is a good source of carbohydrates, fibers, phenolic compounds, and antioxidant. However, it has not been fully utilized. Bitter taste, sandy texture and unpleasant aroma in sorghum cause people do not like it. Therefore, this study aimed to develop tapai from sorghum. This study will focus on the evaluation of starch content, pH, acidity content, sugar content and sensory evaluation tapai made from varieties of sorghum and types of natural packaging. The method used in this study was the experimental method using a completely randomized factorial design consisting of two factors, variable of sorghum (white, red and brown sorghum) and types of natural packaging (banana leaf, pandanus leaf, and guava leaf) with two replications. Duncan's multiple range tests were performed with a 5% significance for statistical analysis. Results revealed that the highest starch content was 27,4% in tapai from brown sorghum with banana leaf packaging. Regarding sugar content, tapai from brown sorghum with banana leaf packaging had the highest result. Whereas, based on sensory evaluation, tapai from white sorghum with banana leaf packaging was the most favored by panelists.

**Keywords:** sorghum, tapai, traditional food, packaging

### INTRODUCTION

Lately, the popularity of traditional foods has increased. The rise in food start-up events is one that can create variations of traditional food preparations. According to Guerrero (2010), traditional food is a food product that is often consumed by a group of people or served in certain celebrations and times, passed down from generation to generation, made with hereditary recipes, and has certain characteristics that are different from other regional culinary delights.

Tapai is a traditional food that is popular in the Southeast Asian region, not least in Indonesia (Law, et al. 2018). Tapai is produced from the fermentation process of carbohydrate with the help of yeast (Berlian, et al., 2016).

Tapai as one of the fermented foods has a distinctive taste. Sweetness, acid, and aromas that arise are the result of the breakdown of carbohydrate components into glucose, organic acids, and alcohol by the activity of yeast during fermentation (Barus and Wijaya, 2011). In Indonesia, there are at least some well-known types of tapai namely white sticky tapai, black sticky tapai, cassava tapai (Peuyeum) and Kuningan tapai.

In general, the tapai is made with banana leaf packaging. However, in West Java, Kuningan regency, tapai is packed with guava leaves. The two types of tapai have a distinct aroma. Different types of packaging are thought to cause the tastes of the two tapai to be different. According to Svensson (2004),



volatile and non-volatile compounds in packaging can migrate from packaging to products. This is also supported by the statement of Hernandez and Gavara (1999) that during packaged food, the mass transfer can occur from and to packaging such as water vapor migration, aroma absorption by packaging and absorption of packaging odor by food.

Sorghum (*Sorghum bicolor* L.) is a cereal plant of the Poaceae family that grows in tropical and subtropical regions in the southeastern Pacific and Australia. By the Javanese people, this plant is better known as cantel. The results showed that sorghum can be processed into various products such as rice, tempeh, white bread, cookies and noodles (Suarni, 2016). However, it is still rare to find processed sorghum products that are circulating in the community. The bitterness and taste of sandy texture cause sorghum less in demand by the people (Berlian, 2016) (Schober et al, 2007). Whereas sorghum has the potential as a functional food because it has been shown to have antioxidant activity and contains polyphenols and tannins (Jeon et al, 2017). Therefore, sorghum has the potency to be processed into a very large variety of products.

Previous research has been carried out on tapai such as the physical and chemical characteristics of onggok tapai (Fahmi, N. and Nurrahman, 2011), sensory characteristics of purple yam tapai with different yeast doses (Handayani, 2013), and the influence of cassava types and fermentation time of cassava tapai (Dirayati, 2017). The results showed that the type of starch source, fermentation time and yeast concentration resulted in the characteristics of the tape with different flavors (Handayani, 2013, Dirayati, 2017). However, research has never been done about the use of sorghum as the main ingredient in making tape. Therefore, our research was conducted on sorghum tape. Besides, the study also carried out an analysis of the effect of the use of traditional packaging of banana leaves, guava leaves and pandanus leaves on the chemical characteristics produced.

## **MATERIALS AND METHODS**

### **Material**

The main ingredient was white, red, and brown sorghum obtained from the Organic

Shop Florensia, Surabaya. Commercial yeast tapai, banana leaves, guava leaves, and pandanus leaves were obtained from the Sopyonyono market, Surabaya. Chemicals such as aquades, Na bisulfite, KI,  $\text{Na}_2\text{CO}_3 \cdot 10\text{H}_2\text{O}$ ,  $\text{Na}_2\text{CO}_3$ , phenolphthalein indicator (PP), obtained from Bratachem stores, NaOH, HCl,  $\text{H}_2\text{SO}_4$  obtained from Merck.

## **Methods**

### **Sorghum tapai production**

300 grams of each white, red, and brown sorghum were cleaned and then soaked in water for 12 hours with the ratio of sorghum and water (w/v) 1:5. Furthermore, sorghum was cooked in a pressure cooker at  $100^\circ\text{C}$  for 20 minutes, then cooking was continued in a steamer for 15 minutes, then cooled at room temperature, then weighed.

One percent of yeast tapai was mixed sorghum (w/w). (Berlian, 2016). The fermentation was conducted in banana leaves, guava leaves, and pandanus leaves packaging. Fermentation was carried out for 48 hours at  $30^\circ\text{C}$  in an incubator. The fermented tapai was analyzed for the starch content, total sugar, total acid, pH, sensory evaluation (preference test).

### **Starch Content**

#### **Sample Preparation**

0.1 gram sample was weighed in a 250 mL Erlenmeyer, 50 mL of aqua dest was added, and 5 mL of 25% HCl, then heated at  $100^\circ\text{C}$  for 3 hours. After being cooled, the suspension was neutralized with 25% NaOH to pH 7. Transfer quantitatively in a 100 mL flask, then adjust until the mark sign with distillate water. This solution is then filtered with filter paper.

#### **Sample analysis**

A total of 25 mL of filtrate plus 25 mL of Luff Schoorl solution in Erlenmeyer (25 mL of Luff Schoorl solution with 25 ml of distilled water) were prepared. Erlenmeyer was connected to the return cooler, then bring to a boil. Boiling the solution was maintained for 10 minutes. Subsequently quickly cooled and added 15 mL 20% KI and carefully added 25 mL 25%  $\text{H}_2\text{SO}_4$ . Then closed and placed in a dark place for 30 minutes. The released iodine was titrated with 0.1 N  $\text{Na}_2\text{S}_2\text{O}_3$  solution using a starch indicator of 2-3 mL. To clarify the color change at the end of the titration, it

was advisable to provide starch when the titration was almost over.

### **Calculation of Starch Content**

Starch content obtained by calculating the difference between standard treatment and sample titration, reducing sugar levels after inversion (after being hydrolyzed with 25% HCl) in ingredients can be searched using the inverse sugar difference table before inverse multiplied by 0.9.

$$\text{Starch content (\%)} = \frac{\text{mg glucose} \times \text{FP} \times 0,9}{\text{mg starch sample}} \times 100\%$$

Note:

mg glucose = number in table of *Luff Schoorl*, based on the mL difference of titration

FP = mL filtrated of titrate

### **Analysis of the Total Sugar Anthrone Method (Morris, 1948)**

20 grams of tapai was put into the beaker glass, then 20 mL of water was added. The tapai then crushed by *waring blender* until all the sugar was extracted, and the tapai crumb was transferred to another glass cup quantitatively and then heated to a 100 ° C water bath for 30 minutes. After cooling, the sample was filtered using Whatman filter paper No. 2. The sample was then added 1.5-2.5 mL of saturated acetate Pb solution, then filtered again. 50 mL of the filtrate then added 2.5 grams of dry Na-oxalate.

Standard glucose solution series of 0.2, 0.4, 0.6, 0.8, and 1.0 mL were prepared into the test tube then diluted up to the total volume was 1.0 mL. 1 ml of distilled water was prepared as a blank solution. Then, 5 mL of each Anthrone reagent was added to each standard glucose solution and blank, then vortexed and heated on a 100 ° C water bath for 12 minutes. the absorbance samples were measured with a UV-Vis spectrophotometer at a wavelength of 630 nm, then a plot of data was made between glucose levels and absorbance on a standard curve.

Samples were analyzed by inserting 5 mL of filtrate tapai into a 100 ml measuring flask, then diluted to the mark with distilled water. 1 mL of the sample was put into a closed test tube, then 5 ml of Anthrone reagent was added quickly, then vortexed and heated on a 100 ° C water bath for 12 minutes. After

cooling, the absorbance samples were measured by a UV-Vis spectrophotometer at a wavelength of 630 nm.

### **Total acid content**

Total acid content based on acid levels equivalent to lactic acid levels (Harjiyanti, 2013). Samples were crushed then filtered. 10 mL of filtrate was added with phenolphthalein indicator then titrated with 0.1 N NaOH until finished.

$$\text{Acid Content} = \frac{V1 \times N \times B}{V2 \times 1000} \times 100\%$$

Note :

V1 = volume of NaOH (mL)

V2 = volume of sample (mL)

N = normality of NaOH (0,1N)

B = molecular weight of lactic acid (90)

### **pH measurements**

10 g of the sample were crushed then added 10 mL (1:1) of aquades. The solution was measured by the pH using a pH meter.

### **Sensory Evaluation (hedonic test)**

Sensory evaluation was referred to Sarlina et al (2017). Determination of the tapai favored by the panelists of each treatment, organoleptic assessments were carried out on the quality of the product which included color, aroma, and texture. This test was based on giving panelists a score on quality in terms of color, flavor, aroma, and texture. The number of panelists was 30 people. Rating scores given based on organoleptic assessment criteria were 5 (very like), 4 (likes), 3 (quite likes), 2 (fewer likes) and 1 (dislikes).

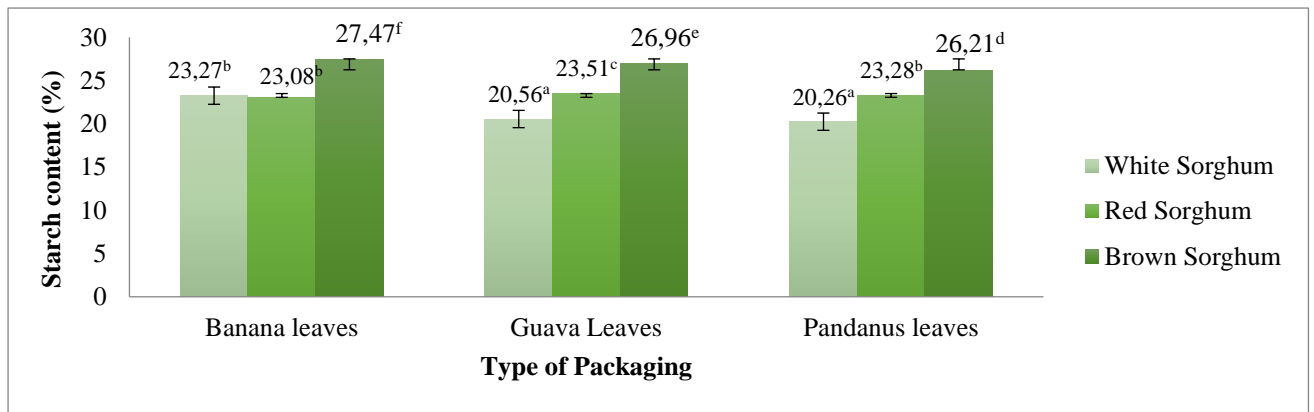
### **Data Analysis**

The data obtained were analyzed with ANOVA (Analysis of Variance) with a confidence level of 5%. If there are significant differences then proceed with the Duncan Multiple Range Test (DMRT).

