

Effect of Drying Methods and Harvest Age on the Quality of Sliced Tiwai Onion (*Eleutherine americana* Merr) as A Potential Functional Food

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Abstract

Tiwai onion (*Eleutherine americana* L. Merr) is renowned for therapeutic properties due to the bioactive constituents, including polyphenol, flavonoid, and tannin. Therefore, this study aimed to characterize water content, ash, fat, protein, polyphenol, flavonoid, tannin, and antioxidant capacity, as well as to observe the functional groups and morphology of dried tiwai powder. The methodology comprised 2 factors, namely harvest age and drying methods. The results showed significant effects of harvest age and drying methods on water, fat, total phenol, total flavonoid, anthocyanin content, and antioxidant capacity, while ash, protein, and tannin content were not significantly affected. In Fourier Transform Infrared Spectroscopy (FTIR) analysis, changes in functional groups were observed, although not significant in wave numbers range of 1,800 - 1,350 and 1,200 - 950 cm^{-1} . Specifically, an increase was observed in signals from C-C, C-O, and C-O-C vibrations. Scanning Electron Microscopy (SEM) at 1000x magnification showed that air drying at 5 months of harvest age produced dense granules, while electric oven and microwave drying of older harvests led to a flatter structure and crumbly texture.

Keywords: Tiwai powder, Proximate, Polyphenol, Functional groups, Phytochemistry, Morphological, Antioxidant capacity

Introduction

Tiwai onion (*Eleutherine americana* Merr) is renowned for a long history as a traditional medicine in various regions in Indonesia. The diversity of active compounds, including polyphenol, flavonoid, and tannin [1,2], provides an interesting scientific basis for exploring the potential as a traditional medicinal plant. According to several previous studies, tiwai has antioxidant properties, protecting body cells from damage caused by free radicals, helping prevent premature aging, supporting cell health, and preventing various diseases [1,3,4].

In practical application, tiwai is usually consumed in dry form as an ingredient in herbal drinks or mixtures in food products [5]. During selection, attention should be paid to characteristics such as dry skin, stiffness, round and symmetrical shape, and color without any spots or damage [6]. These characteristics

can be observed based on different harvest ages, playing a crucial role in determining pharmacological activity which potentially influences chemical properties and bioactive compounds both qualitatively and quantitatively [7,8]. Another study by [9], reported that tiwai harvested at the age of 2 months had a bioactive phenol content of 76.596 ± 0.057 mg/g GAE, higher than those harvested at the age of 1 and 3 months.

Three methods are often used for drying tiwai, namely air, electric oven, and microwave, which affect the physical properties, active compounds, and antioxidant activity [10]. Therefore, this study aimed to characterize the bioactive compounds containing water content, ash, fat, protein, polyphenol, flavonoid, tannin, and antioxidant capacity of dried tiwai using 3 drying methods. In another study [11] it was shown that *B. vulgaris* bamboo leaves using the microwave drying technique had a total active compound content of phenols and flavonoids ranging from 2.69 ± 0.01 - 12.59 ± 0.09 mg gallic acid equivalent (GAE)/ g and 0.77 ± 0.01 - 2.12 ± 0.01 mg quercetin equivalent (QE)/g. Antioxidant activity ranges between 2.92 ± 0.01 - 4.73 ± 0.02 $\mu\text{g/mL}$, superior in retaining active compounds compared to using sun, shade, freeze drying and oven drying techniques.

Materials and methods

Tiwai (*Eleutherine americana* Merr), with the harvest age of 5, 7 and 9 months, were purchased from farmers in Samarinda, Indonesia. The following chemicals were used in the experiment aquadest (Faperta - Unmul, Indonesia), methanol (Fulltime, USA), NaOH (Merck, USA), tannic acid standard (Sigma-Aldrich, USA), gallic acid standard (Sigma-Aldrich, USA), NaNO₂ (Sigma-Aldrich, USA), AlCl₃ (Sigma-Aldrich, USA), and quercetin standard (Sigma-Aldrich, USA). Meanwhile, the instruments used were analytical balance (ACIS, China), blender (Miyako), ultra sonicator (Delta D68H, China), centrifuge (Dragon-type LC-045, China), hot plate (Faithful, type SH-24, China), UV-VIS Spectrophotometer (Eppendorf, Germany), and Vortex (Faithful, China).

