ABSTRACT

Beta-carotene is a pigment that occurs widely and abundantly in nature. Beta-carotene can be found in some fruits and vegetables. Commonly, beta-carotene consistently bonds to other carotenoid compounds. Beta-carotene is a powerful colorant with beneficial effects on human health due to its ability to radical scavenging. Beta-carotene is obtained through the process of extraction with suitable solvent. The extraction method influenced the profile and quality of beta-carotene. The sonochemical approach using ultrasound, microwaves, and ohmic heating is eco-friendly and helps decrease the amount of solvent used, reduce the extraction time, increase the yield of beta-carotene from the sources, and increase the efficiency of the food applied. It is easier for consumers to accept natural dyes with high stability and efficiency. The present review describes detailed information about the quality of beta-carotene extract, isolation methods, and factors that affect the efficiency of natural food colorants applied to food products, which is helpful for the further development of food product formulations.

Keywords: antioxidant activity; beta-carotene; colorant; extraction method; pigment; fruit and vegetables; sensory profile

ABSTRAK


Kata kunci: aktivitas antioksidan; beta-karoten; pewarna; metode ekstraksi; pigment; buah dan sayur; profil sensori
INTRODUCTION

Beta-carotene, a plant pigment, may reduce cancer risk and promote healthy eyes and bones (Liu & Tang, 2016). Over 700 naturally occurring carotenoids or beta-carotene isomers have been identified, and 80-90% of carotenoids are obtained through fruit and vegetable consumption. Human can absorb β-carotene, β-cryptoxanthin, α-carotene, lycopene, lutein, and zeaxanthin in their diet (Maiani et al., 2009). Since the identification of beta-carotene's potency and function as vitamin A, research has been conducted on ways to improve the yield and quality of beta-carotene extracted from plant sources using various techniques (Ghazi, 1999; Strazzullo et al., 2007).

Beta-carotene, a fat-soluble pigment, is found in fruits, vegetables, and microbes. It is soluble in nonpolar solvents such as petroleum ether, hexane, and tetrahydrofuran depending on its polarity (Rivera and Canela, 2012; Susan, 2014). According to Tiwari and Sarkar (2018), the concentration of beta-carotene in carrots extracted using organic solvents ranges from 371.90 mg/L to 631.90 mg/L. The solvent used significantly impacts the beta-carotene extraction process and may have negative health and environmental impacts for humans. Eco-friendly extraction methods, such as maceration, should be used to reduce the environmental damage caused by organic solvents (Rivera and Canela, 2012; Susan, 2014). However, conventional techniques, including maceration, often require a large amount of solvent (Tiwari and Sarkar, 2018). Alternative methods for isolating beta-carotene include ohmic heating extraction (Aamir and Jittanit, 2017), microwave-assisted extraction (MAE), and ultrasound-assisted extraction (Chuyen et al., 2018; Kultys and Kurek, 2022).

Beta-carotene is commonly isolated from plant parts such as peel, seed, leaf, and flesh fruit, and it is also found in the metabolic pathway of algae and Escherichia coli (Kyriakopoulou et al., 2015; Yang and Guo, 2014). The main structure of beta-carotene isomers in food and human tissue is depicted in Figure 1. Algae are a preferred sustainable source of beta-carotene due to food safety and security. The extracted beta-carotene pigment should be added directly to food as a natural coloring agent. Beta-carotene is a pigment easily degraded by air and high temperatures, causing a color change from red to yellow (D’Evoli et al., 2013). The quality of beta-carotene can be assessed based on color, yield, and antioxidant activity using methods such as FRAP, ABTS, and liquid chromatography (Aamir and Jittanit, 2017; Chuyen et al., 2018; Mueller and Boehm, 2011).

![Figure 1. (a) Beta-Carotene Isomers in Food and Plant Tissue; and (b) Beta-Carotene Metabolite](image)

The color of food products affects consumer perception and can impact their quality (Šeregelj et al., 2022) Using beta-carotene as a food colorant may have several benefits for a product. Its antioxidant activity can increase the functional aspects of the product, and the sensory and functional qualities of food products can be improved by using beta-carotene-derived natural colors to promote public health.