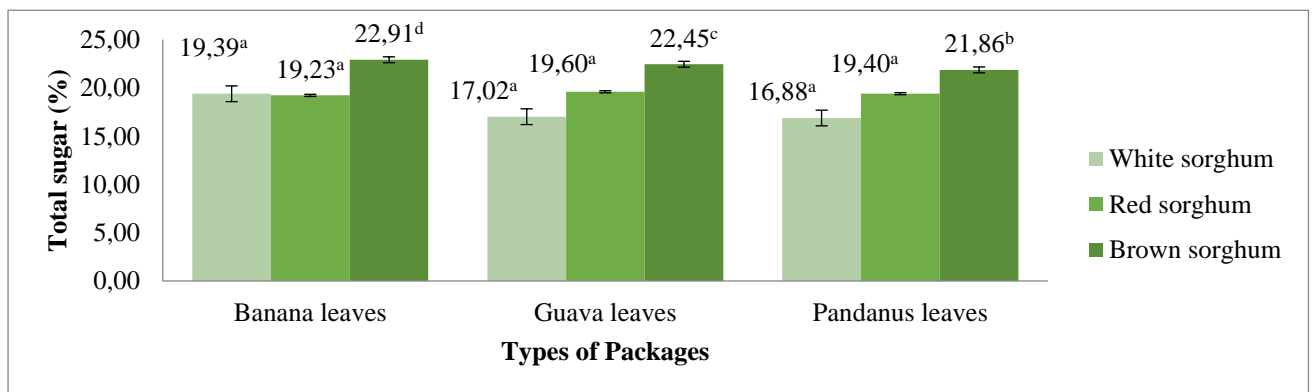
## **RESULTS AND DISCUSSION**

### **Starch content**

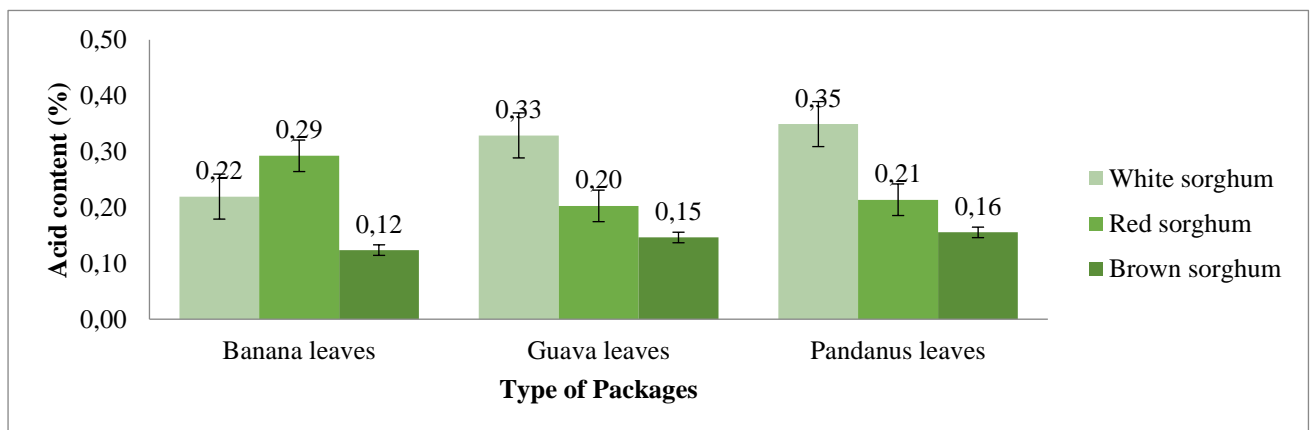
Test of starch content in white, red, and brown sorghum fermented with banana leaves, guava leaves, and pandanus leaves are presented in Figure 1. The results (Figure 1) showed that each sorghum produces a different level of starch after the fermentation process. Sorghum and package significantly influenced starch content ( $p \leq 0.05$ ). The highest of starch content was found in brown sorghum tapai package



**Figure 1.** The relationship between the treatment of sorghum type and type of packaging on the starch content (%) of the sorghum tapii



**Figure 2.** The relationship between the treatment of sorghum type and type of packaging on the total sugar (%) of the sorghum tapii



**Figure 3.** The relationship between the treatment of sorghum type and type of packaging on the total acid (%) of the sorghum tapii

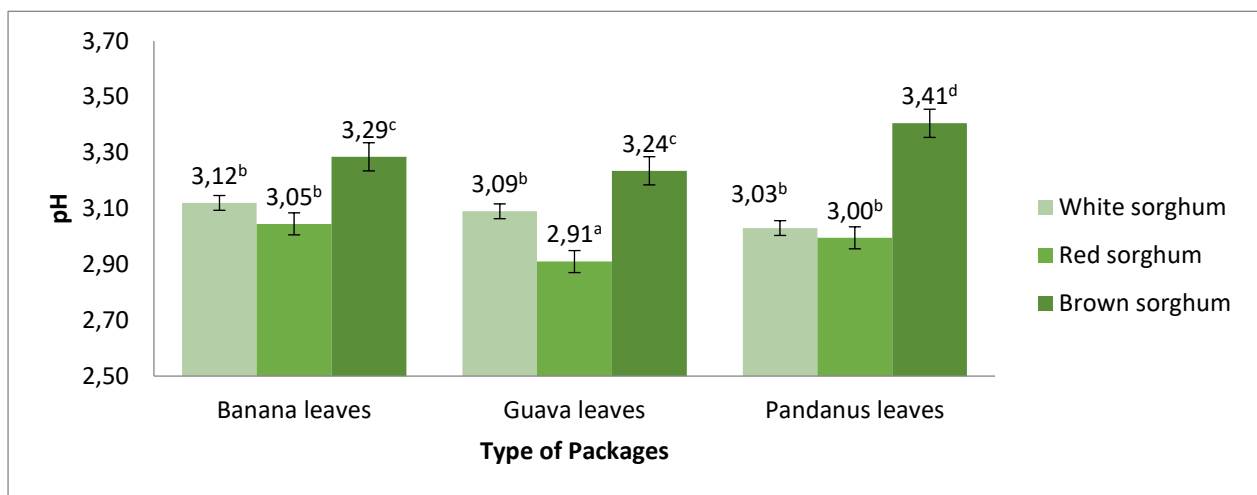
with banana leaves (27,47%), whereas the lowest was white sorghum tapii package with pandanus leaves (20,26%).

According to Boudries *et al*, (2014), the starch content of white sorghum was  $66.81 \pm 0.27\%$  and red sorghum  $65 \pm 0.11\%$ . Starch was used by yeast as the main ingredients of fermentation. Therefore, starch content decrease after the fermentation process. The fermentation process converted starch into

alcohol and organic acids. This was appropriate with Berlian (2016), the cassava fermentation process causes starch levels to decrease after the fermentation process. Microbes in tapii play a role in the fermentation process by remodeling glucose into alcohol. Yeast generally consists of populations of genera *Aspergillus*, *Saccharomyces*, *Candida*, *Hansenulla*, and *Acetobacter* bacteria (Oktaviana, *et. al* 2015).

**Total Sugar**

The total sugar profile presented in Figure 2. Both sorghum and packaging were significantly influenced total sugar after the fermentation process ( $p \leq 0.05$ ). The highest total sugar was brown sorghum tape in banana leaf packaging (22.9%), while the lowest total sugar was the white sorghum tapi in pandanus leaves. Utami and Noviyanti (2010) stated that in making tapi the hydrolysis stage was represented by the boiling stage. In the process of hydrolysis occurs the addition of water molecules in the breakdown of starch. So, the higher the starch content, the more water is absorbed by the hydrolysis process.



**Figure 4.** The relationship between the treatment of sorghum type and type of packaging on the pH of the sorghum tapi

In addition to the production of glucose during fermentation, stachyose (a tetrasaccharide containing glucose, fructose, and two galactose units) is also produced during the hydrolysis of starch. Other oligosaccharides such as maltoheptaose, maltohexaose, maltopentaose (highest sugar monomer), maltotetraose and isomaltotriose (lowest sugar monomer) may be produced too (Azmi and Mel, 2014). The result was similar to that reported by Asnawi *et al*, (2013), who found the value of 18-21% for cassava tapi.

Yeast has a role in the formation of reducing the sugar by hydrolyzing starch to sucrose (maltose) then convert to monosaccharide (glucose and fructose), then convert to alcohol, organic acid and the other compounds (Nuraida dan Owens 2014).

**Total Acid**

Total acid indicates the acidity of a product. In this study, the highest acid content was found in white sorghum tapi with pandanus leaves packaging (0.35%), while the lowest acid content was found in brown sorghum tapi with banana leaves packaging (0.12%). The acids formed were the result of changes from sugars that were converted into alcohol and subsequently undergo changes to organic acids such as lactic acid and acetic acid.

According to Dirayati (2017), the cassava tapi fermentation process begins with the conversion of starch in cassava by the amylase enzyme released by microbes into maltose. Maltose can be converted into glucose by the enzyme maltase. Glucose by the enzyme zymase is converted into alcohol. Furthermore,

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alcohol can be converted to acetic acid, pyruvic acid, and lactic acid by the enzyme alcoholase. The formation of acetic acid, pyruvic acid and lactic acid due to the presence of *Acetobacter* bacteria that are often present in yeast.

Organic acids from alcohols form aromatic esters so that the tape has a distinctive taste. Also, yeast especially *Saccharomyces cerevisiae* can grow at pH 3,5-6,5 well (Oktaviana, 2015).

Prakoso and Santoso in Novianti and Sulandri (2014) in their research stated that the fermentation process in making breadfruit

tapai converted native breadfruit to the soft texture, sour taste, have a sweet aroma and color changes. The sweet taste on the tapai occurred due to the change of carbohydrates into glucose as a simpler carbohydrate, while the sour taste because of the acid in the fermentation process, so fermentation process will increase alcohol levels and total acid content (Fahmi and Nurrahman, 2011).

This is according to the research of Buckle *et al.* (1987) that pyruvic acid is a product formed in the hydrolysis of glucose into ethanol. Pyruvic acid can be converted into ethanol and lactic acid.

Table 1. Organoleptic test

Treatment		Parameter		
Sorghum type	Packaging type	Taste	Aroma	Texture
White sorghum	Banana leaves	2,6 <sup>fg</sup>	4,64 <sup>a</sup>	4,32 <sup>a</sup>
	Guava leaves	2,36 <sup>gh</sup>	3,76 <sup>a</sup>	4,16 <sup>a</sup>
	Pandanus leaves	2,72 <sup>de</sup>	4,24 <sup>a</sup>	4,08 <sup>a</sup>
Red sorghum	Banana leaves	3,08 <sup>ab</sup>	4,12 <sup>a</sup>	3,6 <sup>ab</sup>
	Guava leaves	2,96 <sup>bc</sup>	3,6 <sup>bc</sup>	3,24 <sup>b</sup>
	Pandanus leaves	3,16 <sup>a</sup>	3,64 <sup>ab</sup>	3,28 <sup>b</sup>
Brown sorghum	Banana leaves	2,68 <sup>f</sup>	3,2 <sup>cd</sup>	2,72 <sup>ab</sup>
	Guava leaves	2,76 <sup>cd</sup>	2,92 <sup>d</sup>	2,72 <sup>ab</sup>
	Pandanus leaves	2,32 <sup>h</sup>	2,76 <sup>d</sup>	2,36 <sup>c</sup>

Note: Different letters in the same column indicate significantly different.

### pH value

The pH profile presented in Figure 4. Both sorghum and packaging were significantly influenced pH value ( $p \leq 0.05$ ). The highest pH was brown sorghum tape in pandanus leaves (3,405), while the lowest pH was the red sorghum tapai in guava leaves. The result was lower than that reported by Asnawi *et al.*, (2013), who found the pH value of 5,1 for cassava tapai. The formation of organic acids causes the pH of sorghum tapai to decrease (Sujaya *et al.*, 2001).