Tiwai preparation

Tiwai was sorted and washed under running water and grouped based on harvest age of 5, 7 and 9 months. Subsequently, the samples were sliced using slicing (Tupperware) with a thickness of 1 mm maintained consistently and continued with drying using 3 methods, namely air drying (48 h), electric oven (24 h), and microwave (10 min) with temperatures of 25, 50, and 100 °C respectively. Dried tiwai was grounded using a blender and then sieved with 80 mesh.

Extraction tiwai powder

Extraction process was carried out by weighing 10 g of tiwai powder, and dissolving in a 1:10 ratio of 96 % ethanol solvent, and then left to stand at 25 and 28 °C for 24 h. After filtering using Whatman filter paper, the filtrate was heated to 50 °C in a rotary evaporator (IKA RV10), and evaporation process continued until the sample achieved the consistency of a paste. The amounts of phenol, flavonoid, tannin, and antioxidant capacity were measured in extracted pasta samples examination.

Water, ash, fat, and protein contents analysis

Water, ash, fat, and protein contents were analyzed using standard AOAC methods [12].

Phenol content analysis

Ten milliliters of ethanol were used to dissolve 0.3 g of extract. Next, 0.2 mL of the extract solution was pipetted and mixed with 1 mL of 50 % (v/v) Folin-Ciocalteu reagent and 15.8 mL of distilled water.

After 8 min, 3 mL of 5 % Na₂CO₃ (w/v) was added, and the mixture was allowed to sit/stand in the dark at room temperature (28 ± 2 °C) for 2 h. After that, a wavelength of 725 nm was used to measure the absorbance value. The same method was used to generate a gallic acid standard curve, which was plotted against the absorbance. Gallic acid equivalent dry weight (mg/g) is the unit of measurement for total phenols [13].

Flavonoid content analysis

Extract (1 mg) was dissolved in 10 mL of 95 % ethanol. After the extract had been dissolved in ethanol, 0.7 mL of distilled water and 0.1 mL of 5 % NaNO₂ were combined. 0.1 mL of AlCl₃ 10 % was added after 5 min, followed by 0.5 mL of 1 M NaOH after 6 min. After thoroughly combining all the components, they were incubated for 10 min. At 510 nm in wavelength, absorbance was measured using a blank consisting of 1 mL of sample substituted with 1 mL of 95 % ethanol solvent. We quantified the total flavonoids as mg of catechin equivalents per g of dry weight [14].

Tannin content analysis

The total tannin content of the extract was determined using the Folin-Chiocalteu technique. After dissolving around 0.1 g of Tiwai extract in 7.5 mL of distilled water, the liquid was mixed with 0.5 mL of Folin's reagent. After that, 10 mL of distilled water was added to 1 mL of 35 % Na₂CO₃. After giving the mixture a good shake, 30 min were kept at room temperature. Gallic acid reference standard solutions at the following concentrations (20, 40, 60, 80 and 100 µg/mL) were produced. Using UV/Vis spectrophotometry, the absorbance of test solutions and standards was measured at 725 nm about (with) a blank. The units of measurement for tannin content are mg GAE/g extract [15].

Antioxidant capacity

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) technique was used to determine antioxidant activity utilizing the antioxidant activity test method [16]. One milliliter of the material was placed in a test tube along with 7 milliliters of ethanol. The combination was then treated with 2 mL of DPPH, bringing the final DPPH concentration down to 0.2 mM. Following a 2-to 5-minutes vortex, the material was allowed to sit at room temperature (28 ± 2 °C) for 30 min. Following that, a wavelength of 517 nm was used to measure the absorbance. Antioxidant activity is measured as a percentage of antioxidant capacity and is represented as a percentage of DPPH radical scavenging activity.

$$\% \text{ Antioxidant Capacity} = \frac{(\text{Control absorbance} - \text{Sample absorbance})}{\text{Control absorbance}} \times 100 \quad (1)$$

ATR FTIR analysis

Three harvest ages (5, 7 and 9 months) of tiwai onion powder were subjected to FTIR spectra analysis. Attenuated total reflectance (ATR) using ZnSe crystals and a deuterated triglycine sulfate detector were used for the analysis, which involved 32 iterations and a modest number of samples in the range of 650 - 4,000 cm⁻¹ with a resolution of 4 cm⁻¹ [17].

