

## Effect of varying processing methods (optimal conditions) on chemical properties of herbal leaf tea produced from “Voi” (*Syzygium nervosum*) leaves

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### Abstract

Tea is one of the most preferred food-drink due to its antioxidant potential benefit for human health. *Syzygium nervosum* leaves contain numerous bioactive components with different functional properties. *Syzygium nervosum* leaves can be processed into dried tea convenient for daily consumption. This research evaluates the effect of various thermal treatments like blanching, convective drying, roasting and steeping to the anti-nutritional factors, phenolics and antioxidant capacities in the *S. nervosum* dried leaf tea. Raw *S. nervosum* leaves were treated under different conditions of blanching (100/5, 95/10, 90/15, 85/20, 80/25°C/s), convective drying (65/10, 60/12, 55/14, 50/16, 45/18°C/hrs), roasting (150/2, 145/3, 140/4, 135/5, 130/6°C/mins), and steeping (100/2, 90/3, 80/4, 70/5, 60/6°C/mins). In each processing step, samples were taken to analyze tannin content, phytate content, oxalate content, total phenolic content, diphenylpicrylhydrazyl (DPPH) free radical scavenging and Ferric reducing antioxidant power (FRAP). Results show that *S. nervosum* leaves should be blanched at 90/15°C/s, convective drying at 55/14°C/hrs, roasting 140/4°C/mins, steeping in hot water at 80/4°C/mins to remove anti-nutrients while preserving the highest phenolics and antioxidants in the *S. nervosum* dried leaf tea. Results of this research will contribute important scientific data for processors and consumers in the exploitation of this medicinal plant as a healthy food drink.

## 1. Introduction

Tea (*Syzygium nervosum*) is one of the most popular beverages consumed around the globe, especially in Asia. It is a healthy food drink that contains natural antioxidants amongst other bioactive constituents that are ideal for daily consumption (Zhao *et al.*, 2019; Klepacka *et al.*, 2021). Herbal tea can be processed from different plant sources to exploit bioactive components such as phenolics, flavonoids, alkaloids, carotenoids, saponins, and terpenoids (Chandrasekara and Shahidi, 2018). “Voi” (*Syzygium nervosum*) belongs to the family Myrtaceae which is mostly distributed and grown in Vietnam as well as other Asian countries. Its leaves and flower buds are commonly consumed as a healthy drink by brewing in hot water. Its daily consumption is beneficial to combat influenza, bruise, acne, skin ulcer, and digestive disorder (Giang *et al.*, 2020). The major phytochemical constituents in *S. nervosum* are identified as triterpenoids, flavonoids, and polycyclic phloroglucinols (Su *et al.*, 2018; Song *et al.*, 2019). These bioactive elements contribute to pharmacological properties like antioxidant, anti-inflammatory,

antidiabetic, anticancer, and antiviral (Ha *et al.*, 2016; Tran *et al.*, 2019).

Blanching in advance of drying is required to obtain high quality of dried herbal tea, shorten drying time and minimize energy consumption (Deng *et al.*, 2019). The remarkable benefit of blanching treatment is the prevention of enzymatic browning. This thermal treatment retains more chlorophyll and better rehydration capability of the dried plant (Ahmed *et al.*, 2001). Unfortunately, this thermal treatment also decomposes a great number of bioactive ingredients in the herb (Oboh, 2005).

Convective dehydration is one of the most versatile techniques which are utilized in the food industry. Drying temperature, time, moisture gradient, and air velocity can be comprehensively controlled to overcome the disadvantages of sunlight (Orphanides *et al.*, 2016). To obtain efficiency, blanching should be carried out by a high-temperature short time (HTST) method to minimize the impact of heat on the thermal-sensitive components and maintain the organoleptic traits in the

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tea (Szychowski *et al.*, 2018).

Green tea is usually obtained by drying and roasting while black tea is usually obtained by fermentation (Traore *et al.*, 2013). Roasting is a unit operation in herbal tea production to prevent any excessively biological reactions, eliminate anti-nutrients, and modify functional properties (Minh, 2021). Roasting also enhances the favour of herbal tea (Tizian *et al.*, 2020). Secondary metabolites from the Maillard reaction are produced during roasting (Borrelli *et al.*, 2004). Roasting highly affects the consumer's preference, phytochemical content, and antioxidant property of herbal tea (Rungnattakan *et al.*, 2018). Roasting causes a remarkable influence on thermal-sensitive substances such as chlorogenic acids (Marcason, 2013). The thermal-sensitive decomposition under roasting depends on temperature, duration, and heating methods (Sharma and Gujral, 2011).

