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Effect of Processing Methods on the Chemical Properties of Protium javanicum Burm F. Herbal Tea Powder

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Abstract
Protium javanicum herbal tea powder was a type of herbal tea that was produced from *Protium javanicum* leaves through withering and drying processes. One of the factors that influence the chemical composition of this herbal tea was the processing method. The purpose of this study was to determine the effect of processing methods on the chemical characteristics of *Protium javanicum* herbal tea powder. The processing methods applied in this research were the withering method (PY), the direct drying method (DM) and the steaming method (ST). Results showed that differences in processing methods significantly affected water content, vitamin C, total phenols, flavonoids, tannin levels and antioxidant activity of *Protium javanicum* herbal tea powder. Processing with the ST method produced herbal tea powder with the best chemical characteristics, including water content of 8.0379%, ash content of 6.1673%, vitamin C of 66.6674%, total phenol of 9.1611%, total flavonoids 1.7188% , tannins content 10.7536% and antioxidant activity 92.1944%.
Keywords: *Protium javanicum*, herbal, tea, powder, processing method
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1. Introduction

Protium javanicum Burm F is a plant of the genus Protium which has a spiny stem morphology and a tapered leaf shape at the tip, not too wide in size and pink on very young leaves and dark green on old leaves. In Bali, *Protium javanicum* is used as traditional medicine to cure coughs, treat diarrhea and also often used as vegetables because they have a distinctive flavor and taste.

The peculiarity of the *Protium javanicum* leaf is that it has a slightly harsh taste and smells sour. Based on the phytochemical content, it is known that the chemical composition of the *Protium javanicum* includes : essential oils, triterpenoids, steroids, flavonoids, quinones and tannins. Components of volatile compounds in *Protium javanicum* such as: monoterpenes such as β-ocimen, α-pinen and sesquiterpenes such as β-cariophylene, germacren, α-humulen, β-elements, cariophylene oxide, α-amorpen, and spatulenol (Sukmajaya *et al*, 2012). Sanjaya (2002) also reported that *Protium javanicum* leaves contain essential oils from tinamil tiglat and myristophenone compounds which have antibacterial activity. Therefore, many people use it for traditional medicine.

The utilization of the bioactive components of *Protium javanicum* leaves can be done by processing the leaves into functional food products, including herbal tea. Herbal tea is a general term used for tea beverage products that are not derived from the *Camelia sinensis* plant. Herbal teas can be made from flowers, leaves, seeds or stems of herbal plants (Ravikumar, 2014). Consuming herbal teas is believed to provide various benefits including enhancing the immune system, relaxation, therapeutic effects and others.

Tea is usually available in leaf, packaged and powder form, but powdered processing is much more beneficial because it has a higher nutritional value and all of the nutritional components in tea can be consumed. Fujioka *et al*., (2016) reported that green tea in powder form (*matcha*) had higher scavenging effect on reactive oxygen compounds when compared to leaf tea.

One of the factors that play a role in determining the chemical characteristics of herbal teas is the processing method. In general, herbal tea processing refers to processing tea without enzymatic oxidation. There are several types of processing methods without enzymatic oxidation that are commonly used, namely by steaming and drying, direct drying and by withering at room temperature then drying. The steaming method is a processing method using a steaming technique that aims to inactivate the polyphenol oxidase enzyme (Kosinska and Andlauer, 2014). The direct drying method is the processing of tea without going through rolling or crushing but with rapid deformation and air drying to preserve most of the polyphenol compounds (Saha *et al*., 2017). The withering and drying method is a tea processing technique that is carried out by withering at room temperature and then drying it with a certain temperature and time (Damayanthi *et al*., 2008). Differences in the processing of herbal tea will affect the chemical profile of the products.

Felicia *et al*., (2016) reported that herbal tea powder from avocado leaves processed by the steaming method had higher levels of phenols, flavonoids and antioxidants compared to the roasting method. Other researchers (Laelasari *et al*., 2016) reported that the antioxidant activity of herbal tea powder from mulberry leaves by Indonesian processing method (short withering at room temperature and drying) had higher antioxidant activity compared to Thai processing methods (steaming and roasting). So far there has been no research

conducted to compare the processing method to the chemical characteristics of *Protium javanicum* herbal tea powder. This study aims to determine the effect of the processing method on the chemical characteristics of *Protium javanicum* herbal tea powder.

2. Materials and Methods

2.1. Research methods.

The main ingredient used in this study, namely young *Protium javanicum* leaves, was obtained from around the Jimbaran Hill area. Processing of herbal tea powder was carried out using three methods, namely steaming methods (ST) according to Topuz *et al*., (2014) with modification, withering method (PY) (Damayanthi *et al*., 2008) with modification and direct drying method (DM) (Rohdiana, 2007) with modifications.