Morphological structure

SEM analysis was carried out with SEM-EDD/X (JSM-6510 LA, JEOL, Japan). The sample was placed on the stubs and then coated with gold using a gold sputter coater for 10 min. The coated sample was placed into an SEM microscope and then observed at a magnification of up to 1,000X [18].

Results and discussion

Water content

The primary determinant of adequate drying procedure for a functional food raw material is water content, which typically ranges from 4.5 to 10 % (w/w) [19,20]. Based on the results, the 3 drying methods yielded water content values ranging from 4.33 ± 0.23 to 10.29 ± 0.14 % (w/w). Air drying method produced a higher water content compared to electric oven and microwave at all harvest ages, this can be caused by differences in drying rate, temperature, heat distribution and water evaporation mechanisms. So that it has an impact on the preparation of functional raw materials, the drying method needs to be selected taking into account the balance between quality retention and process efficiency [21].

The lowest water content value was found in electric oven drying at a temperature of 50 °C for 24 h at harvest age of 7 months. The use of microwave drying also produced lower water content values compared to air drying. Previous study indicates that despite the considerable fluctuation in water content, air drying is still a common practice [22]. On the other hand, mechanical dryers offer greater functional qualities and a more favorable drying impact in terms of water content [23]. So it can be concluded that air drying may be suitable for products that are very sensitive to heat, while electric oven or microwave drying may be better for applications that require high efficiency and low moisture content.

Ash content

Irrespective of drying process used, ash content of dried items will inevitably experience alteration because ash is a crude measure of minerals [24]. **Table 1** illustrates this condition, showing no significance for each method and harvest age. The lowest level of 2.01 ± 0.19 % was shown at harvest age of 5 months with air drying and the highest level of 2.53 ± 0.13 % was found at harvest age of 9 months with microwave. In general, microwave drying provides an advantage in maintaining the mineral content of tiwai because the procedure is carried out with short heating such as calcium, phosphorus, magnesium, potassium, sodium, iron and copper. Although other methods are also relatively effective in maintaining the mineral content, only air drying requires quite a long time, only air drying requires a long time. long enough to result in the loss of minerals that are susceptible to oxidation such as iron and copper [25]. With a few exceptions related to pre-drying treatment conditions, ash content is unaffected by drying methods. For instance, mineral removal from the dried material can be achieved by using acid blanching before drying [26].

Fat content

According to previous reports, different drying methods can affect how plant lipids are preserved or changed, factors such as drying methods, temperature, and length of time play a role in determining the final fat composition of plants [27]. In this study, drying methods and harvest age of tiwai had a significant effect on the final fat content obtained, namely in the range of 0.75 ± 0.25 - 6.43 ± 1.40 %. The highest fat content of 6.43 ± 1.40 % was found at 9 months of harvest age using electric oven drying, while the lowest of 0.75 ± 0.25 was found in air drying at 7 months. This result correlates with differences in harvest age as plants tend to accumulate more lipids with age the correlation is influenced by several factors such as plant development stage, metabolic changes in the form of increased fatty acids, Triacylglycerol (TAG) Synthesis, increased levels of abscisic acid (ABA), decrease in gibberellin levels and fat synthesis during the ripening process [28]. Numerous factors, including the stage of plant growth, changes in metabolism, and lipid production in the ripening process, impact the correlation. The choice of drying method also significantly influences fat content, for instance, the use of electric oven entails maintaining a certain

temperature which reduces water content without causing lipid degradation due to excessive heat, reducing the risk of thermal degradation, as well as maintaining the integrity of TAG [28].