Herbal tea is commonly steeped in hot water for minutes to extract the most valuable components. However, inappropriate temperature and time during hot water infusion will be not beneficial for tea consumption (Damiani *et al.*, 2014). Steeping condition plays an important role in defining the healthy beneficial traits of tea (Castiglioni *et al.*, 2015).

To exploit *S. nervosum* leaves as a functional food drink, our study examined the effect of different thermal conditions in blanching, convective drying, roasting, and steeping to the anti-nutritional factors, phenolics, and antioxidant capacities in the *S. nervosum* dried leaf tea. These optimal processing parameters will improve the pharmacological properties of *S. nervosum* herbal tea in decoction.

## 2. Materials and methods

### 2.1 Materials

*Syzygium nervosum* leaves were collected in the Thanh Hoa garden of Binh Phuoc province, Vietnam. They were cleaned under tap water to remove foreign matters. Chemical reagents utilized in this research were all analytical grade. Phytic acid was purchased from Sigma (USA). EnzyChrom™ Oxalate Assay Kit was purchased from BioAssay Systems (USA). Tannin acid standard, phytic acid standard, and gallic acid standard were supplied from purchased from Fluka (Switzerland). Folin-Ciocalteu reagent, DPPH (2,2-Diphenyl picrylhydrazyl) reagent, FRAP reagent, 2,4,6-tripyridyl-s-triazine were supplied from Sigma Aldrich (USA). HCl, FeCl<sub>3</sub>, FeSO<sub>4</sub>.7H<sub>2</sub>O, Na<sub>2</sub>CO<sub>3</sub>, FeCl<sub>3</sub>.6H<sub>2</sub>O, methanol, ethanol, sulfosalicylic acid, Whatman filter paper were supplied from Dong Nam Co. Ltd. Lab apparatus and equipment include pipettor, test tube, cylinder, beaker,

Erlenmeyer flask, vortex mixer, steaming oven, convective drying oven, roasting oven, steeping pot, UV-Vis spectrophotometer.

### 2.2 Researching methods

Raw "Voi" *Syzygium nervosum* leaves were treated under different conditions of blanching (100/5, 95/10, 90/15, 85/20, 80/25°C/s), convective drying (65/10, 60/12, 55/14, 50/16, 45/18°C/hrs), roasting (150/2, 145/3, 140/4, 135/5, 130/6°C/mins), and steeping (100/2, 90/3, 80/4, 70/5, 60/6°C/mins). In each processing step, samples were taken to analyze tannin content, phytate content, oxalate content, total phenolic content, diphenylpicrylhydrazyl (DPPH) free radical scavenging and ferric reducing antioxidant power (FRAP).

### 2.3 Chemical evaluation

#### 2.3.1 Tannin content

Tannin content (mg/g) was measured following the method described by Adegunwa *et al.* (2011). An aliquot of 10 g sample was blended with 100 mL of distilled water. The blend was kept intact for 45 mins at 30±2°C before being filtered by Whatman No. 3 filter paper under vacuum pressure. An aliquot of 5 mL of the extract was dispensed into a 50 mL Erlenmeyer flask. About 5 mL tannin acid standard and 5 mL of distilled water were prepared in other individual Erlenmeyer flasks to serve as a standard and Folin-Ciocalteu reagent was supplemented to each of the flasks and 5 mL of saturated sodium carbonate was added. Each flask was filled up to 50 mL of distilled water and incubated at 30±2°C for 60 mins. Their respective absorbance was read by a spectrophotometer (model: UV5, Mettler Toledo) at 260 nm using the reagent blank to calibrate the instrument at zero. R<sup>2</sup> of the calibration curve was presented at 0.997.

#### 2.3.2 Phytate content

Phytate content (mg/g) was measured by the method described by Latta and Eskin (1998). Standard solution series in range 10-80 µg/mL phytic acid was prepared in distilled water. An aliquot of 2 mL of the standard solutions was dispensed into experimental tubes and a tube containing 2 mL of distilled water was also used as blank. To each test tube, an aliquot of 1 mL of the chemical reagent previously prepared from 0.05% FeCl<sub>3</sub>.6H<sub>2</sub>O and 0.5% sulfosalicylic acid in distilled water was added and the solution was well blended with a vortex. The mixture was then centrifuged for 10 mins and the absorbance of the supernatant was read at 500 nm by a spectrophotometer (model: UV5, Mettler Toledo). The standard curve was plotted by Microsoft Excel software. R<sup>2</sup> of the calibration curve was recorded at 0.995.

