Sensorial and physicochemical characteristics of herbal noodle enriched with *Centella asiatica*

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Article history:
Received: 11 December 2019
Received in revised form: 31 December 2019
Accepted: 2 January 2020
Available Online: 26 February 2020

Keywords:
Herbal noodle, *Centella asiatica*, Antioxidant, Nutritional composition

Abstract

*Centella asiatica* is one of the traditional herbs found commonly in Malaysia. It has been used as an important ingredient in many traditional medicine practices due to its antioxidant and pharmacological properties. This study was done by adding three different percentages of *C. asiatica* extracts (20%, 40% and 60%) in noodles. Sensory evaluation involving appearance, colour, taste and aroma (affective test), physicochemical properties (texture, pH and colour), antioxidant activity (DPPH, FRAP, TPC and TFC assays) and nutritional composition on control noodle and herbal noodles enriched with *C. asiatica* were conducted. For sensory test, control noodle is the most favorable followed by 60% herbal noodle. There was a reduction in the firmness of cooked 20% and 60% herbal noodles. The pH value of herbal noodles also decreased as the concentration of herbal noodles increased due to the addition of *C. asiatica*. Colour analysis showed that the L* values of all the samples were increased, a* values decreased which indicated strong green colour among the herbal noodles while b* values showed the highest value for control noodle. Antioxidant tests such as DPPH showed that 60% herbal noodle exhibited higher free radical scavenging with a value of 49.200% as compared to control noodle (16.027%). The same finding was observed for FRAP assay where 60% herbal noodle displayed higher value (111.335 µg/mL) than control noodle (71.233 µg/mL). TFC of 60% herbal noodle also showed the highest value as compared to other noodles. However, TPC of 40% herbal noodles had greater value as compared to 60% herbal noodle. A decrease in the nutritional value of herbal noodle compared to control noodle was also observed. In conclusion, herbal noodles enriched with *C. asiatica* showed promising antioxidant potential which can be used in functional food applications.

1. Introduction

Wheat-based foods such as noodles are very popular as staple food in Malaysia. There are wide variability among noodles due to different ingredients and processing methods. The consumption of the noodles began 1000 years earlier. Now, noodles have been recognized worldwide and the demand is increasing globally (Hatcher, 2001). The most popular type of noodle is yellow alkaline noodle compared to regular salt noodle in South-East Asian markets where the ingredients used are flour, water and salts either alkaline or sodium chloride (Hatcher, 2007). In general, there are different types of noodles available in markets which are moist or fresh noodles, dry form noodles, boiled and steamed noodles. The preparation of these noodles are in the form of many different preferences such as hot or cold noodles, steamed, stir-fried, boiled or even served in soups. The quality of noodles set by the consumer is based on visual or appearance aspect of noodles such as colour (colour intensity), mouth feel, texture and appearance of the noodles (Hatcher, 2007).

Traditional noodles are known to be lack of other beneficial nutritional components such as antioxidant. Thus, noodle products with health potential benefits such as antioxidant rich are greatly needed. One of the sources of antioxidants is vegetables. In Malaysia, the Ministry of Health has been promoting the consumption of ‘Ulam’ or raw salad and vegetables under the Malaysian Dietary Guidelines (Ministry of Health Malaysia, 2010). The results from the third National Health and Nutrition
Examination Survey revealed that consumers who consumed salad and raw vegetable tend to increase serum level of vitamins, including vitamin C, vitamin E, folic acid, β-carotene, and lycopene (Su and Arab, 2006). In fact, people who always eat raw vegetables also tend to get higher likelihood of optimal mental states such as greater happiness and more life satisfaction (Lesani et al., 2016). *Centella asiatica* or ‘Pegaga’ is one of the traditional plants that is being consumed widely in Malay culture. This plant refers to a small green plant that creeps around wet areas in Malaysia and other countries such as Indonesia, India and China (Li et al., 2014). It grows wildly on the land of tropical and subtropical areas that contain high moisture and humus-rich soil, or sandy and clayey soils (Jamil et al., 2007). There are different types of ‘Pegaga’ or *C. asiatica* which are Pegaga Cina or Nyonya, Pegaga Daun Lebar, Pegaga Salad and Pegaga Renek (Anon, 2011a; Anon, 2011b).