Meanwhile, air drying causes a decrease in fat content due to air and humidity exposure, decreases gradually over a long time, thereby increasing the risk of fatty acid oxidation, TAG hydrolysis and degradation of the hormone ABA, while fast drying microwave drying helps reduce the risk of fatty acid oxidation, maintaining the structure and integrity of TAG. and minimizing thermal and oxidative degradation [28,29]. Another study by Uribe *et al.* [30] reported a significant effect of different drying methods on lipid content in green algae.

Protein content

The use of different drying methods can affect the structural integrity and biochemical composition of protein [31]. Among all treatments, both harvest age and drying methods showed no significant influence. Protein content values ranged from 4.49 ± 0.27 - 6.71 ± 0.27 %, with the highest occurring in 7 months of harvest age treatment using air drying method, while the lowest was in microwave treatment at 9 months (**Table 1**). The Kjeldahl method, based solely on nitrogen content, was used to determine protein content, thereby limiting the observation of other protein components [32]. More specific protein analysis techniques, such as SDS-PAGE and HPLC can be carried out to determine changes in protein integration, protein denaturation and aggregation, changes in protein conformation, protein profiles, and even protein composition [33]. The preservation of protein content is crucial for maintaining flavor in food items. In this study, several drying methods proved to be very efficacious in preserving protein content, as evidenced by the same outcomes obtained [34].

Table 1 Proximate contents of dried tiwai with different harvest ages and drying methods including air drying, electric oven, and microwave.

Sample	Drying method	Moisture content	Ash content	Fat content	Protein content
		(%w/w)	(%w/w)	(%w/w)	(%w/w)
5 months	Air drying	10.29 ± 0.14	2.01 ± 0.19	2.05 ± 0.05	5.72 ± 0.61
	Electric oven	4.73 ± 0.22	2.18 ± 0.05	1.74 ± 0.34	5.60 ± 0.30
	Microwave	6.27 ± 0.14	2.31 ± 0.03	4.49 ± 0.71	5.90 ± 0.53
7 months	Air drying	9.19 ± 0.14	2.14 ± 0.05	0.75 ± 0.25	6.71 ± 0.27
	Electric oven	4.33 ± 0.23	2.35 ± 0.11	5.23 ± 0.75	4.96 ± 0.27
	Microwave	4.86 ± 0.13	2.46 ± 0.02	6.42 ± 0.11	5.19 ± 0.79
9 months	Air drying	5.73 ± 0.86	2.18 ± 0.08	4.49 ± 0.51	5.25 ± 0.46
	Electric oven	5.05 ± 0.05	2.50 ± 0.01	6.43 ± 1.40	5.66 ± 0.66
	Microwave	4.99 ± 0.09	2.53 ± 0.13	5.74 ± 0.36	4.49 ± 0.27

Total phenol content

The total phenol content of tiwai at different harvest ages and air drying, electric oven, and microwave drying methods were determined using spectrophotometric tests. The Folin-Ciocalteu method, based on electron transfer in a basic solution of the complex phenol compound phosphomolybdic/phosphotungstic acid, was used [35]. The results (**Table 2**) showed that harvest ages of 5 and 9 months with air drying

produced the lowest total phenols of 130.26 ± 18.49 and 117.70 ± 13.41 mg GAE/g, respectively. Meanwhile, microwave drying method at the age of 5 and 7 months produced the highest total phenols of 202.69 ± 8.16 and 211.67 ± 22.30 mg GAE/g, respectively, compared to electric oven and air drying. Phenol content tends to decrease in tiwai harvested after 9 months with each drying method due to the effects of enzymatic processes. The phenolic content tends to decrease in tiwai harvested after 9 months with each drying method. This decrease can be attributed to the enzymatic process, namely the degradation of polyphenol oxidase (PPO) and peroxidase (POD) enzymes. These enzymes catalyze the oxidation of phenolic compounds, leading to their breakdown and the formation of quinones, which can then polymerize to form brown pigments. In microwave drying the rapid drying process, is very effective in preserving the phenolic content by quickly denaturing the enzymes responsible for degradation. Oven drying is quite effective if the temperature and drying time can be optimized, while slow air drying inactivates enzymes and allows oxidative reactions to occur, thus affecting phenol levels [36].