Changes of pH in fermentation are caused by the activity of yeast cells in addition to producing ethanol as a primary metabolite also produces acids such as malic acid, tartaric acid, citric acid, lactic acid, acetic acid, butyric acid as a by-product. These acids reduce the pH of the medium (Oktaviana, *et al.*, 2015). The result was corresponding to total acid that

brown sorghum tapai has the lowest of total acid.

### Organoleptic (Hedonic) Test

The hedonic test is an organoleptic sensory analysis used to determine the magnitude of the quality difference between several similar products by giving an assessment or score of certain properties of a product and to determine the level of liking of a product. This level of preference is called the hedonic scale, for example very like, like, rather like, rather dislike, dislike, very dislike and others (Stone and Joel, 2004). In this study, there were three attributes tested, namely taste, aroma, and texture.

Taste is one of the important attributes in organoleptic testing of food products. The results of the research showed that the highest value of the taste parameter was the pandanus leaf packaging in red sorghum tapai with a value of 3.16 (likes), whereas the pandanus leaf packaging in brown sorghum tapai had the

lowest value of 2.32 (somewhat dislike). The red sorghum tapai packaged with pandanus leaves has a sweet and slightly sour taste.

The taste of sorghum tapai was influenced by the results of fermentation which produces sugar, alcohol, organic acids and carbon dioxide. According to Astawan (2004), the basic principle of starchy food fermentation is the degradation of starch components into dextrans and sugars, which are then converted to alcohol or acid to produce fermented foods that have a sweet, alcoholic and slightly acidic or slightly acidic taste.

This is supported by the statement of Fahmi and Nurrahman (2011) which states that the appropriate time will produce a tape that tastes distinctive, sweet taste with a little acid and the smell of alcohol. The sweet taste due to changes in carbohydrates into glucose as a simpler carbohydrate, while the sour taste because in the fermentation process is formed acid, so the longer the ripening there will be an increase in alcohol content and total acid.

The aroma is the sensation of smell received by the sense of smell when food is put in the mouth. The aroma is very influential on the level of preference of panelists because a good aroma can improve the assessment of the organoleptic properties of a food product. Aroma tapai is caused due to their presence of volatile compounds derived from these products, such as organic acid, alcohol, glucose (Handayani, 2013). The white sorghum tapai with banana leaf packaging had the highest score of 4.65 (likes), while the lowest score was the brown sorghum tapai with pandanus leaf packaging.

The results obtained from the texture parameters showed that all samples have a soft texture. However, panelists gave different values. The white sorghum tapai with banana leaf packaging had the highest value of 4.32 (likes), while the lowest value was the brown sorghum tapai with pandanus leaf packaging which was 2.36 (somewhat dislike). White sorghum tapai generally had the highest average texture compared to red and brown sorghum tapai. Allegedly the white sorghum tapai had a soft texture. Following Hasanah et al, (2012), the fermentation process produces sugar, alcohol, acid and also causes soft texture.

White sorghum tapai with banana leaf packaging was the best treatment sample with an average value of 3.87. The tapai had a sweet, slightly acidic acid, the aroma of alcohol was not too sharp, and the texture was soft. *L. plantarum* is a group of bacteria that produce large amounts of lactic acid as the result of sugar (carbohydrate) metabolism. Lactic acid produced will reduce the pH value of the growth environment and cause a sour taste. The mixture of sour taste from lactic acid, sweetness from the result of sugar degradation, and the presence of alcohol due to the activity of yeast *S. cerevisiae* which converts sugar greatly determine the taste of tape so that it has specific characteristics (Barus and Wijaya, 2011).

## CONCLUSION

Results revealed that the highest starch content was 27,4% in tapai from brown sorghum with banana leaf packaging. Regarding sugar content, tapai from brown sorghum with banana leaf packaging had the highest result. Whereas, based on sensory evaluation, tapai from white sorghum with banana leaf packaging was the most favored by panelists.

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# AMYLOGRAPHY PROFILE AND MICROSTRUCTURE OF BENENG TARO BANTEN (*Xanthosoma undipes* K. Koch) STARCH

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## ABSTRACT

High demand of flour import for food applications was a big threaten for food security in Indonesia. The utilization of carbohydrate sources from local food plants can be an alternative to overcome this problem. Beneng taro is a local food from Pandeglang, Banten Province that potential to be used as composite flour. The application of food products, Beneng taro can be used as starch. The purpose of this study is to analyze the characteristics of Beneng taro starch on the amylographic profile of the granules and their morphology. The extraction method used is wet extraction using distilled water. The ratio of taro and distilled water used is 1: 3. The optimum formulation (1: 3) was then tested for morphological properties using SEM instrument and amylograph profile using RVA instrument. SEM test results showed uniform starch granules and polygon-shaped granules. The amylographic profile shown a maximum viscosity of 1075cP with a temperature of 92.4oC at 270 seconds, a set back value of 186 cP and a break down of 214 cP. Based on the amylographic profile, beneng taro starch still has many weaknesses (large set back and break down) so it needs to be modified so that it can be applied to food products.

**Keywords:** beneng taro, starch, amylograph profile, microstructure

## INTRODUCTION

An enormous number of Indonesia's food products were made from carbohydrates, such as biscuits, bread, noodles, pasta, and others. However, these products use wheat flour as basic ingredient, which is still imported. This problem can have an impact on the weakness of food security in Indonesia due to the high use of flour. To reduce the use of flour, there is a need for other carbohydrate sources that can substitute flour for the manufacture of various food products. One source of carbohydrates is Beneng taro. It was a local Banten food which has an abundant presence in Juhut Village, Pandeglang Regency. Beneng taro was more applicable in food products in the form of starch.

Starch is a type of carbohydrate that is abundant in nature and can be obtained from various parts of the body of the plant such as

seeds, roots, stems or cereals. Starch is used in the food, cosmetics and pharmaceutical industries. Some starches that are widely used in the food industry are cassava, corn, potatoes and wheat starch (Deka and Sit, 2016). In the farmer group in Juhut Village, Beneng taro is only made into flour and sold outside the region, so there are no data on the characteristics of the starch. The purpose of this study is to analyze the microstructure and amylography profile of Beneng taro starch. Moreover, the result of this study can provide information about the potency of Beneng taro starch in the food industry.

## MATERIALS AND METHODS

The instruments used were Scanning Electron Microscopy (SEM) USA Zeiss EVO M10, set of kitchen tools (knives, cutting boards, basins, buckets), analytical balance,



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strainer, blender, baking pan, and oven for extracting and drying starch, oven, furnace, cup, desiccator, soxhlet, kjeldahl apparatus, UV-Vis spectrophotometer (Perkin Elmer), HPLC UV-Vis (Agilent), chromameter (Minolta CR-300), Rapid Visco Analyzer (Perten).

The main materials used in this study were Beneng taro obtained from Juhut Village, Karang Tanjung District, Pandeglang Regency, Banten Province, NaCl salt, aquades, hexane, H<sub>2</sub>SO<sub>4</sub> (merck), HgO (merck), NaOH (merck), H<sub>3</sub>BO<sub>3</sub> (Sigma Aldrich), HCl (merck), aquabidest, phenolphtalein (Sigma Aldrich), ethanol 95%, amylose (Sigma Aldrich), acetic acid (merck), iodine (merck), and sodium oxalate (sigma Aldrich).

## Methods

### Beneng Taro Starch Extraction

The process carried out is to observe the diameter, length, weight, skin color, and tuber color of Beneng taro. The next step is the extraction of Beneng taro starch begins with washing, sorting, stripping, slicing, soaking in 1% NaCl salt solution, crushing with water for 5 minutes, filtering, settling for 24 hours, starch, and water separation, drying at 50°C for 24 hours, and size reduction. This extraction process was carried out in five attempts and two repetitions. The ratio of talas and water used for extraction is 1:1, 1:2, 1:3, 1:4, 1:5.

### Morphology Characteristic

Morphological structure was analyzed using Scanning Electron Microscopy (SEM) Zeis EVO MA 10 instrument. A sample was cut 2mm x 2mm size and set on bronze visualization cross by double side tape. The sample surface was coated using thin gold layer. The sputter time and current were set on 60 s and 20 mA, respectively. Sample was inserted into SEM instrument and the surface picture was taken using SE (Secondary Electron) detector with working distance (WD) and electron high tension (EHT) voltage were 8.5 mm and 16.0 kV, respectively.

### Functional Characteristic

Sample suspensions were prepared by mixing 3 g starch (dry basis) and water into total weight of 28.0 g. The suspensions were equilibrated at 50°C for 1 minute, then heated from 50 to 95°C at 6°C/min. A holding period at 95°C was conducted before cooling from 95

to 50 at 6°C/min and another holding phase at 50 for 2 minutes. The RVA pasting parameters which consisted of pasting temperature, peak viscosity, breakdown, final viscosity, and setback were computer recorded and reported. Determination of the best treatment based on the highest yield of starch. Data from Nurtiana, *et al* (2019) are used as the basis for determining the best treatment.

## RESULTS AND DISCUSSION

### Morphological characteristic

The morphological result of Beneng taro starch can be seen in Figure 1.

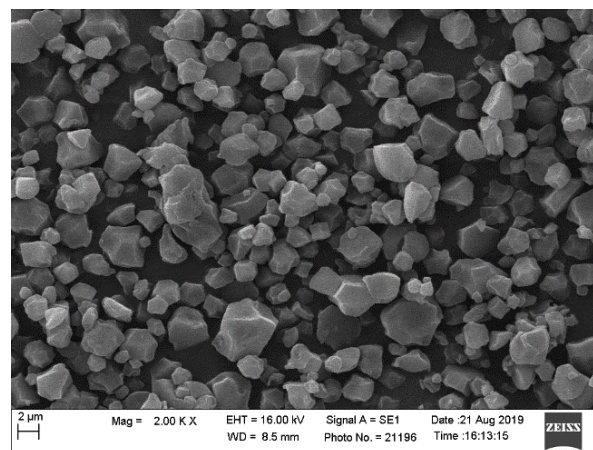


Figure 1. SEM results for beneng taro starch microstructure observation (magnification 2000 times)

Scanning Electron Microscopy results on Beneng Taro Starch showed the form of polygon starch granules. These results are similar to the study of Tattiyakul *et al.* 2006. Other studies even showed a round starch granule (Kartikasari *et al.*, 2016) and irregular (Zeng *et al.*, 2014). Starch granules have a variety of shapes and can be used to identify carbohydrate sources because each source has a different shape. The size of the granules on the Beneng Taro Starch is still relatively large because it has not been modified. In general, starch modification treatment can make starch granules smaller than pure starch granules.

Types of starch granules from various botanical sources vary both in terms of size (diameter ranging from about 0.1 to 200 μm), morphology (ellipsoidal, oval, round, polygonal, elongated, irregular, lenticular, and disk), size distribution (uni-, bi-, or polymodal), and events in amyloplast (individually or as compounds) (Schimer *et al.*, 2014).

**Amylography Profile**

Amylography profile of Beneng taro starch can be analyzed in various ways and can be adapted to the available instruments. Information obtained from the starch amylographic profile is a characteristic of starch paste, including pasting temperature and peak time, minimum viscosity (trough), maximum viscosity (peak), breakdown

viscosity, setback viscosity, and final viscosity. These properties can be measured using Rapid Visco Analyzer (RVA) instrument. Measurement using the RVA instrument requires shorter testing time and a smaller number of samples compared to the Brabender Viscograph (Melissa, 2018). The results of RVA analysis on Beneng taro starch can be seen in Figure 2.