### 2.3.3 Oxalate content

Oxalate (mg/100 g) was examined by the colourimetric method using a high-throughput BioAssay Systems' oxalate assay kit (Danso *et al.*, 2019). Oxalate reacts with oxalate converter and oxalate enzyme mix to form an intermediate, which was, in turn, was visible by a spectrophotometer (model: UV5, Mettler Toledo). The modification in colour intensity of the reaction product at 595 nm was directly proportional to oxalate content in the sample. The assay kit can detect oxalate content in the range of 20-1500  $\mu\text{M}$ .  $R^2$  of the calibration curve was observed at 0.998.

### 2.3.4 Total phenolic content

Total phenolic content (TPC) (mg GAE/ 100g) was quantified by Folin-Ciocalteu reagent assay (Singleton and Rossi, 1965). The extract was dispensed in 90% ethanol (v/v) in a 10 mL tube and centrifuged at 3,000 rpm for 4 mins. Aliquot 1.5 mL of the extract was dissolved with 2.0 mL Folin-Ciocalteu reagent 10% (w/v). After 15 mins of reaction, 5.0 mL of  $\text{Na}_2\text{CO}_3$  (5% w/v) was added. The reaction occurred for 1.5 hrs in a dark place, the absorbance was read at 760 nm by UV-Vis spectrophotometer (Shimazu, UV-1800). The standard curve was plotted by Microsoft Excel software with a pure linear of gallic acid (0-300 mg/L).  $R^2$  of the calibration curve was observed at 0.94.

### 2.3.5 Diphenyl picrylhydrazyl (DPPH) free radical scavenging

Diphenyl picrylhydrazyl (DPPH) free radical scavenging ( $\text{IC}_{50}$ ,  $\mu\text{g}/\text{mL}$ ) was determined using UV-VIS spectrophotometer (Shimazu, UV-1800) with mobile phase methanol and water mixed online in the ratio of 85:15 (v/v), injected at a current speed of 0.8 mL/min. Aliquots of the samples 1.0 mL were supplemented with 4.0 mL of the 0.05 mM DPPH solution in the dark place, and the mixture was well-blended and kept for 30 mins at ambient temperature. DPPH absorbance curve identified by UV-Vis spectrophotometer (Shimazu, UV-1800) at wavelength 517 nm (Andriana *et al.*, 2019).  $R^2$  of the calibration curve was observed at 0.94.

### 2.3.6 Ferric reducing antioxidant power

Ferric reducing antioxidant power or FRAP (mg TE/100 g) was elaborated as power in reducing  $\text{Fe}^{3+}$  to  $\text{Fe}^{2+}$  by an antioxidant utilizing the protocol described by Benzie and Strain (1996). FRAP reagent was diluted with acetate buffer (100 mM, pH 3.5), 5 mM 2,4,6-tripyridyls-triazine in 25 mM HCl, and 15 mM  $\text{FeCl}_3$  at 7.0:1.5:1.5 (v/v/v). All standards and samples were prepared at 400 mg/L in methanol. The 50 mL reagent, the 10  $\mu\text{L}$  standard ( $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ ) or sample, and 10  $\mu\text{L}$  water were

dispensed into the well and gently mixed. The absorbance was read at 600 nm immediately and 45 mins after using a microplate reader. A standard calibration curve of  $\text{Fe}^{3+}$  sulphate pentahydrate ( $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ ) was plotted at concentrations between 500 and 2000 mM as the reference standard.  $R^2$  of the calibration curve was observed at 0.95.

### 2.4 Statistical analysis

The experiments were run in triplicate with different groups of samples. The data were presented as mean  $\pm$  standard deviation. Statistical analysis was performed by the Statgraphics Centurion version XVI. The mean value and standard deviation of a set of data were obtained by analysis of random samples estimating the population statistics. 95% of results will be expected to lie within the range  $\bar{x} \pm 2s$ . The lower and upper bounds of this range were described as the 95% confidence limits of the results. The differences between the pickling samples were analyzed using a one-way analysis of variance (ANOVA). A significant value was set at a 95% confidence interval ( $P \leq 0.05$ ). If significant differences were found, then post hoc analysis was performed using Duncan's multiple range tests.