As tiwai ages, enzymatic activity increases, thereby causing the degradation of phenol compounds. These results are consistent with another study by [37], showing the degradation of phenol compounds due to enzymatic processes in coffee plants.

Total flavonoid

The Two-Way ANOVA test showed a significant effect of harvest age and different drying methods on tiwai. Harvest age of 9 months produced the highest total flavonoid in each drying method compared to harvest ages of 5 and 7 months. Total flavonoid ranged from 11.58 ± 1.89 - 34.24 ± 3.46 mg TAE/g, with air drying method at 9 months of harvest producing the highest, while the lowest was produced with microwave drying at 7 months. Flavonoid is synthesized through the phenylpropanoid and flavonoid biosynthetic pathways. According to [38], the expression of genes coding for enzymes in this pathway, such as chalcone synthase (CHS) and flavonol synthase (FLS), can be influenced by drying conditions and plant age, affecting flavonoid production.

Tannin content

Total tannin in this study was analyzed by Two-Way ANOVA, showing an insignificant influence between harvest ages and drying methods. Microwave drying at 9 months of harvest had the highest total tannin of $2,168.00 \pm 3.93$ and the lowest of $1,806.52 \pm 0.79$ was produced by air drying at 5 months. Some tannins can be stable under various environmental conditions, and the concentrations may not change significantly with drying methods or harvest age. The choice of method used for tannin analysis can influence the results of observations. According to [39], certain methods are more sensitive to changes in tannin content.

Total anthocyanin

The Two-Way ANOVA test showed a significant effect of harvest age and drying methods on tiwai. Air drying method produced high anthocyanin levels of 3.36 ± 0.22 and 5.86 ± 0.07 mg TAC/g at harvest ages of 5 and 9 months, respectively, which decreased to 0.77 ± 0.03 mg TAC/g at 7 months. When tiwai was dried in electric oven, the anthocyanin content tended to increase with higher age, while the use of microwave drying showed a decrease in levels at harvest age of 5 and 9 months. This result was consistent with B Enaru *et al.* [40] stating that changes in anthocyanin levels during drying with higher harvest age could be caused by enzymatic processes. Enzymes implicated in the biosynthesis and degradation of anthocyanin are active during aging and drying process.

Antioxidant capacity

The Two-Way ANOVA test results in **Table 2** showed that different harvest ages and drying methods had no significant effect on tiwai. Harvest age of 5 months with electric oven drying method had IC_{50} value of $69.63 \pm 1.18 \mu\text{g/mL}$, while air drying had IC_{50} of $92.91 \pm 2.14 \mu\text{g/mL}$, both classified as strong in inhibiting free radicals. A study by O Pramiastuti *et al.* [41] showed that tiwai extract had IC_{50} relatively strong in inhibiting free radicals.

Table 2 Polyphenol, anthocyanin, and antioxidant capacity of dried tiwai with different harvest ages and drying using air, electric oven, and microwave.

Sample	Drying method	Total phenol content	Total flavonoids content	Tannins content	Anthocyanin content	Antioxidant capacity IC_{50}
		(mg GAE/g)	(mg Qe/g)	(mg TAE/g)	(mg TAC/g)	($\mu\text{g/mL}$)
5 months	Air drying	130.26 ± 18.49	11.91 ± 1.21	$1,806.52 \pm 0.79$	3.36 ± 0.22	74.90 ± 0.51
	Electric oven	182.82 ± 9.43	14.43 ± 1.94	$1,983.93 \pm 43.21$	0.73 ± 0.03	69.63 ± 1.18
	Microwave	202.69 ± 8.16	12.50 ± 0.16	$2,016.52 \pm 51.07$	2.72 ± 0.13	74.26 ± 0.44
7 months	Air drying	188.21 ± 10.88	19.02 ± 1.52	$2,210.96 \pm 5.50$	0.77 ± 0.03	79.02 ± 0.88
	Electric oven	175.00 ± 13.96	12.80 ± 0.05	$2,115.04 \pm 69.93$	1.89 ± 0.08	70.90 ± 0.08
	Microwave	211.67 ± 22.30	11.58 ± 1.89	$2,079.48 \pm 33.78$	1.49 ± 0.06	70.12 ± 0.95
9 months	Air drying	117.70 ± 13.41	34.24 ± 3.46	$1,930.96 \pm 11.00$	5.86 ± 0.07	92.91 ± 2.14
	Electric oven	147.57 ± 8.52	20.36 ± 0.47	$2,166.52 \pm 17.28$	3.85 ± 0.10	77.56 ± 3.64
	Microwave	138.21 ± 11.60	13.80 ± 1.26	$2,168.00 \pm 3.93$	2.13 ± 0.14	82.32 ± 0.73

