

Difference in Harvesting Time for The Chemical Quality of Red Betel Leaf (*Piper crocatum* Ruiz & Pav.

Perbedaan Waktu Pemanenan Terhadap Mutu Kimia Daun Sirih Merah (*Piper crocatum* Ruiz & Pav.)

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ABSTRACT

Chemical quality of plants can be affected by harvest time. Harvest time is right when the plant parts contain the largest number of active compounds. This study aims to analyze the effect of differences in harvest time on the chemical quality of red betel leaf. The research was conducted from March to May 2019 in Post harvest laboratory (TPP) at Sultan Syarif Kasim State Islamic University and Agricultural Product Technology (THP) Universitas of Riau. The treatments given were time harvest at 07.00 WIB (W1) (Morning), 11.00 WIB (W2) (Afternoon) and 15.00 (W3) (Evening). The study design used a non factorial Complete Random Design (CRD) consisting 6 replications. Extraction used ethanol solvent for 24 hours by immersion. Data were analyzed by analysis of variance and if it had real effect it was followed by Duncan Multiple's Range Test (DMRT). The results of this study concluded that treatment of the afternoon (15.00 WIB) harvest time had a significant affect of red betel leaves chemical quality including alkaloids (5,87 mg/g), total anthocyanin (8,50 mg/g), moisture content (14,54%), total dissolved solids (1,34 °Brix) and color content (153%). The results showed that harvesting time in the afternoon at 15.00 WIB was able to improve the chemical quality of red betel leaves. Chemical quality of red betel leaves accumulates in the afternoon.

Keywords: red betel, harvest time, chemical quality, extraction.

ABSTRAK

Mutu kimia tanaman dapat dipengaruhi oleh waktu pemanenan. Waktu panen yang tepat pada saat bagian tanaman tersebut mengandung senyawa aktif dalam jumlah terbesar. Tujuan penelitian untuk menganalisis pengaruh perbedaan waktu pemanenan terhadap mutu kimia daun sirih merah. Penelitian telah dilaksanakan pada Bulan Maret sampai Mei 2019 di Laboratorium Teknologi Pasca Panen (TPP) Universitas Islam Negeri Sultan Syarif Kasim Riau dan Laboratorium Teknologi Hasil Pertanian (THP) Universitas Riau. Perlakuan yang diberikan adalah waktu pemanenan terdiri atas pukul 07.00 WIB (W1) (Pagi), 11.00 WIB (W2) (Siang) dan 15.00 WIB (W3) (Sore). Rancangan penelitian menggunakan Rancangan Acak Lengkap (RAL) non faktorial terdiri atas 6 ulangan. Ekstraksi menggunakan bahan pelarut etanol 96% selama 24 jam dengan cara direndam. Data dianalisis dengan sidik ragam dan apabila berpengaruh nyata maka dilanjutkan dengan uji Duncan Multiple's Range Test (DMRT). Pengamatan yang dilakukan terdiri atas Alkaloid, Total Antosianin, Kadar Air, Total Padatan Terlarut dan Warna. Hasil penelitian menunjukkan perlakuan (W3) waktu panen 15.00 WIB (Sore) memberi pengaruh nyata dalam meningkatkan mutu kimia daun sirih merah dengan nilai rata-rata alkaloid (5,87 mg/g), total antosianin (8,50 mg/g), kadar air (14,54%), total padatan terlarut (1,34 °Brix) dan warna (153%). Hasil penelitian menunjukkan bahwa waktu pemanenan pada sore hari Pukul 15.00 WIB mampu meningkatkan mutu kimia daun sirih merah. Mutu kimia daun sirih merah terakumulasi pada sore hari.

Kata kunci: sirih merah, waktu panen, mutu kimia, ekstraksi

INTRODUCTION

Red betel plant (*Piper crocatum* Ruiz & Pav.) has long been known as a medicine and is widely grown in Indonesia. The part of the red betel plant that is used as medicine is the leaves. Red betel leaf has been known to have various properties to cure various diseases (Ma'rifah, 2012). Betel has more than 700 species in tropical America and 340 species in tropical Asia, including Indonesia (Arbain dkk., 2018).