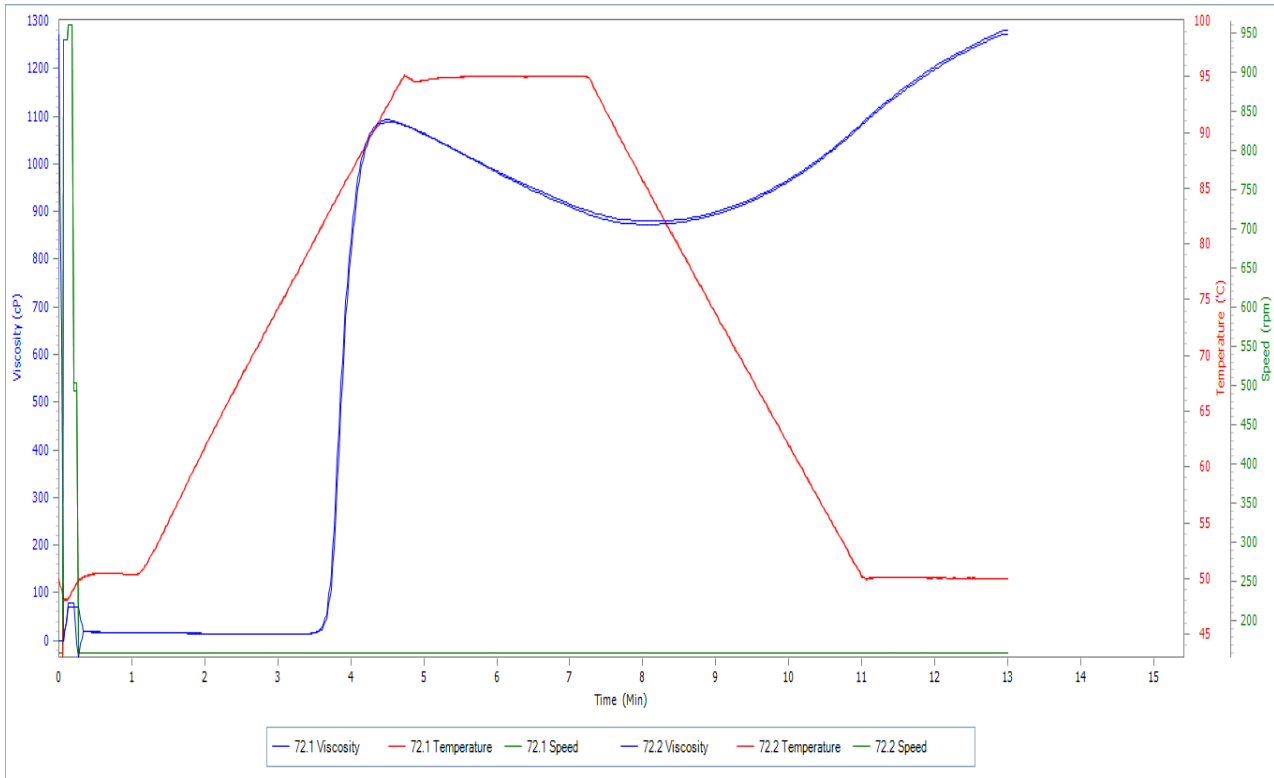


Figure 2. Amylographic profile of beneng taro starch

The results of the Rapid Visco Analyzer on the Beneng taro starch showed a high-temperature gelatinization of 81.9oC, meaning that it needed a temperature high enough for the starch to start gelatinization. The initial temperature of gelatinization or pasting temperature (PT) is the temperature at which viscosity begins to form and indicates starch begins to absorb water (Fetriyuna et al, 2016). The temperature of starch gelatinization is the temperature at which the starch forms a gel completely becomes transparent.

Gelatinization is the process of breaking bonds between starch molecules in the presence of water and heat and allows starch molecules to bind more water. The presence of water penetration will increase between randomness in the structure of starch. The stronger the bonds between starch molecules, the higher the amount of heat needed to break

bonds between molecules so that the higher the gel temperature (Singh-Sodhi and Singh, 2005). The temperature and time of starch gelatinization are influenced by the amylopectin structure, starch composition, granular architecture (Imaningsih, 2012) and amylose content (Murtiningrum et al, 2012). Temperature gelatinization of Banten taro starch higher than sweet potato starch 66,92oC (Irhami et al, 2019); taro flour starch 69oC, corn starch 62-72oC, cassava starch 68-78oC and rice starch 52-64oC (Aryanti et al, 2017).

Besides the gelatinization temperature, other information that can be seen from Figure 2 is maximum viscosity. The maximum viscosity or peak viscosity (PV) value of Beneng Taro Starch is quite small, which is 1075 cP. This result was contrary to the results of research conducted by Fetriyuna et al (2016), that peak viscosity on the taro beneng

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starch is 3899,5 cP. The high peak viscosity on taro is due to an increase in the viscosity of the paste due to the distillation of starch granules,

especially amylose. Amylose content in Beneng Taro Starch is quite high at 28.91% (Nurtiana *et al*, 2019).

Table 1. RVA results of beneng taro starch

RVA parameters	Replication		
	L	2	Average
PT: temperature (°C) at 20cp	81.8	82.1	81.9
Pt: time (s) at 20cP	217	218	218
Peak viscosity (cP) PV	1077	1073	1075
Temp. at peak viscosity (°C) PVT	92.1	92.8	92.4
time at peak viscosity (s) PVt	268	272	270
Hot paste Peak viscosity (cP) HPV	858	865	862
Temp HPV (°C) HPVT	86.6	84.9	85.7
Cold paste viscosity (cP) CPV	1257	1264	1261
Cooking ability (s) CA	51	54	52
Viscosity beginning plateau (cP) VBP	1067	1065	1066
Viscosity end plateau (cP) VEP	883	889	886
Breakdown (cP) BD	219	208	214
Setback (cP) SB	180	191	186
Consistency(cP) CS	399	399	399

Maximum viscosity is the viscosity of the paste produced during heating. Increased absorption of starch granules by heat will increase the viscosity of the paste so that at maximum viscosity perfect gelatinization occurs. The greater the ability to expand starch granules, the higher the viscosity of the paste and eventually will decrease again after the rupture of starch granules (Swinkles, 1985). Starches that have high water absorption ability will also experience high swelling which will result in high viscosity of the peak of the paste. Excessive swelling of the starch granules will be followed by the decay of amylose molecules from the granules as a result of its inability to withstand pressure (Wulandari, 2010).

After achieving maximum viscosity, if the heating process in RVA is continued at a higher temperature, the starch granules will be brittle, break apart and cut to form polymers, aggregates and their viscosity decreases due to amylose leaching. The decrease occurred at a heating temperature of 95°C suspension which was maintained for 10 minutes. The value of viscosity reduction that occurs in these conditions is called viscosity breakdown. The score breakdown of Beneng Taro Starch is

quite high at 214 cP, it shows the characteristics of starch that is less heat resistant.

Breakdown or decrease in viscosity during heating shows the stability of the starch paste during heating, where the lower breakdown value, the paste formed will be more stable to heat (Rostianti et al, 2018). According to Eliason (2004) breakdown is an important factor that has an influence on the application of starch in food products. When starch granules swell and experience heat and shear, starch undergoes fragmentation and results in a reduction in viscosity which indicates starch breakdown. A high breakdown value is undesirable because it causes low viscosity also results in cohesive properties in a starch paste.

After heating to 95°C and held for 10 minutes, the starch paste is cooled. When cooled, the viscosity of the starch paste increases again. The setback value of viscosity is obtained by calculating the difference between the viscosity of the starch paste at 50°C and the maximum viscosity that has been reached upon heating. The set back value in this study was also high, 186 cP. This is caused by the tendency of retrogradation in starch

paste because of its high amylose content. According to Lehmann et al (2003), the viscosity of paste setback shows a tendency towards retrogradation that occurs in amylose molecules because amylose is more easily exposed to water and recrystallized more easily than amylopectin. A similar result was stated by Fetriyuna (2016) that native Beneng taro starch tends to have a high setback viscosity value. The higher the setback value indicates the higher tendency to form a gel (increase viscosity) during cooling, as well as indicate that the starch paste is fast undergoing retrogradation (Marta, 2011).

## CONCLUSION

The microstructure of Beneng taro starch showed polygon and quite uniform shapes. The amylographic profile of the Beneng taro starch respectively for initial gelatinization temperature, maximum viscosity, breakdown viscosity and setback viscosity were 81.9°C, 1075 cP, 214 cP, and 186 cP. These results indicate that Beneng taro starch still has many shortcomings to be applied to food products. So it is necessary to modify native Beneng taro starch.

## ACKNOWLEDGEMENT

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## **SORTING MANALAGI APPLE (*Malus sylvestris* Mill) USING IMAGE PROCESSING APPLICATION**

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### **ABSTRACT**

Manalagi apple (*Malus sylvestris* Mill) is one type of apple that has been widely known by the public in Indonesia. Fruit sorting in Indonesia, particularly Manalagi apple, is carried out manually so it is still at a disadvantage. This study aimed to arrange image processing applications with criteria for size, width, height, color, and defect area of fruits. The samples of Manalagi apple used in this study were 130 with 50 each in the non reject and reject quality classes and 30 for the validation test. Manalagi apple samples were obtained from one of the collectors located in Batu City. The image of Manalagi apple processed gained several variables, including perimeter, area, height, width, r, g, b, and defect area. Based on statistical analysis, image quality variables that could be used as input for making applications in the form of logical sentences were perimeter, area, and defect area. Based on the application, the validation test obtained an accuracy of 86.65%.

**Keywords:** Manalagi apple, sorting, image processing

### **INTRODUCTION**

Apple (*Malus sylvestris* Mill) is one of the leading agricultural products from Indonesia. This plant originates from subtropical regions. Apples were planted in 1934 and could grow well in Indonesia. Malang and Pasuruan regency are centers of apple production in Indonesia. Apple plants in this area began to be developed in 1950. In addition to these areas, apple plants are also widely developed in the Situbondo, Banyuwangi, and East Nusa Tenggara regions (Soelarso, 1996).

Apple production in the three apple production centers, namely Malang Regency, Batu City, and Pasuruan Regency, is quite large. The number of apple production in Malang Regency, which was centered in Poncokusumo District, reaches 150,545 tons/year (Agriculture Service of Malang Regency in Rahaju, 2013). Apple production in Batu City reaches 833,915 tons/year (Theresia Maria & Makmur, 2015). Apple production in Pasuruan Regency reaches 140,284 tons/year (Gutomo, 2015).

Manalagi apple is one type of apple that is widely known by the public. In increasing the selling price, apples need to be sorted to separate between good and bad apples. Sorting is a step for apple products to be accepted on the market. Apple sorting in Indonesia is conducted by human manually. Therefore, it has many disadvantages, including the results of sorting are less in similarity, require more time, and there is a difference in perception of the sorting result due to subjectivity.

Image processing can be used as an alternative way of sorting Manalagi apple. Image processing is the process of processing and analyzing images that involve a lot of visual perception (Ahmad, 2005: 4). Image processing can determine variables from fruit quality such as fruit weight, fruit width, and defect area. Some research on image processing for sorting and qualifying fruit has been carried out, and the results were quite satisfying. Yultrisna and Syofian (2016) conducted a study on the sorting of tomatoes using image processing applications.

Application designed to sort tomatoes based on color differences. The research showed that the result of sorting accuracy reached 100%. Other studies conducted (Saintika, Wijayanto, & Wiguna, 2018) on the classification of carrots based on image processing also showed satisfactory results. The designed image processing application was able to predict fruit quality manual parameters such as weight, volume, length, and diameter. The result showed that the classification accuracy reached 98.88%.