Fourier transform infrared spectroscopy (FTIR)

Figure 1, shows drying methods used, including air, electric oven, and microwave, depicted with codes A, B and C. FTIR analysis was performed to examine alterations in functional groups in tiwai harvested at 5, 7 and 9 months. Pullulan film drying was found to have a comparable phenomenon to the O-H stretching band signal at wave numbers $3,800 - 2,500 \text{ cm}^{-1}$ [42]. However, the O-H bending vibration band, observed at roughly $1,645 \text{ cm}^{-1}$, was unsaturated and offered important details about water molecules. The stretching vibrations of carboxylic groups were attributed to the absorption bands in the $1,600 - 1,350 \text{ cm}^{-1}$ range [43-45]. The absorption bands between $1,200$ and 950 cm^{-1} suggested elevated signals from C-C, C-O, and C-O-C vibrations connected to phenol groups and alkaloids, indicating the bioactivity of components with antioxidant activity [46]. Therefore, this study only observed the spectrum areas between $1,800$ and $1,350 \text{ cm}^{-1}$ as well as $1,200$ and 950 cm^{-1} .

Spectrum $1,800 - 1,350 \text{ cm}^{-1}$

FTIR spectrum of tiwai treated using various drying methods is presented in **Figure 1**, in the wave number range $1,800 - 1,350 \text{ cm}^{-1}$, attributed to increased water evaporation. The absorption bands at $1,415$ and $1,361 \text{ cm}^{-1}$ represent functional groups related to the bending vibrations of methyl and methylan groups (CH_2 , CH_3). These groups are related to changes in the chemical composition of tiwai, including variations in essential oils, lipid content, and other organic compounds [47,48]. The constancy of the wave numbers around $1,637$, $1,415$ and $1,361 \text{ cm}^{-1}$, observed in different drying methods including air drying, electric

oven, and microwave with harvest ages of 5, 7 and 9 months suggest insignificant changes in functional groups in the regions.

Spectrum 1,200 - 950 cm^{-1}

The overall absorption value of the spectrum gradually increases with different drying times in the wind drying method, electric oven and microwave drying time, namely 48, 24 h and 10 min respectively. Long drying times (24 and 48 h) can cause a certain degree of thermal degradation, causing changes in the C-C bond vibration and affecting the integrity of the ether (C-O-C) and hydroxyl (C-O) groups. This can result in partial breaking or weakening of carbon-carbon bonds as well as cleavage or rearrangement of these bonds, thereby reducing the ability to capture free radicals which affect the bioactivity of phenolic compounds and other antioxidants. Drying is fast (10 min) and uniform, resulting in rapid moisture reduction with minimal oxidation. The short exposure time helps maintain the integrity of the C-C bond, preventing significant thermal degradation. Meanwhile, fast drying can maintain C-O and C-O-C bonds thereby maintaining bioactive compounds and antioxidant properties due to minimal degradation of functional groups responsible for this activity [49]. **Figure 1** shows FTIR spectrum in the range 1,200 - 950 cm^{-1} due to the influence of drying time with different drying methods [46]. As illustrated in **Figures 1(A) - 1(C)**, the spectrum in the range 1,148 - 960 cm^{-1} showed almost the same band density and overlap. The second derivative was used in the spectrum to streamline the frequency analysis of the developing bands. The primary bands, which corresponded to the signal increase of C-C, C-O, and C-O-C vibrations, were observed at wave numbers 1,148, 1,075 and 993 cm^{-1} , respectively. In the context of herbal drying, the 1,148 cm^{-1} band, associated with the stretching vibrations of C-O or C-C bonds, may represent alterations in carbohydrates such as cellulose or hemicellulose. The 1,075 cm^{-1} band of this region is often associated with C-O stretching vibrations, suggesting the presence of carbohydrates, including polysaccharide and glycosidic bonds. Meanwhile, the band around 993 cm^{-1} can be associated with various functional groups, including C-O stretching in carbohydrates or the presence of aromatic compounds. In the context of bioactive components, this may be related to changes in the concentration of phenol compounds or aromatic structures [46,49]. The observation implies that the oxygen atoms in each drying method may have hydrogen bonds with water. Inter- or intra-chain hydrogen connections between molecules proliferate during drying process, as shown by functional groups that remain present at wave numbers 1,148, 1,075 and 993 cm^{-1} for each drying method and harvest age.