Red betel, in traditional medicine is widely used for the treatment of hypertension, inflammation of the liver, inflammation of the prostate, inflammation of the eyes, vaginal discharge, ulcers, breast cancer, joint pain, lowering and controlling blood sugar levels, cosmetics, heart disease drugs, Bone tuberculosis, vaginal discharge, breast tumors, antiseptics to eliminate microorganisms from the skin or wounds, for example caused by *Candida albicans*, as a mouthwash can help prevent dental plaque formation and gingivitis and cough medicine expectorants (Parfati dan Windono, 2016).

Red betel leaves contain carotene, xanthophyll, chlorophyll a, chlorophyll b and anthocyanins (Muthoharoh, 2011) also flavonoids, alkaloids, polyphenolic compounds, tannins, carvacrol and eugenol essential oils (Sudewo, 2006). Alkaloids are secondary metabolites that are mostly produced by plants and have antibacterial properties (Ma'rifah, 2012).

The flavonoid compounds in red betel leaf are anthocyanins which are antioxidants and can bind *Free Radical Scavenging* (Hayati et al., 2012). Anthocyanins are colored pigments commonly found in red, purple and blue flowers, fruits, stems, leaves and roots (Ariviani, 2010).

Traditional medicine contains a lot of chemicals and generally it is not known or certain that the active substances play a role in causing the therapeutic effect. The chemical content of plants can be influenced by geographic location, climate, cultivation method, harvest time, harvest method and post-harvest treatment method. One of the factors that influence the chemical content of the betel plant is harvest time (Dewoto, 2007).

Harvest time is closely related to the formation of active compounds in the plant parts to be harvested. The highest nitrite content in spinach at noon was 74.40 mg/g (Aryanda, 2017). Harvest time increases the polyphenol content in *Talinum triangulare* Jacq. (Brasileiro et al., 2015).

Harvest time significantly increased the total anthocyanin production of *Gynura segetum*'s leaves, namely 13.41 mg / plant (Tripatmasari dkk., 2014). The optimum sugar content of sweet seed corn at harvest at 17.00 in the afternoon is 14.82% (Surtinah, 2012).

Red betel ready to harvest at least 4 months old. Red betel consists of 16-20 leaves. The leaves are relatively wide, 15-20 cm long. Harvest time of red betel based on the location of the leaves consists of young leaves (third leaf from shoot), medium leaves (sixth leaf from shoot) old leaves (eighth leaf from shoot). The harvest time for red betel is at moderate physiological age, not too old or young, because of its high levels of active substances (Agoes, 2019). The research objective was to analyze the effect of different harvesting times on the chemical quality of red betel leaves.

MATERIALS AND METHODS

Place and time

This research was conducted from March to May 2019 at Post-Harvest Technology Laboratory of Faculty of Agriculture and Animal Husbandry of Universitas Islam Negeri Sultan Syarif Kasim Riau and Agricultural Product Processing Laboratory of Universitas Riau.

The materials used are red betel leaf, distilled water, 96% ethanol. The tools used were the oven, the maceration bottle, the M150 bottle, cutter knife, scissors, label paper, drop pipette, beaker glass, test tube, measuring glass, beaker, Burette, hand refractometer Abbe, cool box Marina, spectrophotometer Shimadzu Uv mini-1240, desiccator vacuum Duran, Erlenmeyer, analytical scales, stationery, cameras, tissues, masks and filter paper. The study used a completely randomized design (CRD) which consisted of 3 treatments, namely harvesting time (W) and repeated 6 times. W1 = 07.00 WIB (Morning), W2 = 11.00 WIB (Noon), W3 = 15.00 WIB (Afternoon), so we get $3 \times 6 = 18$ experimental units. Each experimental unit consisted of 100g of red betel leaves so that $18 \times 100 \text{ g} = 1800 \text{ g}$ of red betel leaf were obtained.