2.1.1. Withering Method (PY)

A total of 100 grams of *Protium javanicum* leaves were sorted and withered for 24 hours by aerating in an open space, then dried in an oven at 50°C for 4 hours. Dried leaves were reduced in size and sieved with a 100 mesh sieve.

2.1.2. Direct Drying Method (DM)

A total of 100 grams of *Protium javanicum* leaves were sorted and withered for 5 minutes at room temperature. Leaves were dried in an oven at 50ºC for 4 hours. Dried leaves were cooled for 5 minutes, then crushed and sieved with a 100 mesh sieve

2.1.3.. Steaming methods (ST)

A total of 100 grams of leaves *Protium javanicum*, which have been separated from the stems, was steamed at 100°C for 90 seconds then cooled for 5 minutes. Leaves were dried in an oven at 50°C for 4 hours. Dried leaves were ground to become a powder and then sieved with a 100 mesh sieve.

2.2. Experimental design

This study used a completely randomized design with 3 treatments methods of processing : withering method (PY), direct drying method (DM) and steaming method (ST) . Each treatment was repeated 5 times, in order to obtain 15 experimental units. Data were analyzed using variance, and continued with Duncan's Multiple Range Test (DMRT) according to Gomes and Gomes (1995).

2.3. Observed parameters

The parameters observed in this study were moisture content using the gravimetric method ((AOAC, 2005), ash content using the dry ashing method (AOAC, 2005), determination of vitamin C levels with the iodine titration method (Sudarmadji *et a*l., l98l) total phenol using the Folin-Ciocalteu method (Sakanaka *et al*., 2003), total flavonoids with spectrophotometric method (Xu dan Chang (2007), tannin content according to Suhardi (1997), antioxidant activity using the DPPH (2,2-diphenyl-l-picrylhidrazyl) method (Khan *et al*., 2012).

3. Results and Discussion

3.1. Moisture content

The average value of moisture, ash and vitamin C content of *Protium javanicum* herbal tea powder were presented in Table 1.

Table 1. Moisture, ash and vitamin C content of *Protium javanicum* herbal tea powder.

Treatment	Moisture content (%)	Ash content (%)	Vitamin C (%)
PY method	9,1748 ± 0,05b	6,127 ± 0,007	50,7830 ± 1,64a
DM method	10,1413 ± 0,01c	6,0933 ± 0,071	54,1646 ± 1,49b
ST method	8,0379 ± 0,006a	6,1673 ± 0,005	66,6679 ± 0,94c

Note: Data are expressed as means ± SD. Mean values in a column with different letters are significantly different at $p \leq 0.05$

Based on Table 1, it showed that the differences in processing methods had a significant effect ($P < 0.05$) on the moisture content of *Proteum javanicum* herbal tea powder. The highest water content was obtained through DM method while the smallest water content was obtained through ST method. The DM method produces herbal tea powder with a moisture content of 10.1413%, possibly because the processing method did not involve the withering stage, so that the water content of fresh ingredients was still high, and when dried with a certain temperature and time, only part of the water could be evaporated.

The PY method resulted in a product with a moisture content of 9.1748% and lower than the product processed by the DM method. This proved that the withering process at room temperature could reduce the moisture content of fresh ingredients. Arpah (1993) reported that the withering function was to reduce water content by 55% - 70%. This occurs due to the evaporation process by both the air flow and the exhaled heat.

The water content of herbal tea powder processed by the ST method was the lowest compared to other methods, namely 8.0379%. The decrease in water content probably influenced by the heat energy from the

steamer, causing the mass transfer process that occurs in the leaf system to the leaf surface then to the leaf environment (Amanto *et al*., 2015). The higher the temperature and the longer the steaming process, the greater the heat energy carried by the air, so that the amount of mass of liquid evaporated from the material increases. The heat energy received will change the water content in the material into steam, so that water vapor will move to the surface of the material and then released into the environment of the material (Holman, 1995). In addition, it is suspected that the decrease in water content is also closely related to the role of water which affects the rate of reaction that occurs during the blanching process (Ayu and Yuwono, 2013).

3.2. Ash Content

Based on the data in Table 1, it has been shown that differences in processing methods had no effect on the ash content of the *Proteum javanicum* herbal tea powder. Ash content indicates the level of inorganic components present in a material. Minerals in plants are found in plant cell walls. Processing treatment can damage plant cell walls and have an impact on decreasing mineral levels (Bamidele *et al*., 2017). However, the results of this study indicated that the different processing methods did not cause differences in the ash content of the product. This is probably due to the processing did not cause damage to the inorganic components so that the dissolved solids did not get out of the leaf cells. Thus, between treatments there was no significant difference in ash content. The results of this study were in line with those reported by Sayuti (2010) that the withering treatment and drying of mulberry leaf tea did not cause significant changes in ash content. Liliana (2005) also reported that withering and drying treatments did not affect the ash content of celery leaf tea.