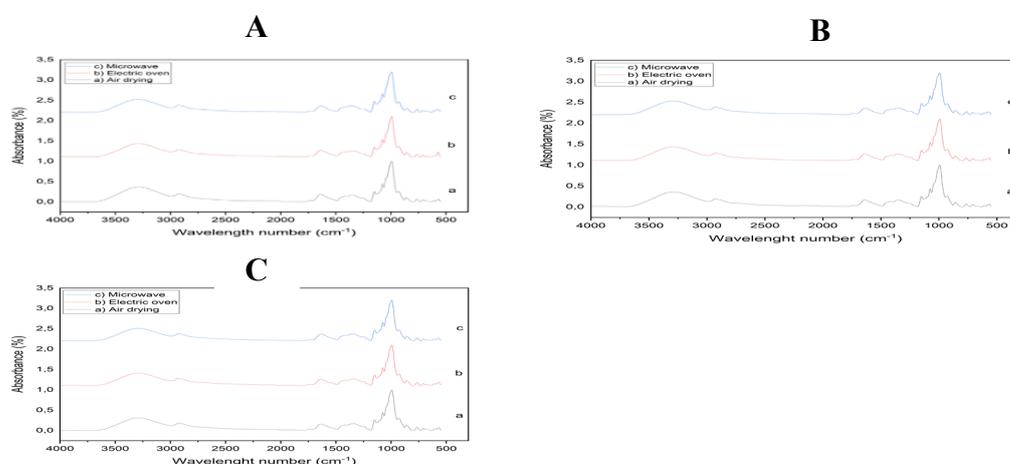


Figure 1 ATR FTIR of tiwai using various drying methods with harvest age (A) 5 months; (B) 7 months; (C) 9 months.

Scanning electron microscopy (SEM)

The effect of harvest age and different drying methods on the microstructural properties of dried tiwai powder samples was observed using SEM (**Figure 2**). Air drying of samples at 5 months of harvest led to the formation of dense granules with irregular size and intact cell shapes. This indicates slower moisture loss, thus giving more time for the particles to gather, forming a larger and denser structure so that resulting in low solubility and rehydration capacity. Larger particle sizes and higher densities mean that the surface area available for water interaction is reduced, making the particles more difficult to dissolve and inhibiting water penetration. Prolonged air exposure can also cause oxidative degradation, potentially reducing the overall retention of bioactive compounds [50].

When drying temperature increased in electric oven and microwave drying methods, the thermal effect became stronger on the microstructure of dried tiwai powder. This analysis was carried out based on changes in whole cells and certain elements, specifically using microwave drying, where tiwai powder tended to have a flatter structure and a brittle texture due to the susceptibility of electromagnetic waves to sudden temperature rise. The rapid drying process causes rapid evaporation of water, resulting in the structure not having time to form solid aggregates and instead becoming more compact and brittle. This structure causes increased solubility and rehydration capacity, faster water absorption and rehydration, because fragile particles can easily break and disperse in water. Rapid drying helps minimize thermal degradation of bioactive compounds due to shorter exposure times, however, higher temperatures can still cause loss of some heat-sensitive bioactive compounds [50]. Another study related to significant structural changes in surface size and shape due to the use of different drying methods including air drying, electric oven, and microwave was carried out by [51]. The results showed that oven and microwave drying led to cell size splitting in stevia leaves. Studies by N Izll and E Isik [52] on beans and R Lemus-Mondaca *et al.* [53] on stevia leaves produced almost the same results which focused on changes in cell structure with different drying methods.