*C. asiatica* has active components known as triterpenes and saponins that exhibit their role in medicine and nutraceutical fields (Loiseau and Mercier, 2000). Asiatic acid, madecassic acid, asiaticosside, and madecassoside are the most important active components in *C. asiatica* (Inamdar et al., 1996) for the usage in the quality assessment as biomarker components (Zheng and Qin, 2007). In Malaysia, the harvested *C. asiatica* contains only 3 types of triterpenes which are asiaticoside, madecassoside and asiatic acid and the highest amount of these components are found in leaves (Zainol et al., 2008). The consumption of *C. asiatica* is very common in Malay culture as it acts as antioxidant in natural ways. It is also very effective in improving the immune system especially in inhibiting free radical actions (Rajadurai and Prince, 2006) and balance the oxidative stress in our body (Kormin, 2005).

Interestingly, the antioxidant content in *C. asiatica* (84%) has been found to be similar with Vitamin C (88%) and grape seed (83%) (Hashim et al., 2011). In addition, other study found that the reducing activity of *C. asiatica* is higher than ascorbic acid, but lower than BHT (Suzanna, 2014). Study conducted by Zainol et al. (2003) showed that leaf extract possessed highest phenolic contents (8.13–11.7 g/100 g) than the other parts of the plants such as root and petiole with the values of 6.46–10.5 g/100 g and 23–4.91 g/100 g, respectively. Previous studies also found the presence of flavonoid compounds in *C. asiatica* such as rutin, quercetin, kaempferol, myricetin and catechin (Zainol et al., 2009; Andarwulan et al., 2010). These compounds are believed to exhibit the antioxidant properties in some vegetables and fruits (Hertog et al., 1992). Hence, this study aimed to develop healthy noodle enriched with *C. asiatica* which is rich in antioxidants and phytochemicals.

## 2. Materials and methods

### 2.1 Preparation of *Centella asiatica* extract

*C. asiatica* was washed and pad dried before blended with a portion of water according to the ratio: 20 g of *C. asiatica* in 100 mL of water for 20% herbal noodles and these same steps were repeated for 40% and 60% herbal noodles.

### 2.2 Noodle processing

In this step, there were three ratios of *C. asiatica* extract; 20%, 40% and 60% where the extract solutions of *C. asiatica* were mixed with 2 eggs and salt until the volume of the extract solution reached 200 mL. Next, the dry ingredient, wheat flour (500 g) was placed in a mixer. The extract solution was slowly added to the flour and the mixture was kneaded to form a solid dough in the mixer for about 1 min where several chemical and physical transformations occurred in order to homogenize all the ingredients (Fennema, 1996). Then, the dough was extruded to form noodle strands. Lastly, the extruded noodle was parboiled in boiled water mixed with oil for about 1 min before it was tossed and coated with oil again to prevent the noodles from sticking. For control, no *C. asiatica* extract was added into the noodle during processing.

### 2.3 Sensory evaluation

An affective test involving fifty untrained panelists from different ages, states and races was conducted at Sensory Analysis Laboratory of Universiti Tun Hussein Onn Malaysia (UTHM). All the samples were blind coded with 3 digits which differed from each other where control noodle, 20% herbal noodle, 40% herbal noodle and 60% herbal noodle were coded as 247, 223, 239 and 210, respectively. The panelists were asked to evaluate the colour, taste, appearance, odour, flavor and overall quality of the samples by using 9-point hedonic scale (varied from dislike extremely or like extremely).

### 2.4 Determination of noodle firmness

Firstly, a calibration of the texture profile analyser was performed which includes the height and weight of load cell before any testing was done. Each sample was evaluated using texture profile analyser model where a few strands (about 4-5 strands) of uncooked herbal noodles were placed on the metal plate and a flat probe was used. The sample was compressed twice at the speed of 50 mm/min and load cell of 50N before the required force needed to shear the noodle was processed using the Texture Expert software (Gull et al., 2016).

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2.5 pH test

A total of 5 g of both parboiled noodles were homogenised in 45 mL of distilled water using homogeniser and the pH values were measured using pH meter (Eutech Instruments, pH 700) (Norlaili et al., 2014).