## 3. Results and discussion

### 3.1 Blanching

The effect of blanching on anti-nutrient factors, phenolics, and antioxidant traits of *S. nervosum* was presented in Table 1. There was a significant difference in anti-nutrient factors by different blanching treatment conditions. Tannin, phytate, oxalate contents have a downtrend by lowering the blanching temperature (100°C down to 80°C) and extending the treatment time (5 to 25 s). Similarly, TPC and FRAP have a downtrend while DPPH (50% Inhibition concentration,  $\text{IC}_{50}$ ) increases by lowering the blanching temperature (100°C down to 80°C) and extending the treatment time (5 to 25 s). There was no significant difference in TPC, DPPH, and FRAP by blanching at 100/5, 95/10, and 90/15°C/s. Time causes much more influence than the temperature in blanching. Blanching at 90/15°C/s was selected to preserve a high amount of phenolics and antioxidants while removing a great amount of tannin, phytate, oxalate contents.

Blanching is proposed as a pre-treatment for the plant before dehydration to limit enzymatic browning and shorten drying time. Dried pepper receives better pungency, flavour, and colour by blanching before the main convective dehydration (Weil *et al.*, 2017). Asparagus blanched in hot water before convective drying obtains high reconstitution and fine structure

Table 1. Effect of blanching (°C/s) on anti-nutrients, total phenolics, and antioxidant traits of *Syzygium nervosum* leaf tea

Blanching (°C/s)	100/5	95/10	90/15	85/20	80/25
Tannin (mg/g)	118.20±0.17 <sup>a</sup>	101.42±0.14 <sup>ab</sup>	87.53±0.11 <sup>b</sup>	69.41±0.15 <sup>bc</sup>	57.15±0.12 <sup>c</sup>
Phytate (mg/100 g)	281.37±0.14 <sup>a</sup>	240.28±0.13 <sup>ab</sup>	203.71±0.15 <sup>b</sup>	165.82±0.10 <sup>bc</sup>	129.09±0.14 <sup>c</sup>
Oxalate (mg/100g)	139.42±0.18 <sup>a</sup>	126.11±0.12 <sup>ab</sup>	110.42±0.16 <sup>b</sup>	96.74±0.14 <sup>bc</sup>	82.51±0.13 <sup>c</sup>
TPC (mg GAE/100 g)	316.29±0.35 <sup>a</sup>	307.15±0.29 <sup>a</sup>	268.41±0.31 <sup>ab</sup>	217.42±0.29 <sup>b</sup>	106.29±0.34 <sup>c</sup>
DPPH (IC <sub>50</sub> , µg/mL)	46.79±0.08 <sup>c</sup>	49.73±0.06 <sup>c</sup>	57.24±0.05 <sup>bc</sup>	64.13±0.07 <sup>b</sup>	79.84±0.09 <sup>a</sup>
FRAP (mg TE/100 g)	89.45±0.14 <sup>a</sup>	87.31±0.12 <sup>a</sup>	80.15±0.11 <sup>ab</sup>	72.67±0.14 <sup>b</sup>	50.29±0.12 <sup>c</sup>

Values are presented as the mean±SD, n = 3. Values with different superscripts within the same row are significantly different ( $\alpha = 0.05$ ).

(Rodriguez *et al.*, 2017). The blanch-dehydrated basil leaves have high colour retention (Rocha *et al.*, 1993). Tannin, oxalate, and phytate content of vegetables is reduced by blanching (Adegunwa *et al.*, 2011). Blanching is proven to reduce the tannin content of green leafy vegetables (Mosha *et al.*, 1995; Ogbadoyi, 2012; Egbonu and Nzewi, 2016). Blanching at 95/5 (°C/minutes) causes a significant reduction of phytate in spinach leaves (William *et al.*, 2020).

### 3.2 Drying

The effect of convective drying condition (°C/hours) to anti-nutrient factors, phenolics and antioxidant traits of *S. nervosum* was presented in Table 2. There was a significant difference in anti-nutrient factors by different convective drying conditions. Tannin, phytate, and oxalate contents had a downtrend with increasing the drying temperature (45°C to 65°C) and shortening the treatment time (18 hrs down to 10 hrs). Similarly, TPC and FRAP had a downtrend while DPPH (50% Inhibition concentration, IC<sub>50</sub>) increases by accelerating the drying temperature (45°C to 65°C) and shortening the treatment time (18 hrs down to 10 hrs). There was no significant difference in TPC, DPPH, and FRAP by drying at 55/14 and 50/16°C/hours. Temperature causes much more influence than duration in drying. Drying was recorded at 55/14°C/hours to preserve a high amount of phenolics and antioxidants while removing a great amount of tannin, phytate, oxalate contents.