Red betel leaves come from Ujung Batu District, Rokan Hulu, Riau. How to pick red betel leaves using sterile scissors spray using technical alcohol. The red betel leaves are washed from dirt using running water three times and then dried. Red betel leaves are taken to the Postharvest Technology Laboratory using land transportation

and a cool box to maintain sample quality during the trip.

Weighing red betel leaves with a weight of 100 g per treatment. Red betel leaves are chopped manually using a cutter knife with a size of 1 cm. Red betel leaves are put into a dark container then added with 96% ethanol extract. Comparison of ingredients and solvents 1: 5 (Senja dkk., 2014).

Then carried out maceration for 1 x 24 hours, stirring occasionally every 2 hours. The maceration process is carried out in a dark colored container which is tightly closed. The obtained macerate is filtered with filter paper. After that, observations were made with 5 variables consisting of alkaloids, analysis of total anthocyanins, water content, total dissolved solids and color.

RESULTS AND DISCUSSION

Cultivation of Red Betel Plants

Cultivation of red betel uses a planting medium in the form of soil, sand and compost in the ratio 1: 1: 1. All planting media materials are mixed first, sieved to clean dirt and rocks. Then put into polybags with a diameter of 10 cm. Seeding is carried out by cuttings of old red betel stems, which are marked by the size of the stems are large and the color of the leaves is dark red. Two book-sized, three-leaf cuttings.



Picture 1. The netting wire around which the tendrils are wrapped

Before planting, soak the cuttings in water for 15 minutes, so that the cuttings absorb enough water to last the growing process. The cuttings were planted in the media for two weeks. The location of the leaf buds is facing upwards. Keep the book (the location of the leaves, buds and fiber roots) covered with the media 0.5-1 cm deep from the surface of the media. After the cuttings are two weeks old \, the cuttings are ready to be transplanted into soil, sand and compost 1: 1: 1 planting medium on 30 x 20 cm polybags. The polybags that have been planted are given 1 m

high stake and 2 m high mesh wire is made on the walls of the house as a place to wrap the red betel plant.

Maintenance includes weed cleaning, lighting and watering. Red betel grows well in shady areas and does not have too much sun. Red betel will grow well when it gets 60-75% sunlight (Hermiati dkk., 2013). Red betel is placed beside the shaded house yard. Watering is done every morning and evening. When it rains, no watering is done. Weeds that grow around the betel are cleaned regularly, so as not to inhibit the growth of red betel.

Harvesting is done after the betel plant is 4 months old. It has strong roots, sturdy stems and relatively wide and fresh leaves. Harvesting is carried out according to the treatment consisting of morning (07.00 WIB), afternoon (11.00 WIB) and evening (15.00 WIB). Red betel leaf harvest criteria are based on the location of the leaves (sixth, seventh and eighth leaves from the shoot).

Alkaloid

Alkaloids are active plant substances that function as drugs and strong activators for immune cells that can destroy bacteria, viruses, fungi and cancer cells. (Olivia dkk., 2006). As an alkaloid antifungal it causes damage to cell membranes. Alkaloids will bind strongly to ergosterol to form holes or channels, causing the cell membrane to leak and lose some intra-cell material such as electrolytes, especially potassium and small molecules. This results in damage to cells and death in fungi (Candrasari dkk., 2011). The average alkaloid values in red betel leaf based on different harvest times can be seen in Table 1.

Table 1. Alkaloid Average of Red Betel Leaf Based on Harvesting Time Differences

| Treatment | Alkaloid (mg/g) ± Stdev |
|----------------|----------------------------|
| W ₁ | 0.59 ± 0.16 ^c |
| W ₂ | 0.84 ± 0.00 ^b |
| W ₃ | 0.98 ± 0.00 ^a |

Note: Different superscripts in the same column show very significant differences (p <0.01)..

Based on the results of the Duncan Multiple Range Test (DMRT), it shows that the highest alkaloid levels are at the time of harvesting in the afternoon at 15.00 WIB (W₃) with an average value of 0.98 mg / g. Meanwhile, alkaloid levels with the lowest average value were

harvesting in the morning at 07.00 WIB (W1) with an average value of 0.59 mg / g. From these results indicate that the optimum alkaloid content of red betel leaves at the afternoon harvest at 15.00 WIB (W3).