Research on the classification of Manalagi apple also been conducted by Anugrahandy *et al* (2013) and Nugraha (2017). Research carried out was the classification of Manalagi apple with various quality classes. Based on the results of the study obtained an accuracy that was quite satisfying, with accuracy reaching 90%. Both of these studies have similarities in the classification of Manalagi apple quality with criteria for size, width, height, and color. Further research needs to be conducted with the sorting criteria, namely the defect area. The criterion of the defect area was the condition of the apple, which has a defect due to animal attack, genetic conditions, and damage during transportation.

Research classification about fruit defect was conducted by (Riquelme *et al*, 2008). This research was classification olive according to external damage. Seven commercial categories of olives, established by product experts, were used: undamaged olives, mussel-scale or 'serpeta', hail-damaged or 'granizo', mill or 'rehu' s', wrinkled olive or 'agostado', purple olive and undefined-damage or 'molestado'. The accuracy of this research reached 75%. The other research about fruit defect area was conducted by (Arjenaki, Moghaddam, & Motlagh, 2013). This research was sorting tomatoes according to surface defects. The accuracy of this research reached 85%.

Therefore, this research aimed to arrange image processing applications with criteria for size, width, height, color, and defect area. The expected result is an application that is accurate enough to sort the Manalagi apple.

## **MATERIALS AND METHODS**

### **Tools and Materials**

The tools used in this study included:

1. CCD camera (Charge Couple Device) as an image capture tool. CCD camera was a special camera for sorting. image capture results had a high pixel value so the results were quite detailed.
2. TL lamp with a power of 5 Watt (220 Volt) as a lighting aid. The white TL lamp was chosen because it had a complete color value content. Thus it would clarify the results of the image that had been obtained.
3. White cloth as the base layer. White cloth was chosen as the background because it had a color that contrasts with the apple object. Thus the resulting image could clearly distinguish between objects and backgrounds. Laptop device with a core i5 processor as image processing hardware

The materials used in this study were two qualified Manalagi apples, no reject and reject, 50 pieces for each, and 15 pieces for the validation process so that the samples of Manalagi apples were 130 pieces. This apple was obtained from one of the collectors located in Batu City.

### **Methods**

This research used a CCD camera to capture images from Manalagi apple. Figure 1 was the stage of research that was being carried out.

#### **a. Determination of image quality variables**

Quality variables of Image processing for the classification of Manalagi apple in this study were:

1. non-reject fruit (normal): Large to medium fruit size, standard fruit shape, no or little defect area.
2. reject fruit: Very small fruit size, abnormal shape, there is a large defect area.

Based on the above criteria, the image quality variables used for Manalagi apple include shape, size, color, and damage. The estimation of the variable of image quality can be seen in Table 1.

#### **b. Image Acquisition**

Image Acquisition was a process of getting the best images of Manalagi apple. This process was carried out by adjusting the distance of the camera and the irradiation position to resemble a natural image, a slight shadow arises, and no excess light affects the

color of the object. The best camera distance and exposure position are shown in Figure 2.

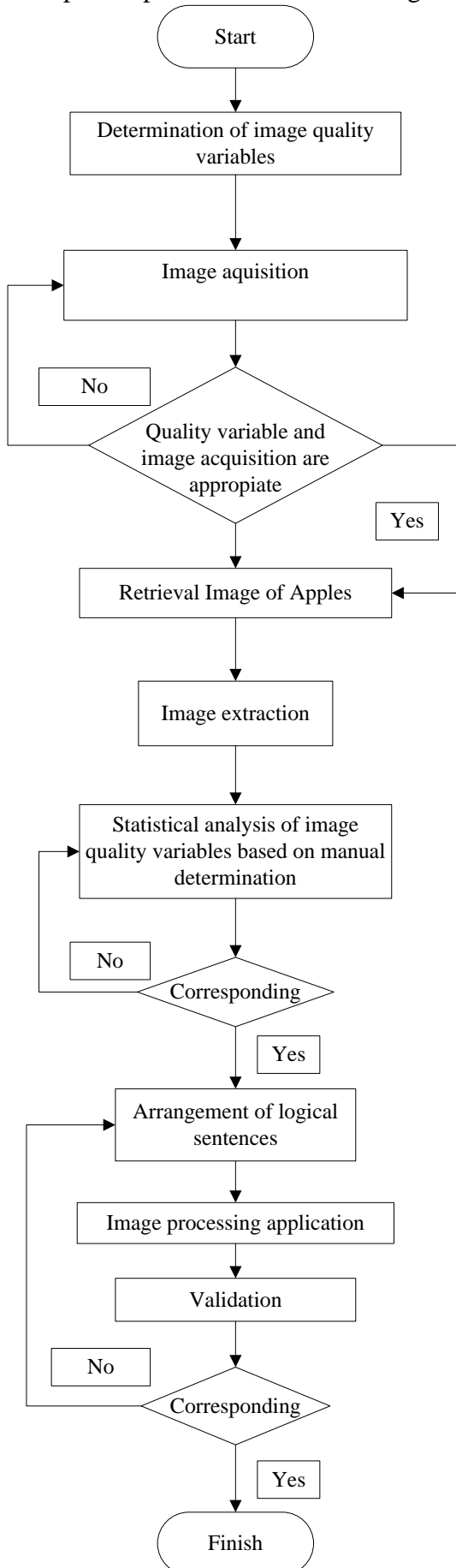


Figure 1. Research procedure

Table 1. Variable quality of Manalagi apple and image processing variable

No	Variable Quality of Manalagi apple	Variable of Image Processing	Description
1	Shape	Perimeter (P)	The image quality variable that can represent the shape characteristics of apples is the perimeter. Perimeters are expressed in units of pixels. Size properties correlate with the variable image quality of the area, width, and height. The area, width, and height of Manalagi apple have pixel dimensions.
2	Size	Area (A), width (L), and height (H)	Quality variables of image processing that can represent the skin color of Manalagi apple are a red color index (r), green color index (G), and blue color index (B). Quality variables representing damage are defective areas. The defect area is obtained based on the threshold function that can separate the area of Manalagi apple fruit from the area of damage. The area of defects of the fruit has pixel dimensions.
3.	Colour	r, g, and b	
4	Defect	Defect area (C)	

c. Image capturing

The apple image was captured by using a CCD camera. The image was taken according to the image acquisition process. The steps are below:

1. place the Manalagi apple on a white cloth as a background and under the CCD camera with the height and angle obtained from the image acquisition procedure.
2. recording images in RGB format.
3. save the file in the BMP format.

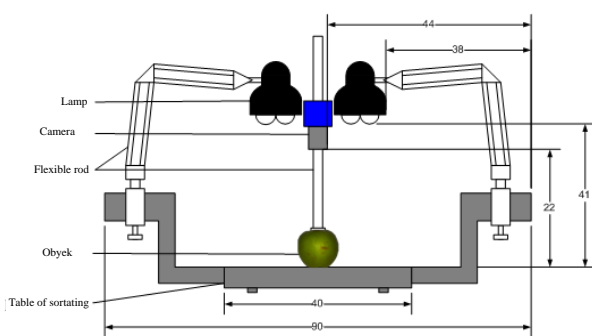
d. Image extraction

The data source of Image extraction was a converted image with a resolution of 1024 x 768 pixels. Image extraction was a process to measure the perimeter of the object, area, width, height, area of defect, and a color index of RGB (red, green, blue). Image extraction was conducted with a computer program made in advance using the Sharp Develop 3.2 programming language.

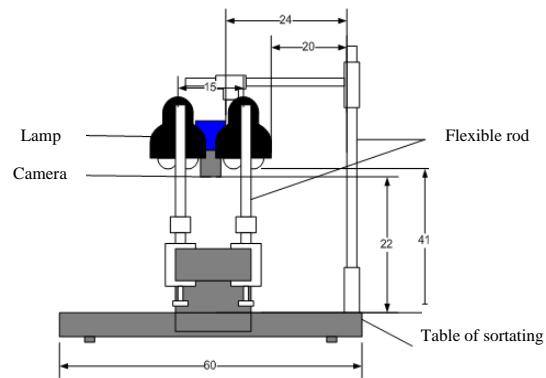
The procedure in image extraction are:

1. creating an Image Processing program in Sharp Develop 3.2, which could calculate object areas, and the RGB color index (red, green, blue).

2. calculating the perimeter of the Manalagi apple
  3. calculating the area of Manalagi apple by counting the number of pixels constructing the apple area.
  4. calculating the width of the Manalagi apple
  5. calculating the color index of R (red), G (green), and B (blue) of the apple
  6. calculating the defect area of the binarization process with the threshold function
- analyzing collected data with statistics.



(a)



(b)

Figure 2. Image acquisition from the capturing table (a) the table looks from the front, (b) the table looks from the side

e. Evaluation of the correlation between image quality variables and manual sorting

The results of the data processing of the Manalagi apple image were carried out with a statistical analysis model. The statistical variables used were the average, standard deviation, Q1 (first quartile), median, Q3 (third quartile), minimum value, and maximum value. Furthermore, the values of quality variables that have been tabulated were illustrated in the boxplot graph.

f. Data processing

Procedures in data processing were:

1. collecting data on the quality variable of the apple image obtained from image extraction.
2. tabulating the value of the perimeter image quality variable with the sample number as the independent variable and the perimeter as the independent variable classified by each quality class.

3. calculating the average value, standard deviation, maximum value, and minimum value of the object area for each quality class. These values provided limits to the amount of the object area for each quality class.
4. plotting perimeter variables in the Box graph and whisker plot.
5. determining the boundary values that can be used to separate each sample based on their grade quality.
6. compiling logical statements based on the value limits derived from the statistical analysis.
7. repeating procedures 1-6 for other image quality variables, namely: Area and diameter, index r, g, and b, and area of defect.
8. merging the combination of logical statements with trial and error until the best level of conformity is obtained between the image quality parameters and quality class with the formula



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(compatibility level = the number of correct predictions/number of samples x 100%).

g. Validation

Validation was an assessment of the performance or accuracy of the prediction of the Manalagi apple image processing application to the examples given during the training process. This process was conducted by providing data samples other than the training process and seeing the ability of the program to present the correct answer. This validation test was carried out by analyzing the confusion matrix analysis.

Confusion matrix analysis was used to obtain results that illustrate the validation of the program. Besides, the confusion matrix was a table that showed the visualization of the performance of an algorithm explicitly, especially at the supervised training stage. Each column in the matrix shows the predicted class, while each row shows the actual class. Outside the realm of artificial intelligence, confusion matrices were known as contingency tables or error matrices (NRCan in Soedibyo, 2012: 57).

Confusion matrix analysis was a step in the completion of the program in the form of a matrix. The results of predicted sorting were compared with the actual results of sorting. Validation values could be made according to the target in the program that had been made. If the validation value results in a large enough accuracy then there was no need to make improvements to the program and vice versa if the verification value was low then an improvement in the program was needed.

## RESULTS AND DISCUSSION

### Sorting Application of Manalagi Apple

Chemical analysis results are shown in Table The sorting application of Manalagi apple in this study was created using the SharpDevelop 4.2 program. This program was built to gain the quality variable of the Manalagi apple. Quality variables employed in the sorting process of the fruit were perimeter, area, height, width, the area of defects, and color indexes consisted R, G, and B. Appearance of the fruit sorting application can be seen in Figure 3.