There is a tight relationship between drying

temperature and moisture removal, especially with a lower boiling point (Barbosa *et al.*, 2006). By convective dehydration, water is eliminated from raw materials to obtain dried particles (Calin-Sanchez *et al.*, 2020). There are two kinds of moisture including free and restricted water. Free moisture is conveniently escaped from raw material by diffusion, migration, and evaporation (Pydi-Setty and Ramana-Murth, 2003). Water reduction from raw materials supports for retardation of pathogen and spoilage microorganisms, minimization of an enzymatic reaction, and adaptation of sensory acceptance and extended shelf life (Ozbek and Dadali, 2007). The inappropriate temperature in convective drying induces major degradation of herbal flavours and colours (Fennell *et al.*, 2004). Too much shrinkage on the dried plant is a serious obstacle in convective drying (Orphanides *et al.*, 2016). Moisture removal will be a suitable approach to lower weight and volume hence supporting packaging, handling, and commerce. By low water activity, the dried herbal tea shows higher protection to microbial contamination and enzymatic reaction with prolonged stability (Mathlouthi, 2001). Low energy requirement in convective dehydration is very vital to minimize production cost towards sustainable development with minimal environmental impact.

A low air dehydration temperature is not suitable due to slow drying kinetic resulting in low production yield and more risk of fungi cross-contamination (Hevia *et al.*, 2002). Dehydration of surian leaves at 90°C needs a shorter duration than that of dehydration at 50°C

Table 2. Effect of convective drying (°C/hours) on anti-nutrients, total phenolics, and antioxidant traits of *Syzygium nervosum* leaf tea

Convective drying (°C/hours)	65/10	60/12	55/14	50/16	45/18
Tannin (mg/g)	17.42±0.11 <sup>c</sup>	29.36±0.12 <sup>bc</sup>	37.74±0.10 <sup>b</sup>	50.41±0.13 <sup>ab</sup>	64.23±0.16 <sup>a</sup>
Phytate (mg/100 g)	42.53±0.15 <sup>c</sup>	79.41±0.11 <sup>bc</sup>	107.28±0.12 <sup>b</sup>	136.54±0.16 <sup>ab</sup>	178.46±0.14 <sup>a</sup>
Oxalate (mg/100g)	19.50±0.13 <sup>c</sup>	37.18±0.16 <sup>bc</sup>	65.33±0.15 <sup>b</sup>	82.46±0.13 <sup>ab</sup>	103.15±0.11 <sup>a</sup>
TPC (mg GAE/100 g)	121.29±0.24 <sup>c</sup>	152.47±0.20 <sup>bc</sup>	231.27±0.25 <sup>a</sup>	204.42±0.21 <sup>ab</sup>	182.39±0.29 <sup>b</sup>
DPPH (IC <sub>50</sub> , µg/mL)	99.14±0.05 <sup>a</sup>	91.03±0.03 <sup>ab</sup>	55.27±0.04 <sup>c</sup>	71.25±0.06 <sup>bc</sup>	82.47±0.04 <sup>b</sup>
FRAP (mg TE/100 g)	20.04±0.16 <sup>c</sup>	29.91±0.14 <sup>bc</sup>	63.07±0.12 <sup>a</sup>	51.80±0.13 <sup>ab</sup>	40.36±0.10 <sup>b</sup>

Values are presented as the mean±SD, n = 3. Values with different superscripts within the same row are significantly different ( $\alpha = 0.05$ ).