2.6 Colour analysis

The colours of each sample which comprised of control and herbal noodles were measured using Colour Spectrophotometer (Hunter Lab 4500L Model MiniScans E2, USA) (Norlaili et al., 2014). Data were collected in three different values; L* value: lightness, a* value: greenness (negative value) and redness (positive value) and b* value: blueness (negative value) and yellowness (positive value) (Norlaili et al., 2014).

2.7 Determination of free radical scavenging activity

2.7.1 2,2-diphenyl-1-pyrylhydrazyl (DPPH)

Firstly, the extraction of sample was done according to Gull et al. (2016). A total of 2 g of each sample was extracted with 10 mL of 80% methanol. The mixture was stirred using magnetic stirrer for 2 hrs. Then, it was filtered using a filter paper and the supernatants were collected and stored in a freezer at -20°C. DPPH test was conducted according to Bakar et al. (2017) where DPPH solution was prepared by dissolving 5.9 mg of DPPH powder in 100 mL of 99.8% of methanol. Then, 77 µl of the extracted sample was pipetted into the test tube and 3 mL of DPPH solution was added to the test tube. The sample was left for about 15 mins before any reading was taken. After 15 mins, the absorbance reading was taken at 515 nm using UV-Vis spectrometry (Model T6u, PG Instrument, USA). The antioxidant activity was calculated as % of discoloration as the following equation:

\[
\text{% of discoloration} = \frac{\text{Blank DPPH} - \text{Samples}}{\text{Blank DPPH}} \times 100
\]

2.7.2 Ferric reducing/ antioxidant power (FRAP) assay

Extracted samples were diluted by dissolving 100 µL of the extracted sample with 300 µL of deionised water. Then, 2.5 mL of FeCl₃ 2.5 mL of TPTZ and 25 mL of buffer solution were added into the same test tube to form FRAP reagent (Benzie and Strain, 1996) before the addition of 100 µL of the sample. The sample was left for about 4 mins before the absorbance reading was taken at 593 nm using UV-Vis spectrometry (Model T6u, PG Instrument, USA).

2.8 Total phenolic content (TPC)

TPC was determined using Folin-Ciocalteu reagent (Hassan and Bakar, 2013). A total of 20 g of sodium carbonate was dissolved in 100 mL of distilled water to obtain a 20% (w/v) of sodium carbonate. Then, 200 µL of the sample was mixed with 3 mL of distilled water, followed by the addition of 0.5 mL of Folin-Ciocalte solution and left for about 3 mins. Next, 2 mL of 20% (w/v) sodium carbonate was added and left for about 1 hr. All these steps were prepared in the dark. Lastly, the mixture was vortexed and measured at 650 nm using UV-Vis spectrometry (Model T6u, PG Instrument, USA). Gallic acid was used as a standard and the results were expressed as µg gallic acid equivalents/ml (µg GAE/ml).

2.9 Total flavonoid content (TFC)

TFC was determined using aluminium chloride colorimetric method (Bakar et al., 2015). An aliquot (1 mL) of the extracted sample was mixed with 4 mL of distilled water in a test tube followed by the addition of 0.3 mL of 5% sodium nitrate solution and left to stand for 6 mins. Then, 0.3 mL of 10% aluminium chloride hexahydrate was added and the mixture was left to stand for 5 mins. 2 mL of 1M sodium hydroxide was added and left for another 5 mins. The mixture was vortexed and measured at 520 nm using UV-Vis spectrometry (Model T6u, PG Instrument, USA). Rutin was used as a standard and results were expressed as µg rutin equivalent/ml (µg RE/ml).

2.10 Determination of nutritional compositional

Analysis of protein and fat content was conducted based on AOAC (1984) while ash content was determined according to AOAC (2005) using moisture analyzer (AND MX 50). Carbohydrate content was determined by the difference in the following equation (Dusuki et al., 2019):

\[\text{Carbohydrates} = (\text{protein%} + \text{fat%} + \text{ash%} + \text{moisture%})\]

2.11 Statistical analysis

All experiments were carried out in triplicate and statistical analysis was done using one-way analysis of variance (ANOVA) followed by Duncan’s multiple range test. Differences were considered significant at p<0.05 and data were analyzed using Statistica software.