3.3. Vitamin C Content

Analysis of variance showed that the difference in processing methods had a significant effect ($P < 0.05$) on the vitamin C levels of *Proteum javanicum* herbal tea powder. Herbal tea processed by the ST method produced the highest levels of vitamin C, while processing with the PY method produced the lowest levels of vitamin C. Based on the data listed in Table 1, the processing of herbal tea using the ST method is better at maintaining vitamin C levels in the product compared to the processing of the PY and the DM methods. The use of the correct temperature and time in the steaming process was necessary to maintain the product's vitamin C levels. The time and temperature required may vary greatly depending on the type and size of the material. Underblancing conditions can stimulate enzyme activity, while overblancing conditions can cause changes in flavor, color and loss of vitamins and minerals.

Fafunso *et al*., (1976) reported that the loss of vitamin C in various types of vegetables caused by the blancing process, however, the blancing process carried out at the right temperature and time did not cause significant loss of vitamin C. The study by Musa *et al* (2017) also reported that the rate of damage to vitamin C in *Moringa oleifera*, *Hibiscus sabdarifa* and *Hibiscus esculentus* vegetables decreased due to the blancing process. Meanwhile, Kaur *et al* (2018) reported that the steam blancing was better than water blancing and resulted in higher vitamin C levels (289 mg / 100g) in broccoli. The steam blanching was also reported to have no significant effect on vitamin C levels in *Moringa* herbal tea (Wickramasinghe *et al*., 2020).

Processing of *Proteum javanicum* herbal tea using the DM method produces herbal teas with lower vitamin C than the ST method but the vitamin C content is higher than the PY method. Withering, which is carried out by spreading the material at room temperature and being exposed to the surrounding oxygen for a long period of time, makes vitamin C more susceptible to damage. The drying process, heating for a long time, heating in iron or copper tools and exposure to environmental temperatures cause damage to vitamin C (Fafunso *et al*., 1976; Vishwanathan *et al*. (2013).

3.4. Total Phenol

The average value of total phenol, flavonoid, tannin and antioxidant activity of *Protium javanicum* herbal tea powder presented in Table 2.

Table 2. Phenol, flavonoid, tannin and antioxidant activity of *Protium javanicum* herbal tea powder.

Treatment	Phenol (%)	Flavonoid (%)	Tannin (%)	Antioxidant activity (%)
PY method	6,5654 ± 0,01a	1,2816 ± 0,004a	7,1667 ± 0,003a	69,8085 ± 0,09a
DM method	8,9301 ± 0,04b	1,6552 ± 0,001b	9,3237 ± 0,004b	84,5361 ± 0,20b
ST method	9,1611 ± 0,0007c	1,7188 ± 0,003c	10,7536 ± 0,003c	92,1944 ± 0,10c

Note: Data are expressed as means ± SD. Mean values in a column with different letters are significantly different at $p \leq 0.05$

Analysis of variance showed that the difference in processing methods had a significant effect ($P < 0.05$) on the total phenol of *Proteum javanicum* herbal tea powder (Table 2). Herbal tea processed by the ST method have the highest total phenol, namely 9, 1611%, while the lowest total phenol was obtained from herbal tea processed by the PY method (6.5654%). These results have shown that the ST method was able to recover polyphenol

compounds in *Preoteum javanicum* leaves. The increase in total phenolic content due to the steam blanching treatment was most likely caused by the presence of polyphenol compounds did not undergo enzymatic oxidation (Nurhayati *et al* 2018). Decreased degradation of polyphenol compounds as a result of the inactivation of the polyphenol oxidase enzyme (Bamidele *et al*., 2017) and be due to the release of bound phenolic acids from the breakdown of cellular constituents of the plant cell walls (Francisco *et al*., 2010).

The increase in total phenol content is probably due to the degradation of tannins into simpler phenol compounds. Kim *et al*., (2010) reported that tannic acid will undergo hydrolysis to become galloyl such as gallothanin in the presence of heat. Turkmen *et al*., (2005) also reported that there was an increase in total phenol content in green beans and chilies that were boiled for 5 minutes. The same thing was also reported by Roy *et al*., (2009) that broccoli blanched for 5 minutes can increase the total phenol level from 135.66 mg EAG / 100g to 144.33 mg EAG / 100g.

Proteum javanicum herbal tea powder processed by the DM and PY methods produced lower total phenol compared to the ST method. Processing with the DM method involves a shorter withering process compared to the PY method. Withering for a long time will cause damage to phenolic compounds.