(Ismanto *et al.*, 2017). The drying time of black tea at 130°C is shorter than that of 90°C (Teshome *et al.*, 2014). Drying at high temperatures can decompose thermal-sensitive constituents such as catechin of green tea (Cheong *et al.*, 2005). The tannin, total phenolic content and antioxidant capacity of herbal tea decelerate as accelerated drying temperature (Ismanto *et al.*, 2017). Convective dehydration utilizes hot air as a middle agent to exchange vapour between the sample and hot air current in the drying cabinet. The common benefits of convective dehydration can be easily realized as universal equipment, simple manipulation, effective cost, and stable shelf life. However, this dehydration technique also presents various hindrances like long-lasting drying time, decomposition of phytochemical substances, browning appearance, off flavour, and coarse outer layer on the dried samples (Saavedra *et al.*, 2017; Raghavi *et al.*, 2018). Convective dehydration produces the final product with changed structural traits like hard stiffness, high bulk density, more shrinkage, dense structure, low porosity. A remarkable decomposition of antioxidant properties, volatile evaporation can be highly recorded on this dehydration technique. Drying tends to degrade anti-nutrient in food (Youssef and Mokhtar, 2014). Sun-drying method is effective to eradicate anti-nutritional factors in okra (Matazu and Haroun, 2004), scent, and basil leaves (Abiodun *et al.*, 2012). Oxalate content in *Moringa oleifera* leaf is significantly reduced after drying (Joshi and Mehta, 2010). Anti-nutritional factors are significantly eliminated by convective drying and sun drying (Mercy *et al.*, 2015). Drying causes a reduction of oxalate due to the high thermal intensity which disrupts the plant tissue leading to the evaporation of oxalate in raw material (Danso *et al.*, 2019).

### 3.3 Roasting

The influence of roasting condition (°C/min) on anti-nutrient factors, phenolics, and antioxidant traits of *S. nervosum* was presented in Table 3. There was a significant difference in anti-nutrient factors by different roasting conditions. Roasting at 140/4°C/min shows the highest reduction of tannin, phytate, oxalate contents while preserving the highest phenolics and antioxidant

attributes. Roasting was recorded at 140/4°C/min for further experiment. Longer exposure time and higher temperature result in low phenolic content and low antioxidant capacity. The appropriate thermal treatment causes cell wall disruption and emission of antioxidants from insoluble particles.

Roasting utilizes dry thermal to modify the physicochemical, functional, nutritional, and phytochemical characteristics of plants (Oliviero *et al.*, 2008). There were 3 levels of roasting including brown, charred, and carbonized traits affecting the aroma, colour, and appearance of herbal tea (Xu *et al.*, 2018). Duration and temperature of roasting greatly affect water removal and browning (Zzaman and Yang, 2014). Roasting produces a mildly bitter taste and a light green for herbal tea. Roasting reduces a huge amount of tannin contributing to a lower astringent taste (Liang *et al.*, 2005; Zhang *et al.*, 2013). Thermal-treated samples induce chain-breaking and oxygen-scavenging capability (Jung *et al.*, 2004). Roasting barley at 280/20 (°C/s) causes an 8% reduction of phenolic content (Sharma and Gujral, 2011). DPPH of spearmint is greatly degradable up to 60 % under roasting (Lim and Murtijaya, 2007).

### 3.4 Steeping

The effect of steeping condition (°C/min) to anti-nutrient factors, phenolics, and antioxidant traits of decoction from *S. nervosum* leaf tea was presented in Table 4. There was a significant difference in anti-nutrient factors by different steeping conditions. Steeping at 80/4°C/min showed the lowest contents of tannin, phytate, oxalate while preserving the highest phenolics and antioxidant attributes. This is explained by the appropriate contact time between the sample and hot water. Several components from tea may be dissolved in hot water.

The longer steeping time (120 mins at ambient temperature) of green and white tea results in a higher total phenolic content than those in short time steeping (90/7°C/min) (Castiglioni *et al.*, 2015). However, a longer steeping duration also may cause the oxidation of phenolics (Reblova, 2012). The extended steeping

Table 3. Effect of roasting (°C/min) on anti-nutrients, total phenolics, and antioxidant traits of *Syzygium nervosum* leaf tea

Roasting (°C/min)	150/2	145/3	140/4	135/5	130/6
Tannin (mg/g)	35.02±0.08 <sup>a</sup>	29.24±0.11 <sup>ab</sup>	11.17±0.07 <sup>c</sup>	18.13±0.05 <sup>bc</sup>	23.19±0.04 <sup>b</sup>
Phytate (mg/100 g)	101.27±0.10 <sup>a</sup>	83.29±0.08 <sup>ab</sup>	25.03±0.09 <sup>c</sup>	41.27±0.07 <sup>bc</sup>	74.65±0.04 <sup>b</sup>
Oxalate (mg/100g)	47.12±0.12 <sup>a</sup>	36.07±0.10 <sup>ab</sup>	9.72±0.05 <sup>c</sup>	20.26±0.06 <sup>bc</sup>	28.73±0.05 <sup>b</sup>
TPC (mg GAE/100 g)	115.18±0.19 <sup>c</sup>	134.72±0.19 <sup>bc</sup>	223.19±0.21 <sup>a</sup>	201.05±0.23 <sup>ab</sup>	174.28±0.18 <sup>b</sup>
DPPH (IC <sub>50</sub> , µg/mL)	78.24±0.11 <sup>a</sup>	73.39±0.05 <sup>ab</sup>	59.37±0.06 <sup>c</sup>	64.28±0.11 <sup>bc</sup>	69.13±0.12 <sup>b</sup>
FRAP (mg TE/100 g)	21.17±0.03 <sup>c</sup>	29.43±0.07 <sup>bc</sup>	50.42±0.04 <sup>a</sup>	43.04±0.07 <sup>ab</sup>	34.26±0.09 <sup>b</sup>