EXTRACTION OF BETA-CAROTENE

Extraction is the process of isolating a chemical or pigment from a plant using a solvent. The extraction of beta-carotene from plants can be
facilitated through the use of organic materials because extraction can be used to separate plants from pigments. Solvents with various solubilities have been used to enhance the yield of the extract (Dai and Row, 2019). Most carotenoids are insoluble in water and soluble in organic solvents such as acetone (Rivera and Canela, 2012), alcohol, tetrahydrofuran (D’Evoli et al., 2013; Tonucci et al., 1995), ethyl ether (Mumtaz et al., 2019), chloroform, and ethyl acetate (Ludwig et al., 2021). However, their solubility depends on the availability of specific functional groups. Beta-carotene and lycopene have similar solubility in hexane, but they can be separated using a combination of methanol and tetrahydrofuran (6:4 v/v) to increase the yield of beta-carotene from maize (Rivera and Canela, 2012).

In the past decade, various techniques have been developed for the isolation of beta-carotene. For example, a mixture of beta-carotene and oil from Gac aril fruit (Momordica cochinchinensis) can be obtained using a combination of water and hexane with a ratio of 1:6, with distilled water used as a conductive medium for heating. Ohmic heating is a technique that uses the principle of generating heat by applying electrical energy to materials with electrical resistance (measured in ohms) using a heat conductor. The electrical energy is converted into heat due to the material’s electrical resistance, known as the Joule effect. (Aamir and Jittanit, 2017; Indiarto and Rezaharsamto, 2020). Ohmic heating can induce the rupture of the plant cell matrix, increasing the surface area between the powder and the solvent and allowing nonpolar components like beta-carotene to flow into the solvent. As a result, an ohmic heating method can be 245% more effective in extracting beta-carotene than a conventional extraction method (Aamir and Jittanit, 2017).

**Conventional**

The process of obtaining bioactive compounds from plant materials through the use of a solvent is referred to as solid-liquid extraction, though it is also known as leaching or lixiviation in chemical terminology (Sasidharan et al., 2018). Conventional extraction techniques can be divided into two categories: hot extraction by temperature, which involves the use of high temperatures (e.g., Soxhlet), and cold extraction, which does not use heat or high temperatures (e.g., maceration) (de Andrade Lima et al., 2019). Maceration, distillation, perforation, and reflux are conventional extraction methods that use a significant amount of solvent and have a prolonged duration, making them less efficient in the extraction process (Garcia-Vaquero et al., 2020; Khamitova et al., 2020; Uribe et al., 2015; Zhang et al., 2018), different methods will produce extracts with varying characteristics. Hot extraction (soxhlet, dist is faster but may result in lower antioxidant quality due to the sensitivity of the pigment to heat, while cold extraction takes longer but may produce extracts with higher antioxidant quality. Additionally, conventional extraction can be performed using a mechanical process, such as shaking and mixing at low temperatures (D’Evoli et al., 2013; Tonucci et al., 1995). One limitation of conventional techniques is the high solvent usage and the required prolonged extraction time. However, this method is simple for isolating beta carotene from solid matrices (Garcia-Vaquero et al., 2020). The quality of the beta-carotene yield extract may be influenced by various factors such as solid particle size, pressure, and co-solvent (Prado et al., 2013).

**Modern method**

Conventional extraction methods using solvents such as n-hexane, acetone, and ethyl alcohol for extracting beta-carotene from gac fruit flesh yield lower amounts of beta-carotene compared to microwave-assisted extraction (MAE) and ultrasound-assisted extraction (UAE) methods using ethyl acetate. Extraction using microwave-assisted (MAE) and ultrasound-assisted (UAE) methods yields higher amounts of beta-carotene compared to the maceration method, with values of 2623 ± 35 mg/L and 2677 ± 12 mg/L, respectively (Chuyen et al., 2017). Eco-friendly extraction methods yield nearly three times more beta-carotene than the maceration method.