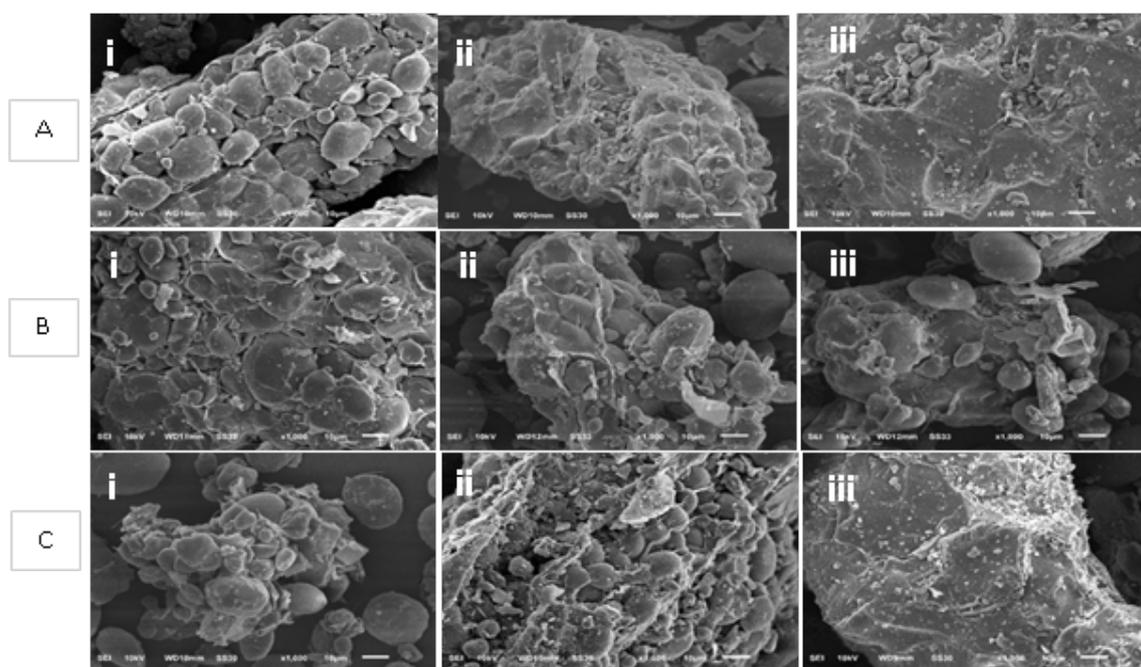


Figure 2 SEM of tiwai with harvest age (A) 5 months; (B) 7 months; (C) 9 months with different drying methods i) air drying, ii) electric oven, and iii) microwave.

Conclusions

In conclusion, the combination of harvest age and different drying methods influenced the proximate content, total phenol, total flavonoid, total tannin, and antioxidant capacity in tiwai. Based on the results, drying methods affected water content, with the lowest occurring in electric oven drying at harvest age of 7 months. Ash content was stable, while the highest fat content was produced by electric oven drying and harvest age of 9 months. Protein levels did not vary significantly, and total phenol was highest in microwave method, specifically at harvest ages of 5 and 7 months. Furthermore, the highest total flavonoid was obtained using air drying at 9 months of harvest and total tannin did not change significantly. Air drying method produced the highest anthocyanin levels at harvest ages of 5 and 9 months. Antioxidant capacity was relatively stable, with electric and air drying methods at 5 months of harvest showing significant results. FTIR analysis showed that changes in functional groups were not significant, while SEM images depicted transformations in the morphological structure during drying process.

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