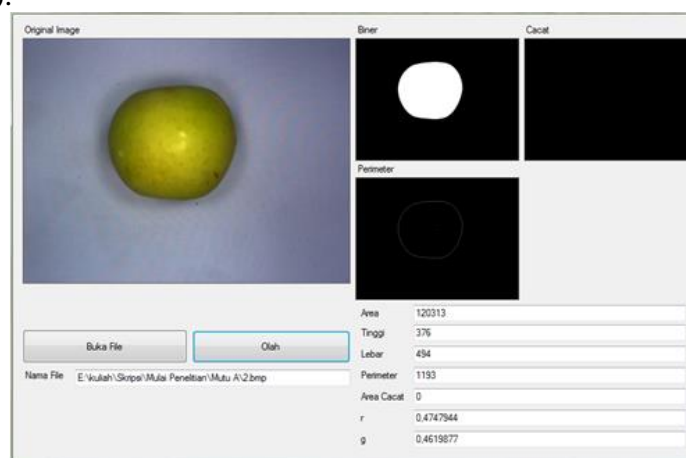


Figure 3. Sorting application of Manalagi Apple

Figure 3. is an appearance of the application used to analyze the image variables of Manalagi apple. In the application, there are two buttons designed to open and process images, four picture boxes arranged to display images, and eight boxes used to display file names and processed images. The procedures performed in the Manalagi apple image processing application are:

1. press the "Open File" button.
2. press the "process" button.

3. repeat procedure 1 and 2 until all the Manalagi apple samples have been processed.

### Determination of Segmentation Boundary Value of the Background (Thresholding)

Thresholding is the process of separating a region with a background. The result of image thresholding is called a binary image. Threshold values are obtained from differences in the values of R, G, and B belonging to the object and values of R, G, and B belonging to the background.

Determination of the threshold value was obtained from sampling values of R, G, and B of objects and backgrounds. The sample points used in determining threshold values were ten points. Furthermore, the sample values were analyzed using graphs to determine the differences in the values of R, G, and B objects and background. From these different values, were determined the boundary values of R, G, and B that were able to distinguish between the objects and the background. The graph used to determine the background threshold function can be seen in Figure 4.

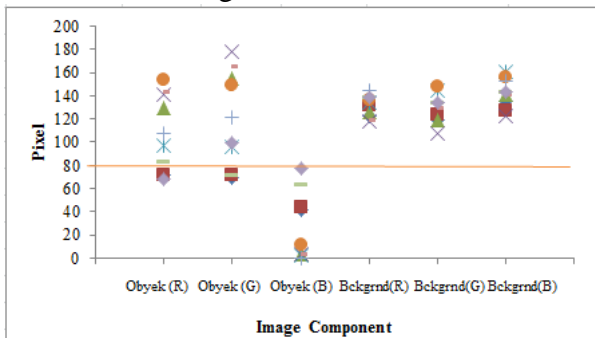


Figure 4. Distribution values of the objects and backgrounds RGB

Figure 4 showed the distribution values of R, G, and B of the ten sample points of Manalagi Apple. Based on Figure 4, in general the distribution values of R, G objects (apples) were almost the same as the values of background (cloth) distributions. The different distribution values were at value B.

The boundary value that distinguished between objects and background was right on B equal to 80. Thus, the background threshold function was when the original image had value  $B > 80$ , then the image would be black, otherwise, it would be white. Based on the boundary value, the black image was the background, while the white one was the object.

**Determination of the Segmentation Boundary Value (Threshold) of Defect Areas**

The threshold of the defect area is used to differentiate fruit defect area pixels from fruit area pixels. The determination of the threshold value was obtained from taking the ten sample points of Manalagi apple. Then, the sample values were analyzed using graphs to determine the differences in the values of R, G, and B in the defects area and fruit areas. The

graph used to determine the threshold function area for defects can be seen in Figure 5.

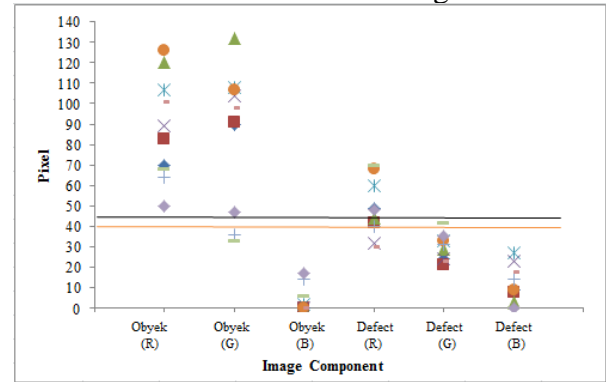


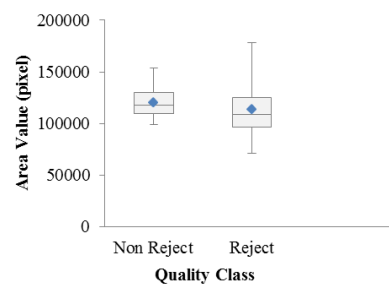
Figure 5. Distribution value of RGB of the defect and fruit Area

Figure 4 shows the distribution values of R, G, and B from the ten sample points of the Apple Manalagi defect. Based on Figure 4, the different distribution values are at the values of G and B.

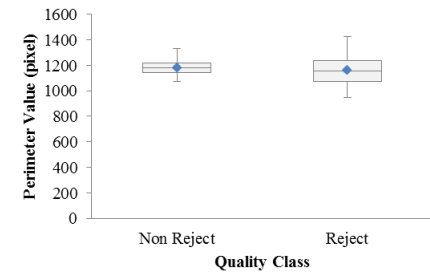
Based on Figure 5, the threshold function of the defect area was when the morphological image was white-colored (R-value = G value = B value = 255) AND (R-value < 45) AND (G value < 40), so that it displayed the defect area image as white. Otherwise, it displayed the image of the defect area as black.

**Statistical Analysis of Image Quality Variables**

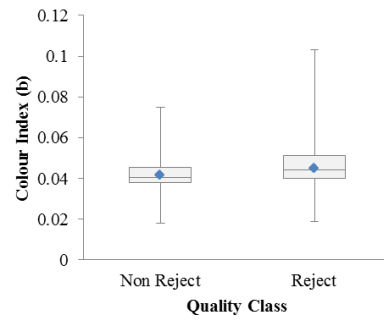
The process of image extraction is the process of getting the value of the image quality variable through a 100 Manalagi apple sample base on the criteria of non-reject and reject. Statistical analysis of image quality variables was accordingly the result of the image extraction process on the quality variables perimeter, area, width, height, area of defects, and color index (r, g, and b). The results can be seen in the following figures.



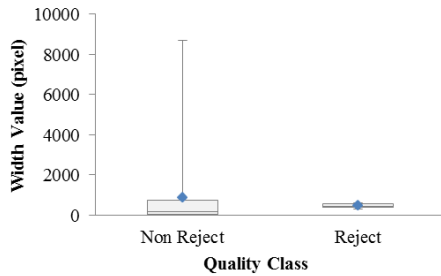
(a)



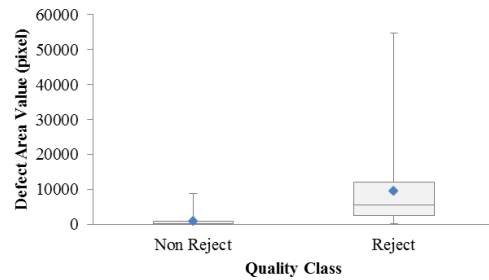
(b)



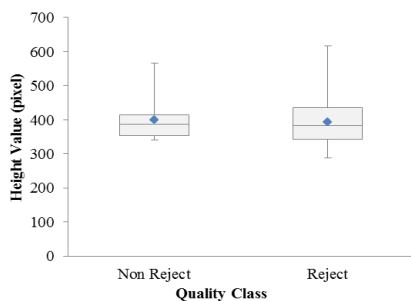
(g)



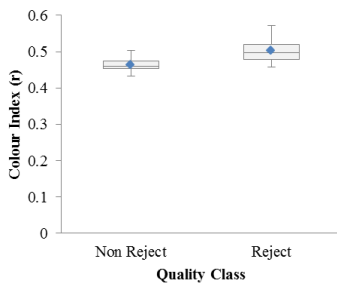
(c)



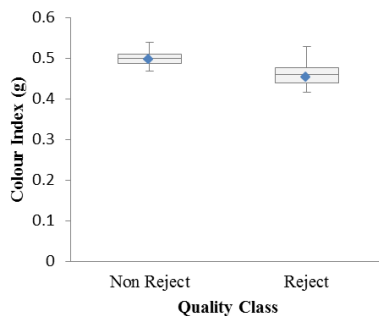
(h)



(d)



(e)



(f)

Figure 6. Statistical analysis of image quality variables (a) area, (b) perimeter, (c) height, (d) width, (e) red, (f) green, (g) blue, (h) defect area.

Based on figure 6 graphic, the image quality variables used in compiling the Apple apple sorting application were the area, perimeter, and defect area. These three variables have consistency that can classify apples by non reject and reject apple categories.

### Determination of Logical Sentences

The first process in determining a logical sentence was figuring out the boundary value of the image variable used as a qualification reference. Then, statistical analysis was applied to analyze the image quality variables of the apples. Image quality variables that were employed as input in determining the boundary value were perimeter, area, and defect area. The value limits used for the sorting of apples can be seen in Table 2.

Table 2. Variable quality of Manalagi apple and image processing variable

Quality Variables	Quality class	
	<i>Non Reject</i>	<i>Reject</i>
Perimeter (P)	$1400 \geq P > 1070$ ;	$P > 1400$ Or $P < 1070$ ;
Area (A)	$154000 \geq A > 99000$ ;	$A > 154000$ Or $A < 99000$ ;
Defect Area (C)	$800 \geq C > 0$ ;	$C > 800$ Or $C < 1500$ ;

The combination of logical sentences based on the boundary values was used as input for building the application were as follows:

1. And if  $perimeter \leq 1400$ ,  $perimeter > 1070$ ,  $area \leq 155000$ ,  $area > 99000$ ,  $defect\ Area > 0$ ,  $defect\ area \leq 350$ , then sorting decision = "Non Reject"
2. Or if the  $perimeter > 1400$ ,  $perimeter < 1070$ ,  $area > 155000$ ,  $area < 99000$ ,  $defect\ area > 350$ ,  $defect\ area \leq 1500$ , then sorting decision = "Reject"

The combinations of quality variables above were the best combinations by trial and error based on the results of the boundary value on each quality variable. Furthermore, the combination was inputted into the program making the Apple Sorting application again. The Apple Sort application has a sorting analysis result box named "Sort Results." The appearance of the Apple Sorting application can be seen in Figure 7.

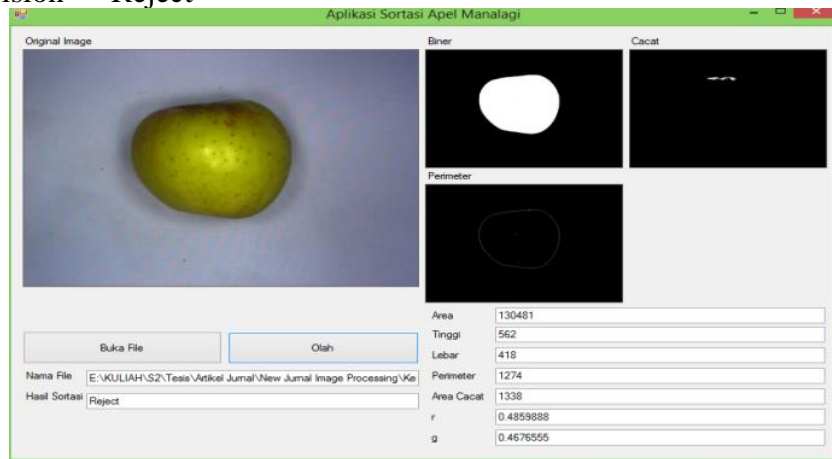


Figure 6. Application of Manalagi apple sorting

### Application Validation of Manalagi apple Sorting

The assessment of the accuracy of the Manalagi apple sorting application was carried out on 30 samples of quality Manalagi apple.