The extraction of beta-carotene using microwave-assisted extraction (MAE) has several advantages including stable temperature conditions, pure yield extract, faster extraction compared to conventional
methods, and low energy requirements. (Chuyen et al., 2018; Elik et al., 2020; Kultys and Kurek, 2022). The use of ultrasonic waves, known as cavitation forces, in the UAE method results in increased diffusion of beta-carotene and high antioxidant activity. The cavitation effect causes bubbles to form on the solid surface of the material matrix in a liquid solvent medium, creating a microjet flow that increases the rate of mass transfer from inside the cell wall to the solvent. The UAE extraction process can be further accelerated by using suitable solvents. In comparison, the MAE technique involves the use of microwave energy and nonpolar solvents to extract beta-carotene, resulting in a purer yield extract and faster process with lower energy requirements (Abid et al., 2021; Chuyen et al., 2018; Jha et al., 2017; Lavilla and Bendicho, 2017; Li et al., 2013; Mumtaz et al., 2019). In addition, UAE extraction has a significant antioxidant activity due to various polyphenols extracted during the processing.

The yield of beta-carotene extract from vegetables and fruits is influenced by the number of components extracted during the extraction process. The suitability of the solvent with the ingredients being extracted plays a significant role in the success of the extraction process. Polar components dissolve in polar solvents and nonpolar components dissolve in nonpolar solvents. N-hexane, a nonpolar solvent, is effective for extracting nonpolar materials like beta-carotene (Arion et al., 2017; Gucu1ü-Ustünda1 and Temelli, 2005) The solubility of beta-carotene in organic solvents, such as carbon disulfide, benzene, chloroform, acetone, ether, and petroleum ether, can be improved by the addition of aceton. The combination of polar acetone and nonpolar n-hexane is most effective for extracting beta-carotene. Some studies have shown that essential oils and certain combinations of semi-polar solvents can be used as alternatives to n-hexane to extract beta-carotene from cells surrounded by aqueous fluid (Grootaert et al., 2021; Ordóñez-Santos et al., 2015).

The concentration of beta-carotene in fruits and vegetables can vary due to factors such as cultivar, source, fruit clone, and extraction process. Dry extraction processes that use high heat and treatment can cause carotene degradation. These factors should be considered when using beta-carotene as a food additive (Lasunon et al., 2021). Carotene is highly vulnerable to isomerization and heat-induced oxidation by the conjugated double-bond system. The cis isomer of beta-carotene, which has provitamin A and lowers antioxidant activity, can be altered by high pressure, mechanical processes (such as mixing), the addition of essential oil, or replaced by more efficient methods. A summary of beta-carotene extraction methods for various sources can be seen in Table 1.

**THE USE OF BETA-CAROTENE AS A FOOD DYE**

The food industry often uses synthetic dyes to improve the appearance of products, as they are typically more cost-effective and efficient than natural colors. However, research suggests that excessive consumption of synthetic colors may negatively affect health, including allergies and abnormalities, as well as an increased risk of cancer. Natural colorants, such as beta-carotene, can be used to enhance the color of food without these potential negative consequences (Kobylewski and Jacobson, 2012).

Beta-carotene is a pigment colorant with the highest provitamin A activity. In addition to its role as a pigment, beta-carotene can be metabolized into the vitamin A by-products retinoic acid (RA) and retinal. RA is a regulator of various biological functions and is recognized by two classes of nuclear receptors: retinoic acid receptors (RARs) and retinoid X receptors (RXRs) (Grune et al., 2010; von Lintig et al., 2005). Since (9Z)-beta carotene is a precursor of (9Z)-RA, the ligand for RXRs, isomers (all-E)-beta carotene is a precursor for (all-E)-RA, it binds to RAR (von Lintig, 2020; von Lintig et al., 2005). Natural colorants also have biological features of antimicrobials. Applying natural colorants to food can have additional benefits for extending the product's shelf life (Assadpour et al., 2020; Cavalcanti et al., 2022). Beta carotene can be obtained naturally from raw materials like sweet potato, yam, rice, maize, pumpkin, and potatoes, or it can be added as a food.
enhancer or emulsion product (de Andrade Lima et al., 2019; Jati et al., 2022; Rivera and Canela, 2012; Tang et al., 2012).