The difference in alkaloid levels in red betel leaves is caused by metabolic processes that take place in the morning, afternoon and evening. Alkaloid levels accumulate in the afternoon. According to Aryanda (2017), the longer the harvesting time, the higher the chemical content of the plant. The afternoon is the optimum harvesting condition, this is because the many elements are absorbed by the plant over the length of harvesting time and other factors that support such as humidity, soil conditions, weather and weather intensity.

Alkaloids are secondary metabolite compounds derived from amino acid precursors. Secondary metabolites come from primary biosynthesis. Generally, takes place in the morning and the starting material is water and carbon dioxide (CO₂) (photosynthesis). Photosynthesis is the basic biochemical process that underlies life. Water is the starting material for alkaloid formation (Saifudin, 2014).

Alkaloids are the most common organic compounds found in nature. All alkaloids come from plants with various levels (Tengo dkk., 2013). Based on the identification results using TLC obtained 6 isolates which stained the TLC plate. All of them provide an R_f value that matches the standard for alkaloid compounds, namely 1. The value of R_f can be defined as the distance traveled by the compound from the origin divided by the distance traveled, therefore the R_f number is always less than 1 (one) (Windayani, 2014).

Alkaloid content of several types of plants, jembirit leaves (*Tabernae sphaerocarpa* BL.) with the maceration extraction method is 0.727 mg / g (Salamah dkk., 2017). *Carica papaya* L. amounting to 0.029 mg / g (Mukhaimin et al., 2018). The total alkaloid in leaf linkages was 0.230 (Kadarwenny, 2017).

Total Anthocyanin Analysis

Anthocyanins are the pigments that cause almost all of the red to blue colors in the flowers, leaves and fruit of plants. Anthocyanin extract color differs depending on the pH of the medium. In the acidic medium the color is red, the base medium is blue and the neutral medium is purple. Anthocyanins have high antioxidant activity, can

prevent cancer, reduce the risk of coronary heart disease and prevent cataracts (Lestario, 2017). The average value of the total anthocyanin analysis based on different harvesting times can be seen in Table 2.

Table 2. Mean Total Anthocyanin Analysis Based on Harvesting Time Differences

| Treatment | Total |
|----------------|---------------------------|
| | Antosianin (mg/g) ± Stdev |
| W ₁ | 4.74 ± 0.65 ^c |
| W ₂ | 7.69 ± 0.34 ^b |
| W ₃ | 8.50 ± 0.40 ^a |

Note: Different superscripts in the same column show significant differences (P <0.01).

The results of the Duncan's Multiple Range Test (DMRT) continued test showed that the highest total anthocyanin levels were at harvest time at 15.00 WIB (W3) with an average value of 8.50 mg / g, while the lowest total anthocyanin levels were at morning harvesting time. day at 07.00 WIB (W1) with a mean value of 4.74 mg / g.

According to Lestario (2018), The formation of anthocyanins in plants depends on a number of environmental factors such as light, temperature, water availability, sugar supply, injury and infection. The most important factor of all is light. Plants that are exposed to sunlight for a long time have a high total anthocyanin content. The effect of light on anthocyanin synthesis is seen in the activity of several different enzymes involved in their biosynthesis. Anthocyanin synthesis is directly affected by the activity of the enzyme Phenylalanine ammonia-lyase (PAL).

Anthocyanins in plants are located in vacuole cells, most anthocyanins are found and can be taken from several plant organs, such as flower crowns, leaves, fruit, seeds, and tubers. In various types of fruit and tubers, anthocyanins are not only contained in the pulp and tubers, but also in the skin. The color in anthocyanins is not only a differentiator, but also important information regarding its nutritional content. The darker or stronger the color produced on the plant indicates that the greater the concentration of anthocyanins in the plant (Priska dkk., 2018).