The quality sample includes non-reject and reject apples. Testing the accuracy of this program is accomplished by the validation process using the confusion matrix. The results of the validation can be seen in table 3.

Table 3. Validation of the image processing application of Manalagi apple

Quality Class	Prediction		Total row	Prediction accuracy	Error
	Non Reject	Reject			
Actual Non- Reject	11	4	15	73,33%	26,67%
Actual Reject	0	15	15	100%	0%
Accuracy Total			86,65%		

Based on table 3, the total accuracy value was 86.65%. This value was obtained from the average prediction accuracy between non reject and reject. It can be concluded that the sorting application of the Manalagi apple could be applied with high accuracy.

### CONCLUSION

Image quality variables used in the sorting of Manalagi apple were perimeter, area, width, height, index (r, g, and, b), and defect area. Furthermore, based on these variables, the variables used as input in the

sorting application are the perimeter, area, and defect area. The three variables were then combined in the form of logical sentences, which were then inputted in the Manalagi apple sorting application. Based on the validation test, obtained a total accuracy value of 86.65%. Thus, it can be concluded that the application sorting of Manalagi apple can execute the sorting with quite high accuracy.

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## EVALUATION OF ANTIOXIDANT PROPERTIES OF *Cucumis melo* L cv. Hikapel DURING STORAGE AT ROOM TEMPERATURE

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### ABSTRACT

The changes in antioxidant compounds and antioxidant activities of melon (*Cucumis melo* L.) cultivar Hikapel during postharvest storage at room temperature were evaluated. Melon with three ripening stages (27 DAA, 29 DAA, and 32 DAA) were harvested and stored at 25°C for 20 days. Melon cv. Hikapel were evaluated for their antioxidant compounds such as ascorbic acid, total phenolic (TPC), and total flavonoid content (TFC). Antioxidant capacity was also evaluated using DPPH radical scavenging assay (DPPH-RSA) and ferric reducing power assay (FRPA). The result showed that there were different levels of antioxidant compounds (TPC, TFC, and AAC) and antioxidant activities (DPPH-RSA & FRPA) in different ripening stages of this melon. Antioxidant compounds and antioxidant activity decreased during postharvest storage. In conclusion, melon cv. Hikapel provides various natural antioxidant compounds such as phenolic, flavonoid which can be the main contributors to the overall antioxidant activity of melon cv. Hikapel and its antioxidant properties were influenced by the postharvest storage period.

**Keywords:** *Cucumis melo* L. cv. Hikapel, antioxidant, ripening stage, room storage

### INTRODUCTION

*Cucumis melo* L. cv. Hikapel, a new cultivar of melon, had been developed in the Laboratory of Genetic and Breeding, Faculty of Biology, Universitas Gadjah Mada (UGM), Indonesia. The melon cv. Hikapel is one of the non-netted orange-fleshed melons. This melon has several quality values, such as an orange-fleshed, sweet taste, crunchy texture, and good shelf life. This melon has been known for its antioxidant properties, such as ascorbic acid, carotenoid, phenolics, and flavonoid (Wulandari *et al.*, 2017). These compounds are suggested to be major bioactive compounds for health benefit. Nowadays, the consumption of fruits has been associated with reduced risk of chronic diseases. The awareness about the importance of nutritional requirements among consumers has increased due to people's healthy lifestyle.

It has been reported that the levels of health-promoting bioactive compounds of fruits are strongly influenced by the ripening stage and postharvest storage time (Arancibia-Avila *et al.*, 2008; Zainuddin *et al.*, 2014; Zhang *et al.*, 2008). The study of changes in bioactive compounds during ripening of fruit and during storage time has great relevance both to human health and commercial purposes. That study provides valuable information to understand the alteration of those compounds and to evaluate the best harvesting time and postharvest storage time to get the highest antioxidant potential. Harvesting time is essential to get high-quality fruit with good nutrition potential. Postharvest storage time is crucial to determine how long fruit can be stored at room temperature storage.

As a new melon cultivar, the study of antioxidant potential in this fruit is still rare. The study focused on antioxidant activity

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alteration during postharvest storage of melon cv. Hikapel has never been done. Therefore, this research aimed to study the changes in antioxidant activity and antioxidant compounds during postharvest storage at room temperature.

## MATERIALS AND METHODS

### Plant Materials and Chemicals

Melon (*Cucumis melo* L.) cv. Hikapel was cultivated in Agricultural Training, Research and Development Station of Universitas Gadjah Mada (KP4 UGM), Desa Kalitirto, Berbah Sub-District, Sleman Regency of the Special Region of Yogyakarta. Melon with three ripening stages S1 (27 days after anthesis (DAA)), S2 (29 days after anthesis (DAA)), and S3 (32 days after anthesis (DAA)) was harvested and transferred to the laboratory immediately.

The melon was washed with NaOCl 50 ppm, rinsed with water, air dried, and stored for 20 days at room temperature. Melon was cut, deep freeze, and freeze-dried. The freeze-dried samples were ground and extracted using methanol: acetone (80:20) acidified with acetic acid, sonicated and centrifuged at 4000 g for 10 min at 4°C. The supernatant was used for further analysis such as total phenolic, total flavonoid, DPPH-RSA, and FRP assay.

All chemicals used in this experiment have a pro analysis grade. H<sub>3</sub>PO<sub>4</sub>, AlCl<sub>3</sub>.6H<sub>2</sub>O, FeCl<sub>3</sub>.6H<sub>2</sub>O, Na<sub>2</sub>CO<sub>3</sub>, K<sub>3</sub>Fe(CN)<sub>6</sub>, methanol, ethanol, acetone, petroleum ether, acetic acid were purchased from Merck. Folin-Ciocalteu phenol reagent, TCA, 2,2-dipyridyl, quercetin, gallic acid, ascorbic acid, 2,2-diphenyl-1-picrylhydrazyl radical (DPPH).

### Methods

#### Total phenolic content (TPC) determination

The amount of total phenolic content (TPC) in melon extracts was determined according to the Folin-Ciocalteu method (Dewanto *et al.*, 2002). Samples 125 µL were introduced into test tubes in which containing 125 µL of Folin-Ciocalteu's reagent and 250 µL of sodium carbonate (7.5%, w/v), mixed by vortex and allowed to stand in darkness at room temperature for 90 min. The absorbance was measured spectrometrically at 760 nm. The total phenolic content was expressed as mg gallic acid equivalents per 100 g of fresh

weight (mg GAE/100 g DW). All measurements were done in triplicate.

#### Total flavonoid content (TFC) determination

Total flavonoid content was estimated according to the procedure of Santas *et al.* (2008) based on the aluminum chloride complex formation. To 1 mL of supernatant added with 1 mL of 2% (w/v) AlCl<sub>3</sub> methanolic solution. The mixture was then allowed to react for 2 min at room temperature and the absorbance was read at 410 nm. Total flavonoid content was calculated as mg quercetin equivalent per 100 g of fresh weight (mg QE/100 g DW). All measurements were done in triplicate.

#### Ascorbic acid content (AAC) determination

Ascorbic acid content was determined by the 2,2-dipyridyl method (Giudice *et al.*, 2015). This method is based on the reduction of Fe<sup>3+</sup> to Fe<sup>2+</sup> by ascorbic acid and detection of Fe<sup>2+</sup> complexed with 2,2-dipyridyl. Five grams of frozen flesh were homogenized in 5 mL of 5% (w/v) trichloroacetic acid (TCA). The homogenate was filtered and centrifuged for 10 min at 12,000 g (4°C). Then, 20 µL of the supernatant mixed with 20 µL 0.4 M phosphate buffer (pH 7.4) and 10 µL distilled water.

Eighty microlitres of colour reagent solution, prepared by mixing solution 1 (31% H<sub>3</sub>PO<sub>4</sub>, 4.6% (w/v) TCA, and 0.6% (w/v) FeCl<sub>3</sub>) with solution 2 (4% 2,2'-dipyridil (w/v) made up in 70% ethanol) at a proportion of 2.75:1 (v/v), were added. The mixture was incubated at 37 °C for 40 min, then cooled down immediately to room temperature. Absorbance was read at 525 nm. Ascorbic acid content was expressed in mg AA/100 g DW. All measurements were done in triplicate.

#### Ferric reducing power assay (FRPA)

The ferric reducing power of the melon fruit extracts was performed by using the potassium ferricyanide-ferric chloride method (Berker *et al.*, 2007). One mL of extract was mixed with 2.5 mL phosphate buffer (0.2 M, pH 6.6) and 2.5 mL of 1% (w/v) potassium ferricyanide (K<sub>3</sub>Fe(CN)<sub>6</sub>). The mixtures were incubated at 50°C for 20 min, and 2.5 mL of 10% (w/v) trichloroacetic acid (TCA) was added. 2.5 mL of the mixture was taken and mixed thoroughly with 2.5 mL of distilled water and 0.5 mL of 0.1% FeCl<sub>3</sub> (w/v). The

absorbance of the blue green color was measured at 700 nm. Ferric reducing power activity was expressed as mg ascorbic acid equivalents per 100 g of fresh weight (mg AAE/100 g DW). All measurements were done in triplicate.

#### DPPH radical scavenging activity assay (DPPH-RSA)

The DPPH radical-scavenging activity of the melon fruit extracts was estimated as described by Sharma *et al.* (2009). Briefly, 0.3 mL sample extract was mixed with 2.7 mL of 100  $\mu$ M DPPH in a methanolic solution. The mixtures were left for 40 min in the darkness at room temperature, the absorbance was then measured at 517 nm. All measurements were done in triplicate. The percentage of DPPH radical-scavenging activity was calculated using the following equation:

$$\% \text{ RSA} = [(\text{Abs. Blank} - \text{Abs. Sample}) / (\text{Abs. Blank})] \times 100\%$$

#### Statistical analysis

The effects of postharvest storage time on the antioxidant properties of melon cv. Hikapel at different ripening stages were analyzed by one-way analysis of variance (ANOVA). Tuckey test was carried out to identify significant differences between ripening stages and postharvest storage time. Mean values with  $p < 0.05$  were considered statistically significant. For analysis of the correlation between antioxidant capacity and antioxidant compounds, the Pearson correlation was carried out. All statistical analysis was performed using SPSS 20.

## RESULTS AND DISCUSSION

*Cucumis melo* L cv Hikapel at ripening stage S3 can only be stored until the 10th day of storage. *Cucumis melo* L cv Hikapel at ripening stage S1 can be stored until 20<sup>th</sup> and ripening stage S2 can be stored until the 15<sup>th</sup> day of storage. Melon cv. Hikapel provides wide variety of antioxidant compounds such as carotenoid, phenolic, flavonoid, and ascorbic acid. The statistical analysis revealed that TPC decreased significantly during the on tree-ripening.

Total phenolic content reached the highest concentration at ripening stage S1 (124.75  $\pm$  13 GAE/100 g DW) and decreased to

92.20  $\pm$  04 mg GAE/100 g DW (S3). Similarly, significantly decreased in total phenolic as in cantaloupe melon (Abu-Goukh *et al.*, 2011). There was no change of total flavonoid content during ripening of melon cv. Hikapel. Ascorbic acid content and total carotenoid content increased during ripening stages. Ascorbic acid content reached the highest value at ripening stage S3 (243.92  $\pm$  11 mg AA/100 g DW). It showed that ascorbic acid was synthesized during on-tree ripening of melon cv. Hikapel. These results are in agreement with several studies reported in teasel gourd (Singh *et al.*, 2015) and carambola fruit (Zainuddin *et al.*, 2014).