Values are presented as the mean±SD, n = 3. Values with different superscripts within the same row are significantly different ( $\alpha = 0.05$ ).

Table 4. Effect of steeping ( $^{\circ}\text{C}/\text{mins}$ ) on anti-nutrients, total phenolics, and antioxidant traits of decoction from *Syzygium nervosum* leaf tea

Steeping ( $^{\circ}\text{C}/\text{mins}$ )	100/2	90/3	80/4	70/5	60/6
Tannin (mg/g)	64.28 $\pm$ 0.09 <sup>a</sup>	51.47 $\pm$ 0.08 <sup>ab</sup>	20.13 $\pm$ 0.05 <sup>c</sup>	31.18 $\pm$ 0.07 <sup>bc</sup>	43.28 $\pm$ 0.06 <sup>b</sup>
Phytate (mg/100 g)	87.12 $\pm$ 0.07 <sup>a</sup>	72.18 $\pm$ 0.11 <sup>ab</sup>	39.49 $\pm$ 0.10 <sup>c</sup>	47.65 $\pm$ 0.09 <sup>bc</sup>	61.23 $\pm$ 0.07 <sup>b</sup>
Oxalate (mg/100g)	39.74 $\pm$ 0.05 <sup>a</sup>	31.56 $\pm$ 0.09 <sup>ab</sup>	14.37 $\pm$ 0.07 <sup>c</sup>	18.61 $\pm$ 0.04 <sup>bc</sup>	24.05 $\pm$ 0.11 <sup>b</sup>
TPC (mg GAE/100 g)	289.14 $\pm$ 0.30 <sup>c</sup>	318.40 $\pm$ 0.25 <sup>bc</sup>	408.35 $\pm$ 0.49 <sup>a</sup>	386.24 $\pm$ 0.37 <sup>ab</sup>	352.17 $\pm$ 0.45 <sup>b</sup>
DPPH (IC <sub>50</sub> , $\mu\text{g}/\text{mL}$ )	61.16 $\pm$ 0.08 <sup>a</sup>	55.38 $\pm$ 0.07 <sup>ab</sup>	38.15 $\pm$ 0.08 <sup>c</sup>	43.07 $\pm$ 0.13 <sup>bc</sup>	49.72 $\pm$ 0.11 <sup>b</sup>
FRAP (mg TE/100 g)	59.13 $\pm$ 0.06 <sup>c</sup>	66.42 $\pm$ 0.04 <sup>bc</sup>	89.74 $\pm$ 0.07 <sup>a</sup>	81.25 $\pm$ 0.09 <sup>ab</sup>	73.14 $\pm$ 0.10 <sup>b</sup>

Values are presented as the mean $\pm$ SD, n = 3. Values with different superscripts within the same row are significantly different ( $\alpha = 0.05$ ).

duration greatly improves the total phenolic content of the extract prepared from the lotus petal that dried at 50 and 60 $^{\circ}\text{C}$  (Kaewka *et al.*, 2021). The longer steeping time has the potential to enhance the radical scavenging activity of the extract. The high total phenolic content of the extract contributes to high antioxidant activities from both DPPH and FRAP assays. The total phenolic content of herbal tea is positively correlated with antioxidant potential (Fu *et al.*, 2011). However, extended steeping duration or high temperature also greatly degrades the antioxidant capacity. This can be explained by the instability of flavonoid and anthocyanin during long steeping duration and thermal degradation. Phenolics possess numerous biological characteristics with powerful antioxidant activity against cancer and cardiovascular syndromes (Toor and Savage, 2005). Flavonoids are highly evaluated as promising health-promoting components owing to their anti-oxidative, anti-cancer, and cardiovascular defence (Ginwala *et al.*, 2019). DPPH may be beneficial in limiting the degree of reactive nitrogen species in life cells (Tetty *et al.*, 2014). The evaluation of antioxidant capacity to reduce iron reflects the power of one constituent to move an electron or hydrogen atom from another element to and an antioxidant capability to reduce the oxidized secondary substances in peroxidation (Gomez-Ordenez *et al.*, 2012). Phytochemical constituents are beneficial in scavenging toxic free radicals in the body emitted by numerous biological tension and ailments (Swain *et al.*, 2014).