Anthocyanin content is also influenced by nutrients in the form of sugars and phytohormones. Anthocyanin synthesis requires free sugar. The presence of sugar is a triggering of

anthocyanin accumulation. There is a close relationship between sugar levels and anthocyanin levels. The accumulation of sugar initiates an increase in anthocyanin levels. Phytohormone (ethylene) is a ripening hormone, increasing the accumulation of anthocyanins in plants (Gross, 1987).

According to National Agency of Drug and Food Control (2013), regarding the maximum limit for the use of food additives that the number of anthocyanins allowed in food is 2.5 (SN) body weight. This means that per kilogram of body weight people can only consume 2.5 mg of anthocyanins. If the average body weight is 50 kg, the amount of anthocyanins that can be consumed is 125 mg.

Anthocyanin content varies in plants, in flowers: roses (0.925% / 10 g), hibiscus (0.739% / 10 g), four o'clock (0.977% / 10 g). Leaves, fruit and tubers, anthocyanins are found in caladium leaves (0.057 mg / g), red spinach (6350 ppm on leaves and 2480 ppm on stems), purple sweet potato (11.02 mg / g fresh sweet potato), red cabbage (185 mg / 100 g fresh ingredients), strawberries (20.8 mg / g), grapes (190 mg / 100 g), mulberry (1993 mg / 100 g), dragon (8.8 mg / 100 g dragon fruit flesh), and jamblang (161 mg / 100 g of fresh ripe jamblang flesh) (Priska dkk., 2018).

Water Content

Water content is the amount of water contained in the material and is expressed in percent. Water content is one of the most important characteristics of food, because water can affect the appearance, texture and taste of fruit. (Winarno, 2004). The results of the variation in moisture content showed that the difference in harvesting time was significantly different from the moisture content of red betel leaves. The average value of water content in red betel leaves based on different harvesting times can be seen in Table 3.

Table 3. Average Moisture Content Based on Harvesting Time Differences

| Treatment | Water Content (%) ± Stdev |
|----------------|---------------------------|
| W ₁ | 16.21 ± 0.61 ^a |
| W ₂ | 15.19 ± 0.33 ^b |
| W ₃ | 14.54 ± 0.50 ^c |

Note: Different superscripts in the same column show significant differences (P <0.01).

Based on the results of the Duncan's Multiple Range Test (DMRT), it showed that the highest water content was at the morning harvest time at 07.00 WIB (W1) with an average value of 16.21%, while the lowest water content was at the afternoon harvest time at 15.00 WIB (W3) with an average value of 14.54%.

Reduced water content in plants is caused by temperature and the plants carry out the transpiration process. The process of transpiration is the process of losing water due to evaporation through the inside of the plant body, namely the water absorbed by plant roots is used to form tissue and then released through the leaves into the atmosphere. (Purba, 2011).

The transpiration process is influenced by external and internal factors. External factors are wind speed, humidity, temperature, sunlight, air pressure. Factors in the thickness of the leaves, the number of stomata, presence of cuticles, the number of trichomes / leaf hairs (Haryanti, 2010). Temperature is a factor that affects evaporation. At high temperatures many molecules have the energy to evaporate. The high temperature causes the molecules to move at high speed so that beyond the attractive forces in the liquid or solid, the water molecules will come out through the surface and become a gas (Putri dkk., 2017).

In the morning the ambient temperature is still in balance with the plant's body temperature, so that the evaporation of plant water is still controlled. During the day the temperature has risen while the temperature of the plant is still low, therefore the plant must reduce its evaporation (transpiration)(Haryanti dan Meirina, 2009).

From these results indicate that the water content of red betel leaves in the entire sample is > 10%. These samples do not meet the quality characteristics of red betel leaves regarding the quality requirements of traditional internal medicine Kemenkes RI No. 55/Menkes/SK/I/2000 and regulations BPOM RI Nomor 12 Tahun 2014. This is because the samples tested were fresh samples, while red betel leaves as a requirement for traditional medicine were in the form of simplicia or dry powder.