3.5. Flavonoid

Analysis of variance showed that the difference in processing methods had a significant effect (P <0.05) on the total flavonoids of *Proteum javanicum* herbal tea (Table 2). The processing of herbal tea using the ST method produced the highest levels of flavonoids, while the processing with the PY method produced the lowest levels of flavonoids. Steaming is one way of wet heating. It was reported that wet heating treatment for one minute did not affect the phytochemical compounds and the integrity of the cell structure which would result in component migration (Zainol *et al*., 2009). The results of this study are in line with those reported by Pujimulyani *et al*., (2010) that white turmeric blanched for five minutes in 0.05% citric acid medium has higher levels of total flavonoids compared to fresh ingredients. Likewise, reported by Roy (2007) that heating the *Cosmos caudatus* leaves for one minute did not cause changes in flavonoid levels.

Processing of *Proteum javanicum* herbal tea powder using the DM and PY methods resulted in lower levels of total flavonoids than the ST method. The withering process can increase or decrease certain components (Arpah 1993). The withering process for 24 hours is thought to cause oxidation of *Proteum javanicum* leaves.

3.6. Tannin Content

The difference in processing methods showed a significant effect (P <0.05) on the tannin content of *Proteum javanicum* herbal tea powder. Herbal tea processed using the ST method had the highest levels of tannins, while herbal tea processed using the PY method produced the lowest levels of tannins. This was presumably because steaming causes protein denaturation so that the tannins that are with the protein become free so that the condensed tannins are more easily extracted. In addition, it is also possible that blancing/steaming will deactivate the polyphenoloxidase enzyme so that tannins do not undergo enzymatic oxidation. Barret and Theerakulkalit (1995) reported that one of the goals of blancing is to deactivate the enzyme. Similar results were also reported by Pujimulyani *et al*., (2010) that blanching white turmeric in citric acid medium 0.05%, temperature 100 ºC for 5 minutes can significantly increase the condensed tannins from 6.10 to 10.59 mg EC/g compared to those without blanching.

Processing with the DM method produced lower levels of tannins than the ST method, but higher levels than the PY method. This was probably because the enzymatic oxidation was more inhibited if the withering process is brief. Withering for a long time will cause enzymatic oxidation, changing some of the tannins into derivative compounds (theaflavins and thearubigins) so that their levels were reduced (Rohdiana, 2007). According to Rollyanroza (1990) *in* Sayuti (2010) that during withering, the tea shoots will undergo compound changes due to plant metabolic processes that take place in leaf cells. Therefore, at the time of withering, an enzymatic oxidation reaction has occurred, resulting in a change in the content of the compounds in it. The results of this study are in line with those reported by Kusumaningrum *et al*., (2013) that the different treatment of enzymatic oxidation processes and differences in withering time had a significant effect on the tannin content of *Lotus* flower tea. The longer the withering time, the lower the tannin content of the *Lotus* flower tea.

3.7. Antioxidant Activity

Based on the data in Table 1, it is known that differences in processing methods had a significant effect on the antioxidant activity of *Proteum javanicum* herbal powder tea (P <0.05). The highest antioxidant activity was obtained from processing with the ST method, namely 92.1944%, while the lowest antioxidant activity was obtained from processing with the PY method, namely 69.8085%. Antioxidants are electron-giving compounds or compounds that can counteract or reduce the negative impact of oxidants in the body. Antioxidants work by donating one electron to an oxidant compound so that the activity of the oxidant compound can be inhibited (Winarsi, 2007).

The results showed that the processing of *Proteum javanicum* herbal tea using the ST method was better to maintain the content of antioxidant compounds. The high antioxidant activity in herbal teas was supported by the levels of phenols, flavonoids, tannins and vitamin C which was also the highest among the three processing methods tested. This indicates that there are polyphenol molecules that act as H + donors so that they play the most role in antioxidant activity. So the more total polyphenol content, the more OH groups donate H + atoms so that the antioxidant activity can be higher (Supriyanto *et al*., 2014). Volioglu *et al*., (1998) also reported that phenol content and antioxidant activity had a strong relationship and were very significant factors in increasing and decreasing antioxidant activity. Several studies have shown that steaming methods increased antioxidant activity. Puuponen (2003) reported that the antioxidant activity of steamed cabbage increased by 9%.

4. Conclusion

The differences in processing methods had a significant effect on moisture content, vitamin C, total phenols, flavonoids, tannin levels and antioxidant activity of *Protium javanicum* herbal tea powder. *Protium javanicum* leaves processed by the ST method produced herbal tea with the best chemical characteristics indicated by the highest content of phytochemical compounds compared to other methods.

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