### Table 1. Extraction of Beta-Carotene

<table>
<thead>
<tr>
<th>Source</th>
<th>Part of fruit</th>
<th>The Extraction method and solid ratio matrix by solvent</th>
<th>Condition extraction</th>
<th>Advantage</th>
<th>Total beta-carotene (mg/L)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carrot</td>
<td>Whole</td>
<td>Maceration (1:5)</td>
<td>n-hexane at ambient temperature for 6 hours</td>
<td>Simplest without specialization apparatus</td>
<td>321.35</td>
<td>(Li et al., 2013)</td>
</tr>
<tr>
<td>Carrot</td>
<td>Whole</td>
<td>UAE (1:5)</td>
<td>Sunflower oil at 40°C temperature for 20 minutes</td>
<td>Rapidly processed and higher beta-carotene</td>
<td>334.75</td>
<td>(Li et al., 2013)</td>
</tr>
<tr>
<td>Carrots</td>
<td>Whole</td>
<td>Maceration</td>
<td>n-hexane and acetone (1:1) at ambient temperature</td>
<td>Simplest without specialization apparatus</td>
<td>33.7</td>
<td>(Rifqi et al., 2020)</td>
</tr>
<tr>
<td>Cherry</td>
<td>Whole</td>
<td>Maceration</td>
<td>Tetrahydrofuran, calcium carbonate, and dichloromethane</td>
<td>Simplest without specialization apparatus</td>
<td>10.0 ± 0.5</td>
<td>(D’Evoli et al., 2013)</td>
</tr>
<tr>
<td>Gac fruit</td>
<td>Aril</td>
<td>Maceration (1:7)</td>
<td>n-hexane at 50°C for 7 hours</td>
<td>Simplest without specialization apparatus</td>
<td>39.6</td>
<td>(Aamir and Jittanit, 2013)</td>
</tr>
<tr>
<td>Gac fruit</td>
<td>Aril</td>
<td>Ohmic heating (1:7)</td>
<td>Water and n-hexane (1:6), 50 V at 50°C for 7 hours n-hexane:acetone:ethanol</td>
<td>Yield extract is 91% higher</td>
<td>582.2</td>
<td>(Aamir and Jittanit, 2017)</td>
</tr>
<tr>
<td>Gac fruit</td>
<td>Fresh</td>
<td>Maceration (1:10)</td>
<td>Ethyl acetate at 40.7°C for 30 minutes</td>
<td>Simplest without specialization apparatus</td>
<td>907 ± 22</td>
<td>(H. V Chuyen et al., 2017)</td>
</tr>
<tr>
<td>Gac fruit</td>
<td>Dried</td>
<td>Maceration (1:80)</td>
<td>Ethyl acetate, 120 W at 60°C for 25 minutes</td>
<td>The temperature of extraction is relative stable, low energy needed, and rapidly processed by adding water as an ohmic heating medium</td>
<td>2623 ± 35</td>
<td>(Chuyen et al., 2018)</td>
</tr>
<tr>
<td>Gac fruit</td>
<td>Dried</td>
<td>MAE (1:80)</td>
<td>Ethyl acetate, 200 W at 60°C for 80 minutes</td>
<td>Great antioxidant activity, rapidly processed, higher yields, and without using an extra medium solvent or co-solvent</td>
<td>2677 ± 12</td>
<td>(Chuyen et al., 2018)</td>
</tr>
<tr>
<td>Gac fruit</td>
<td>Dried</td>
<td>UAE (1:80)</td>
<td>Hexane and acetone (1:1) at ambient temperature for 15 minutes</td>
<td>Simplest without specialization apparatus</td>
<td>2700</td>
<td>(Ghazi, 1999)</td>
</tr>
<tr>
<td>Orange fruit</td>
<td>Peel</td>
<td>Maceration (1:15)</td>
<td>Chloroform and methanol (2:1) at room temperature for 1 hour</td>
<td>Simplest without specialization apparatus</td>
<td>54.6</td>
<td>(Sarangallo et al., 2015)</td>
</tr>
<tr>
<td>Pandanus</td>
<td>Pericarp</td>
<td>Maceration</td>
<td>Acetone: ethanol in petroleum ether (2:1:1) at ambient temperature</td>
<td>Simplest without specialization apparatus</td>
<td>42.2</td>
<td>(Mumtaz et al., 2019)</td>
</tr>
<tr>
<td>Conoideus</td>
<td>OIl</td>
<td>Maceration</td>
<td>Sunflower oil, 80 W at 35°C for 30 minutes</td>
<td>The solvent was used as more eco-friendly with a yield similar to organic solvent and rapidly processed</td>
<td>1634.7</td>
<td>(Ordóñez-Santos et al., 2015)</td>
</tr>
<tr>
<td>Papaya</td>
<td>Flesh</td>
<td>Maceration</td>
<td>Tetrahydrofuran</td>
<td>Simplest without specialization apparatus</td>
<td>2.3 ± 0.4</td>
<td>(Tomucci et al., 1995)</td>
</tr>
<tr>
<td>Peach fruit</td>
<td>Peel</td>
<td>UAE (1:2)</td>
<td>Simplest without specialization apparatus</td>
<td>Simplest without specialization apparatus</td>
<td>2.3 ± 0.4</td>
<td>(Tomucci et al., 1995)</td>
</tr>
</tbody>
</table>