Total -phenolic, -flavonoid, -ascorbic acid content of melon cv. Hikapel were changed during postharvest storage at room temperature as presented in Fig. 1. Total -phenolic and -flavonoid, ascorbic acid content were decreased during postharvest storage. Ascorbic acid content decreased during storage might be related to the increased of *ascorbate peroxidase* activity during early storage time. But when the *ascorbate peroxidase* starts to decrease in their activity, the ascorbic acid content still decreased. It might be caused by the use of ascorbic acid for oxalate or tartrate synthesis.

Ascorbic acid catabolism occurs with tartrate and oxalate as major products (Smirnoff and Pallanca, 1996). Therefore, it is also possible that ascorbic acid in melon cv. Hikapel is being driven to other routes, such as oxalate and tartrate production, such as in guava and mango (Gomez and Lajolo, 2008). Generally, when fruits become over-ripe, vitamin C content declines, concurrently with the degradation of fruit tissues (Kalt, 2005).

Total -phenolic, -flavonoid content decreased during fruit storage can be related to the expression of phenolic biosynthetic enzyme genes, *phenylalanine ammonia-lyase* (PAL) and the phenolic oxidizing enzyme, *polyphenol oxidase* (PPO) and *peroxidase* (POX). Increasing in PPO activity and decreasing PAL activity was also the possible contribution to the TPC and TFC decreasing value as reported in carambola fruit (Zainuddin *et al.*, 2014).

Another possible reason for the decrease in ascorbic acid, phenolic, and flavonoid content might be due to the decrease of internal



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antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), APX, and guaiacol peroxidase (G-POD) making the cells turn to other reserved antioxidant compounds

such as phenolic, flavonoid, and ascorbic acid in maintaining cellular integrity (Huang *et al.*, 2007).

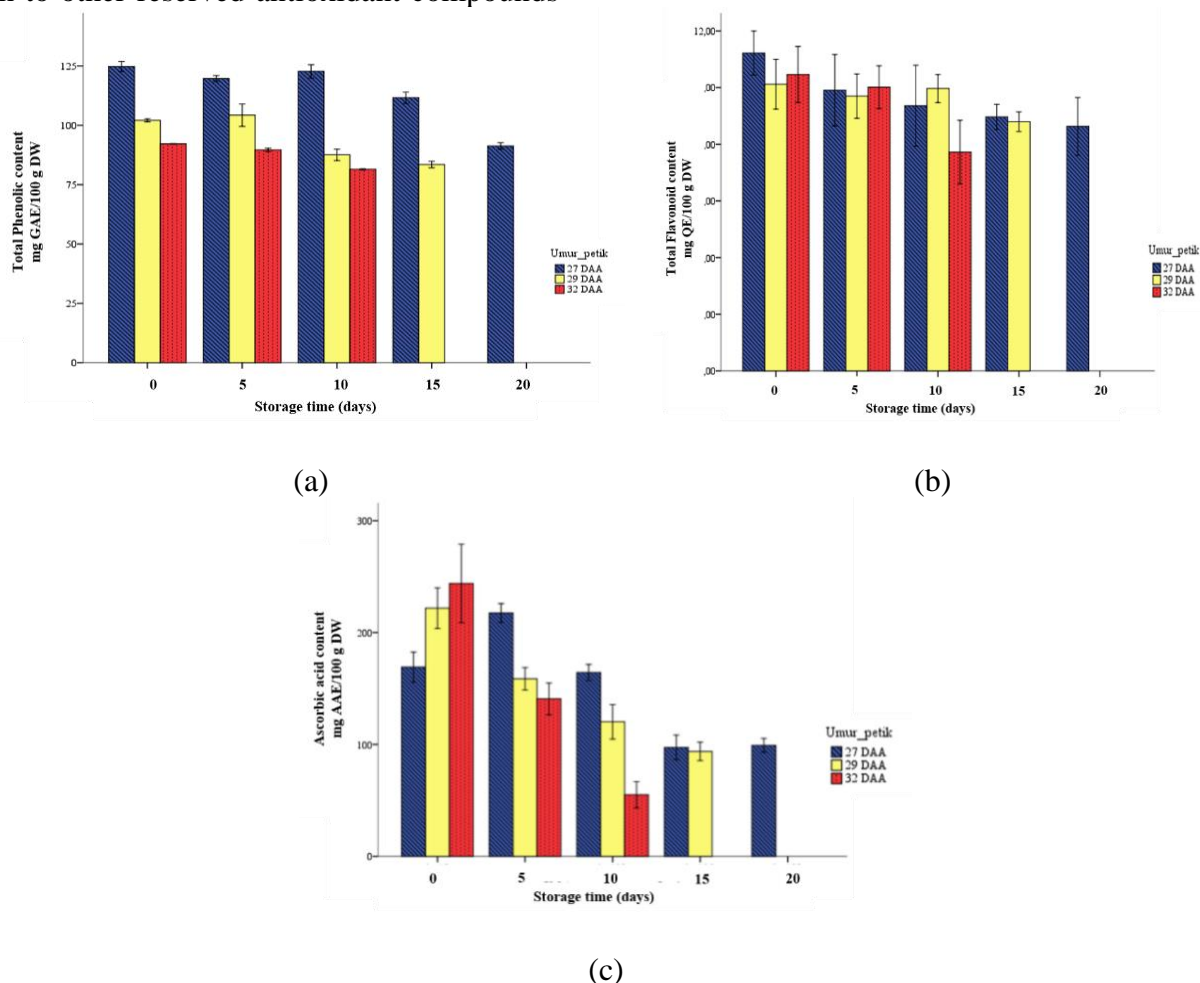


Figure 1. Total Phenolic Content (a), Total Flavonoid content (b), Ascorbic acid content (c) of *Cucumis melo* L. cv. Hikapel during postharvest storage at room temperature.

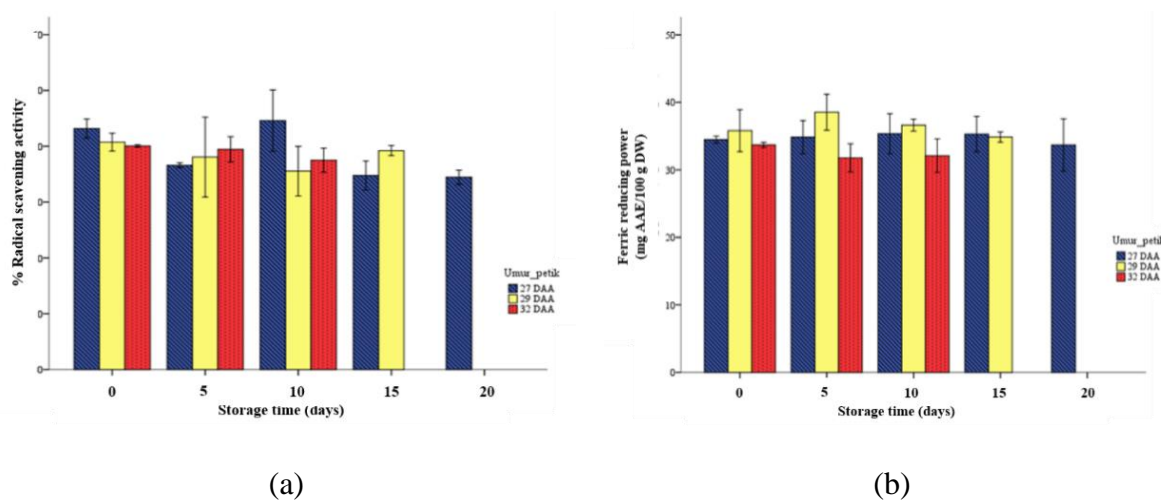


Figure 2. DPPH radical scavenging activity (a), Ferric Reducing Power (b) of *Cucumis melo* L. cv. Hikapel during postharvest storage at room temperature

Based on the DPPH-RSA the fruit of all ripening stages was capable to scavenge free

radicals via the hydrogen-donating mechanism. All samples indicated high

antioxidant potential as strong as Vitamin C at a concentration of 50 ppm. The level of DPPH radical scavenging activity decreased significantly up to S2 and stable to the end-stage (S3). The ferric reducing power may serve as a significant indicator of the antioxidant potential. This assay evaluates the ability of the extracts to reduce  $Fe^{3+}$  to  $Fe^{2+}$ , recorded as Perl's Prussian blue formation. All ripening stages of melon cv. Hikapel showed reducing power activity, which was more than 30 mg AA equivalent. The reducing power of the fruits was not significantly different among S1 and S2; S1 and S3.

In general, the antioxidant capacity of melon cv. Hikapel also decreased during on-tree ripening due to a decrease of the total phenolic, flavonoid, ascorbic acid content as observed in carambola (Zainuddin *et al.*, 2014) and durian (Arancibia-avila *et al.*, 2008). It is known that ascorbic acid and phenolic compounds, especially flavonoid compounds responsible for free radical scavenging. However, antioxidant activity does not only depend on phenolic concentration itself but also depend on any other compounds such as ascorbic acid, flavonoid, and carotenoid (Tavarini *et al.*, 2008; Gardner *et al.*, 2000).

Therefore, the antioxidant activities in fruit cannot be attributed solely to their phenolic contents, but also to the actions of different antioxidant compounds present in the fruits. There was a correlation between antioxidant activity and antioxidant contents of melon cv. Hikapel (data not shown). Antioxidant activity (DPPH-RSA, FRPA) was positively correlated with total phenolic, total flavonoid, and ascorbic acid content. It is also known that fruits with high antioxidant capacity generally contain more antioxidants and most of these antioxidants has been showing to be phenolic compounds and in particular flavonoid.

## CONCLUSION

All ripening stages of melon cv. Hikapel provides various dietary antioxidants such as phenolics, flavonoids, and ascorbic acid and showed free radical scavenging and reducing power activity. There were different levels of ascorbic acid, total phenolic, and total flavonoid content in different ripening stages of this melon. Antioxidant compounds and

antioxidant activity decreased during storage at room temperature. At the end of storage time, S1 has the highest antioxidant compound and capacity, whereas S3 has the lowest antioxidant compound and capacity. In conclusion, the ripening stage and postharvest storage time are important factors on the antioxidant properties of this melon cultivar Hikapel. Therefore, the grower should determine the appropriate harvesting time and postharvest storage duration at room temperature to meet commercial purposes with high antioxidant potential.

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# SCOPE, POLICY, AND AUTHORS GUIDELINES FOR FOOD SCIENTECH JOURNAL

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## Journal Scope

Food ScienTech Journal (FSJ) publishes high quality research articles on food sciences, food technology or its applications in food industry. The published articles can be in the form of research articles or short communications which have not been published previously in other journals(except in the form of an abstract or academic thesis/dissertation or presented in seminar/conference).

## Types of manuscript Research article

A research article is an original full-length research paper which should not exceed 5.000 words (including table and figures). Research article should be prepared according to the following order: title, authors name and affiliations, abstract, keywords, introduction, materials and method, result and discussion, conclusion, acknowledgement (optional), and references.

## Short communication or Review

A short communication or reviews up to 3.500 words (including table and figures) and consists of title, authors name and affiliations, abstract, keywords, introduction, materials and method, result and discussion, conclusion, acknowledgement (optional), and references. A short communication should contribute an important novelty for science, technology, or application.

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## Guideline for the manuscript content Title

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The abstract should state briefly background, material and method, the main findings supported by quantitative data which is relevant to the title, and the major conclusions.

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The introduction states background of the research supported mainly by the relevant references and ended with the objectives of the research.

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