3. Results and discussion

For sensory evaluation, the control noodle and 60% herbal noodle have a similar rating. However, for 20% and 40% herbal noodle, they showed a lower preference in terms of taste attribute (Figure 1). Table 1 shows the results obtained for physicochemical analysis of control and herbal noodles. The texture of uncooked noodles had
greater value than cooked noodles. The decrement from 20 014.085 g force (uncooked) to 14 933.380 g force (cooked) was observed for control noodle while the greater decrement was observed in 60% herbal noodles as it decreased from 19 563.709 g force to 1, 140.245 g force. For pH value, it was observed that the pH value was slightly decreased from 6.743 to 6.557 as the concentration of herbal noodles increased. Colour analysis showed an increment in L* and b* values as the concentration of herbal noodles increased. Meanwhile, a* value which indicates the greenness showed decrement from -2.143 to -1.570 as the concentration of herbal noodle increased.

From the results obtained, it can be said that different concentration of *C. asiatica* extract incorporated in the noodles gave different preferences towards each sensory attribute as significant different (p<0.05) was observed for all attributes among the samples. 60% herbal noodle and control noodle showed similar preferences from the panelists except for taste. However, control noodle had no significantly higher value than 60% herbal noodle for all the attributes. Comparing between 20% and 40% herbal noodles, it was observed that the preference of panelists towards both of samples were almost the same. However, 20% herbal noodle had slightly lower value as compared to 40% herbal noodle. However, for aroma, both samples had observed that the preference of panelists towards both of samples. 60% herbal noodle and control noodle showed similar preferences from the panelists except for taste. However, control noodle had no significantly higher value than 60% herbal noodle for all the attributes.

Table 1. Results obtained for overall physicochemical analysis for both control and herbal noodles.

| Sample           | Texture (g force) | pH value | Colour value |
|------------------|-------------------|----------|--------------|
|                  | Uncooked          | Cooked   | L*           | a*         | b*         |
| Control          | 20 014.085        | 14 933.380 | 6.743±0.042 | 52.220±0.120 | -2.143±0.386 | 39.030±1.204 |
| 20% herbal noodle| 15 264.326        | 19 708.443 | 6.757±0.125 | 56.983±0.747 | -1.787±0.100 | 22.343±1.115 |
| 40% herbal noodle| 19 921.022        | 10 919.917 | 6.627±0.015 | 57.433±0.237 | -1.637±0.042 | 23.603±0.295 |
| 60% herbal noodle| 19 563.709        | 1 140.245 | 6.557±0.006 | 53.810±0.420 | -1.570±0.118 | 24.283±0.086 |

Table 2 shows the antioxidant activity of control and herbal noodles which include DPPH, FRAP, TPC and TFC tests. Based on the results obtained, 60% herbal noodle displayed the highest value as compared to that of control noodle for all the antioxidant assays; DPPH inhibition (60% herbal noodle: 49.200; control noodle: 16.027), FRAP (60% herbal noodle: 111.335; control noodle: 71.233), TFC (60% herbal noodle: 9.285; control noodle: 4.823) and TPC (60% herbal noodle: 69.875; control noodle: 55.866). In addition, the nutritional composition analysis showed that protein, fat, and carbohydrate of 60% herbal and control noodles were observed to be in the ranges of 5.9 to 6.0%, 2.3 to 2.7%, and 15.54 to 16.37%, respectively (Table 3).

Table 2. Results obtained for overall antioxidant assays of four different samples.

| Sample           | control noodle | 20% herbal noodle | 40% herbal noodle | 60% herbal noodle | % Inhibition DPPH | FRAP (µg/mL) | TFC (µg/mL) | TPC (µg/mL) |
|------------------|----------------|-------------------|-------------------|-------------------|------------------|--------------|-------------|-------------|
|                  | 16.027±0.960  | 28.007±0.898      | 29.319±0.458      | 49.200±2.943      | 57.433±0.237     | 72.885±1.112 | 69.875±1.499 |
|                  | 71.233±0.128  | 73.023±0.094      | 99.648±0.466      | 111.335±0.094     | 57.433±0.237     | 72.885±1.112 | 69.875±1.499 |
|                  | 6.005±0.093   | 6.274±0.081       | 4.823±0.000       | 9.285±0.047       | 55.866±1.150     | 72.885±1.112 | 69.875±1.499 |