Beta-carotene can be made more stable and soluble for use as a food colorant by evaluating it as an emulsion product. However, the use of a single emulsifier to create an oil-in-water emulsion of beta-carotene may not be suitable for industrial food production due to sensitivity to various factors.
such as temperature, light, pH, heat, oxygen, and free radicals. Microencapsulation of beta-carotene can also be used to act as a fat replacer in cupcakes and enrich the product, potentially replacing up to 36% of the used fat. (Mueller and Boehm, 2011; Nayana et al., 2021). Table 2 summarizes the use of beta-carotene as a food dye.

### Table 2. The use of Beta-Carotene as a Food Dye

<table>
<thead>
<tr>
<th>Sample</th>
<th>Treatment</th>
<th>Parameter</th>
<th>Research result</th>
<th>Preferences</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flake product with the addition of Orange Sweet Potato and Red Rice</td>
<td>Ratio Orange (100:0, 80:20, 60:40, 40:60, 20:80, and 0:100)</td>
<td>Total beta-carotene, antioxidant activity, color intensity, and sensory profile</td>
<td>Flake product with the addition of orange sweet potato and red rice (60:40) has total beta-carotene of 25.77 mg/L; The antioxidant activity of 85% is in a strong category; The best color intensity is in flakes with a ratio of orange sweet potato and red rice (100:0), as well as on the sensory profile of all flakes favored by consumers.</td>
<td>(Jati et al., 2022)</td>
</tr>
<tr>
<td>Ras Cheese with adding 99% beta-carotene of carrots pells</td>
<td>Ras cheese with 99% beta-carotene of carrots pells</td>
<td>Sensory evaluation of Ras cheese during 180 days of storage</td>
<td>Consumers enjoy the sensory characteristic of ras cheese, which has a yellowish hue and a smooth, slightly salty flavor after being held for 180 days.</td>
<td>(Al-Surmi et al., 2021)</td>
</tr>
<tr>
<td>Orange-fleshed sweet potato composite flour bread</td>
<td>Orange-fleshed sweet potatoes (56–70 grams in 100 grams)</td>
<td>Antioxidant activity</td>
<td>Orange-fleshed sweet potato composite flour bread has antioxidant activity ranging from 12.31 mg/L to 40.36 mg/L.</td>
<td>(Oloniyo et al., 2021)</td>
</tr>
<tr>
<td>Incorporated beta-carotene in palm and flaxseed oil as an emulsion to develop cupcake</td>
<td>Replaced oil with microencapsulation powder oil for formulation cupcakes</td>
<td>Color texture profile and sensory properties</td>
<td>The texture, color, and sensory evaluation of cupcakes using microencapsulation oil are similar to butter as the fat.</td>
<td>(Nayana et al., 2021)</td>
</tr>
<tr>
<td>Functional mangiferin (mango peel extract) drink</td>
<td>The drink contains mangiferin extract</td>
<td>Antioxidant activity, beta-carotene content, physicochemical properties (pH, total soluble solid, total acidity) and sensory properties using the hedonic test (color, sweetness, sourness) during 2 months of storage</td>
<td>Mangiferin functional drink is more stable than control after 2 months of storage-based sensory evaluation and physicochemical properties</td>
<td>(Imran et al., 2016)</td>
</tr>
</tbody>
</table>
### Nopal marmalades

Optimizing the base mix (raw material and water) and supplementary materials by: 87.40%, 79.40%, 66.30%, 66.00%

- Antioxidant activity, beta-carotene, and sensory properties (color, odour, flavour, spreadability) at different temperatures
- The formulation of 66.30% base mix, adding 33.0% lactitol, and aspartame 0.1%, has been more accepted by the customer than other formulations and is similar to standard marmalades (Leopoldo et al., 2012).