Total Dissolved Solids

The total value of dissolved solids or commonly known as brix levels is obtained from measurements using a refractometer. The total dissolved solids indicates the content of the

ingredients dissolved in the solution (Yulianti dkk., 2014). The results of the variance of total dissolved solids showed that the difference in harvesting time was significantly different for red betel leaves. The average value of total dissolved solids based on the difference in harvesting time can be seen in Table 4.

Table 4. Mean of Total Dissolved Solids Based on the Difference of Harvesting Time

| Treatment | TPT (°Brix) ± Stdev |
|----------------|--------------------------|
| W ₁ | 1.32 ± 0.01 ^c |
| W ₂ | 1.33 ± 0.01 ^b |
| W ₃ | 1.34 ± 0.01 ^a |

Note: Different superscripts in the same column show significant differences (P <0.01).

The results of Duncan's Multiple Range Test (DMRT) show that the highest total dissolved solids content in red betel leaves is harvest time at 15.00 WIB (W3) with an average value of 1.34 °brix. The lowest level of total dissolved solids was harvesting in the morning at 07.00 WIB (W1) with an average value of 1.32 °brix

The increase in total dissolved solids is due to the fact that complex components such as carbohydrates and proteins are broken down into simpler compounds resulting in an increase in total dissolved solids. The components measured as total dissolved solids are sucrose, reducing sugars, organic acids and proteins (Yulianti et al., 2014).

The level of total dissolved solids in plants is influenced by light intensity where in conditions of high light intensity photosynthesis becomes effective. and at high light intensity is usually positively correlated with temperature, and temperature will affect the work of enzymes, plants effectively carry out photosynthesis between temperatures of 30-40 °C. Photosynthesis that takes place in the afternoon and evening accumulates into the leaves and in the afternoon is the time limit for harvesting, after that time the total dissolved solids content will decrease because there will be a change of sugar into flour. (Surtinah, 2012).

Color

Analysis of color content using a UV-Vis spectrophotometer where the data obtained is an absorbance value. Absorbance is the ratio of the intensity of the absorbed light to the intensity of the incident light. This absorbance value will

depend on the content of the substance contained in it, the more substances contained in a sample, the more molecules will absorb light at a certain wavelength so that the absorbance value is greater or in other words the absorbance value will be directly proportional to the concentration. substances contained in a sample (Neldawati dkk., 2013).

The results of the color variable variance showed that the difference in harvesting time was significantly different in the color content of red betel leaves. The average value of red betel leaf color content based on the difference in harvesting time can be seen in Table 5.

Table 5. Color Average Based on Harvesting Time Differences

| Treatment | Color (%) ± Stdev |
|----------------|-------------------------|
| W ₁ | 80 ± 0.59 ^c |
| W ₂ | 148 ± 0.00 ^b |
| W ₃ | 153 ± 0.45 ^a |

Note: Different superscripts in the same column show significant differences (P <0.01).

The results of Duncan's Multiple Range Test (DMRT) show that the highest color content is at the afternoon harvest time at 15.00 WIB (W3) with an average value of 153%. The color content with the lowest value is harvesting time in the morning at 07.00 WIB (W1) with an average value of 80%.

The more colorful it is, the higher the color content in a leaf. This happens because the higher the color content, the more molecules contained in the plant leaf extract will increase the number of molecules that will absorb light at a certain wavelength. (Neldawati dkk., 2013).

Optimal time The levels of color and other compounds in the plant vary between parts, tissue and plant age, and are influenced by environmental factors. Environmental factors consist of temperature, ultraviolet light, nutrients, water availability and CO₂ levels in the atmosphere (Carbonaro & Grant, 2005). Color levels continue to increase until harvest time in the afternoon, this is because the need for light for the photosynthesis process is fulfilled. High light intensity photosynthesis becomes effective (Surtinah, 2012). According to the Minister of Health of the Republic of Indonesia No.003/MENKES/PER/2012 regarding food additives, states that the maximum level of use of

color content as a natural dye is 171%.

CONCLUSION

The best harvest time is in the afternoon at 15.00 WIB (W3) against alkaloids (0.98 mg / g), analysis of total anthocyanins (8.50 mg / g), water content (14.4%), total dissolved solids (1.34 g) and color (153%).

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