Tannin highly migrates to infusions especially at long-time brewing due to its high molecular weight and high solubility in water (Garbowska *et al.*, 2018). Tannin is responsible for the organoleptic quality of tea infusions. Highly polymerized tannin consisting of abundant hydroxyl groups has antioxidant potential; however, it can be considered as an anti-nutritional factor in high dietary intake (Khasnabis *et al.*, 2015). Tannin is structured as a hydroxyl group bonded to an aromatic ring (Ozidal *et al.*, 2013). Tannin has a powerful binding capacity with proteins and minerals as well as non-specific enzyme inhibition. Tannin binds strictly

with the  $-\text{NH}_2$  segments of peptides and proteins limiting their metabolism (Shahidi and Naczka, 2004). Tannin retards  $\alpha$ -amylase, trypsin, and lipase resulting in lower metabolism of carbohydrates, proteins and lipids, respectively (Sugiyama *et al.*, 2007; Arimboor and Arumughan, 2011; Goncalves *et al.*, 2011). Moreover, tannin also combines with minerals leading to deficiency of micronutrients (Dixon *et al.*, 2005). Tannin is mostly decomposed in the gastrointestinal route by intestine microflora to release metabolites ready for absorption in the blood, the remaining is discharged via manure (Gu *et al.*, 2007; Ou and Gu, 2014). Immersion in the blanching process may reduce tannin content via a diffusion mechanism. Phytate is the salt form of phytic acid, myo-inositol hexakis dihydrogenphosphate. Phytate has a negative charge, therefore, it is easily bound with cation to create an insoluble complex. Phytate chelates with amino acid resulting in deficiency of protein digestibility and bioavailability (Angel *et al.*, 2002). Phytate also interacts with carbohydrates via hydrogen bonding with a phosphate group limiting starch solubility and bioavailability (Rickard and Thompson, 1997). Phytate is degraded in the gastric and tiny gut by phosphatase. In gastric, phytate interacts with minerals to form mineral-phytate in dissolved status. In the tiny gut, mineral-phytate begin precipitation induced to low digestibility of mineral (Schlemmer *et al.*, 2009). At the colon segment, phytate is degraded by phytase originating from gastrointestinal microbiota. High content of  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  retards the phytate dissolution and decomposition. Microbials in the colon participate in phytate hydrolysis to lower phytate content in manure (Schlemmer *et al.*, 2001). Phytate is mostly metabolized in the tiny gut, a small amount is absorbed in the blood (Schlemmer *et al.*, 2009). Immersion in the blanching process results in phytate loss through leaching (Duhan *et al.*, 1989). Around 96% phytate in legumes is decomposed in fermentation (Sathe and Venkatachalam, 2002). Oxalate has a strong ability to create insoluble particles via chelating different minerals, and vital micronutrients like iron, zinc, and calcium (Diana *et al.*, 2018). In oral intake, oxalate constrains with minerals in the gastrointestinal route and limited their metabolism.

Metabolism of oxalate happens in the gastric, tiny colon, and massive colon (Hatch and Freel, 2005). The huge gut is the main location for oxalate metabolism; soluble oxalate is metabolized via negative disperse (Hughes and Norman, 1992). Unavailable oxalate will be discharged in manure (Noonan and Savage, 1999). Soluble oxalate combines with calcium to turn into insoluble calcium oxalate, condense in renal and urinary route, saturate into calcium oxalate crystals (Marengo and Romani, 2008).

#### 4. Conclusion

Raw *S. nervosum* leaves can be converted into dried herbal tea after being blanched, convective-dehydrated, roasted, and steeped in hot water. This research has successfully investigated the impact of different thermal conditions on the anti-nutrients, phenolics, and antioxidant potentials in the dried *S. nervosum* leaf tea. Appropriate temperature and time during blanching, drying, roasting, and steeping will facilitate for removal of anti-nutritional factors like tannin, phytate, and oxalate while maintaining the most phytochemical constituents like phenolics and antioxidants. Consumers will have more chances to utilize this functional beverage in daily consumption.

#### Conflict of interest

The author strongly confirms that this research was conducted with no conflict of interest.

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