### Enrichment yoghurt with carrot juice and sugar

Yoghurt with added 10% and free sugar; concentration of carrot juice (0%, 15%, and 20%)

- Viscosity, syneresis, titratable acidity, pH, yeast and mould counts, color measurements, beta-carotene contents and sensory qualities during 21 days of storage time
- The best treatment for all parameters is yoghurt with 10% sugar and 15% carrot juice; had beta-carotene content appropriate to 80 µg/100 g (Cakmakci et al., 2014).

### Beta-carotene fortified Gari based Orange-fleshed sweet potato (OFSP) root composite flour

Optimizing the substitution of OFSP composite flour and fermentation time

- Swelling capacity, taste, texture, flavour, color, beta-carotene content
- The optimization of OFSP composite adding is 10% with 43 hours of fermentation to provide gari: 16.72 µg/ml beta-carotene content, 85.16 brightness, 40.2 yellowness, 4.93 redness, 350% Swelling capacity, 6.75 scores for appearance, 7.0 for taste, 7.35 for texture, 7.0 for flavour, and 7.2 for overall acceptability (Cakmakci et al., 2014).

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Beta-carotene levels dropped in the flake product, including orange sweet potato and red rice. The heat degradation of beta-carotene may be the cause of this occurrence. Carotenoids are further recognized as being light- and heat-sensitive compounds. Red rice and orange sweet potatoes both contain different kinds of carotenoids. After boiling, there was an increase in carotenoids, which could be related to the thermal disruption of the protein-carotenoid system (Trono, 2019). They have sophisticated and matrix-based carotene release. The amount of orange sweet potato used in the flake formulation impacted the carotenoid level. The amount of carotenoid in the flakes increases with the percentage of orange sweet potato.

The fortifications of the food product by adding the sources of beta-carotene (fruit and vegetable) would change the chemical composition of polymer binding, i.e., protein, carbohydrate, and lipid to the hydrogen bound or matrices of the intramolecular network in a food product (Abano et al., 2020; Cakmakci et al., 2014). The inclusion of orange-fleshed sweet potato flour in Gari resulted in a low swelling capacity due to the weak protein and carbohydrate network's ability to absorb water. Beta-carotene, which is sensitive to heat and may change color as a result, significantly affects the intensity of the color. Observations show that the hue of beta-carotene can range from yellow to red (Trancoso-Reyes et al., 2016). The Maillard process can alter the color of flakes, with more significant Maillard reaction products resulting in a darker color. Many panelists found the dark orange hue of beta-carotene-containing food to be unappealing, unappetizing, and less fresh in terms of sensory characteristics. Beta-carotene can improve the brightness and redness of a product, increasing color preference. Most panelists
preferred the reddish and brighter flakes. The white endosperm of the material is associated with a higher level of brightness.

As shown in previous research, the breakdown of carotenoids during food processing is caused by heat treatment. This can also decrease the antioxidant activity of the carotenoids. The sensitivity of bioactive substances to heat can also impact their antioxidant activity, as there is a positive correlation between the decrease in bioactive substances and the decrease in antioxidant capability (Bagchi et al., 2021). According to a recent study, rising temperatures hasten the start of oxidation, impairing the effectiveness of antioxidants. Bioactive chemicals deteriorate, experience structural modifications, and ultimately become inert substances.

CONCLUSION

In summary, beta-carotene is a pigment found in plants with several health benefits, including reducing cancer risk and promoting healthy eyes and bones. It is commonly extracted from plant parts using organic solvents, but eco-friendly extraction methods, such as maceration, are also available. Alternative methods for extracting beta-carotene include ohmic heating, microwave-assisted, and ultrasound-assisted extraction. Beta-carotene can be used as a natural colorant in food products and has antioxidant activity. The solubility of beta-carotene depends on the solvent used and the presence of specific functional groups. Beta-carotene is easily degraded by air and high temperatures, and its quality can be evaluated based on color, yield, and antioxidant activity. The use of beta-carotene as a food colorant can improve the sensory and functional qualities of food products and promote public health.

ACKNOWLEDGEMENT

The author would like to gratefully acknowledge Djuanda University Bogor, Universitas Padjadjaran, IPB University (Bogor Agricultural University), The Ministry of Education and Culture of The Republic of Indonesia for the support provided, and The Research Center for Appropriate Technology, National Research, and Innovation.

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