Table 3. Results obtained for overall nutrition composition between control and 60% herbal noodles.

| Sample | Control noodle (%(w/w)) | 60% herbal noodle (%(w/w)) |
|--------|--------------------------|---------------------------|
| Protein| 6                        | 5.9                       |
| Fat    | 2.7                      | 2.3                       |
| Carbohydrate| 15.542              | 16.367                  |
| Ash    | 8.072                    | 7.983                    |
| Moisture| 67.687                | 67.45                     |

In this study, the cooking time was standardized to 1 minute and the results showed that cooked noodles had lower firmness than uncooked noodles. This might be due to the absorption of water during the parboiling process, resulting the decrease of compact structure of gluten. For uncooked noodles, it was observed that 60% herbal noodle had a higher value than 20% herbal noodle, but slightly lower than the control noodle. In general, the addition of herbal extract in the noodle has an effect on the disruption of the protein network.

For cooked noodle, it was observed that the firmness of herbal noodles decreased as the concentration of herbal noodle increased.
herbal incorporated in noodles increased. This happened likely due to the transition caused by thermal treatment during the parboiling process which resulted in denaturation of gluten when heated above its denaturation temperature (Nasehi et al., 2009). Firstly, monomer of proteins when being heated above 70 to 80°C causes other reactions to occur at disulphide or sulphhydryl chain (Schofield et al., 1983; Payne et al., 1987; Delcour et al., 2012). Secondly, the reduction of firmness occurs due to the overcooking process of noodles as the different concentration of herbal noodles have specific cooking time. In the previous study, it was stated that the cooking time for herbal noodles was shorter (i.e. 5% herbal noodle: 150 seconds, 10% herbal noodle: 130 seconds and 15% herbal noodle: 70 seconds) as compared to control noodle (Norlaili et al., 2014). Thus, overcooked noodles appeared to be soft and little soggy which make it easy to break (Park and Bail, 2004).

The highest value of L* obtained in this study (i.e. 40% herbal noodle: 57.433) was due to the colour loss during cooking (Gull et al., 2016). In addition, a* value indicates greenness of the noodle whereby the green colour of the noodles increased as the concentration of the herbal extract increased. This was clearly seen for the 60% herbal noodle which had a higher value (-1.570) and nearest to the zero while the lowest value was shown by control noodle (-2.143). Besides, b* value indicates yellowness of the sample which means, theoretically, it refers to the control noodle as the highest value was obtained (39.030).

A significant different (p<0.05) was observed for all the samples tested for pH value. Herbal noodle had a slightly higher pH than the control noodle. However, among the different ratios of herbal noodles, the highest ratio of herbal noodle; 60% herbal noodle in this case, had slightly lower pH value than the other herbal noodles. In previous study, the addition of herbal plant (Cosmos caudatus) resulted in a lower pH as the amount of herbal plant incorporated in the noodle increased (Norlaili et al., 2014). Hence, in this study, it was believed that lower pH values obtained in herbal noodles (i.e 4.5-5.5) were due to acidic properties exhibited by C. asiatica.

In addition, the results showed that they were statistically significant difference between all samples for DPPH scavenging free-radical activity (p<0.05). DPPH inhibition increased gradually as the concentration of herbal incorporated in the noodles increased. From this, it can be concluded that C. asiatica extract is one of the effective radical scavenging substances which is probably due to its phenolic compounds. As stated in the previous study, phenolic compound is known as hydrogen donor which has shown good antioxidant properties (Rice-Evans et al., 1995).

Based on Table 2, the results obtained have shown an increment of FRAP value from control noodle to herbal noodles and significant difference (p<0.05) was observed among the samples. There was 2.5% increment for 20% herbal noodle, 40% increment for 40% herbal noodle and 56% increment for 60% herbal noodle. This might be due to the presence of phenolic compound which acts as a reducing compound (hydrogen donor and singlet oxygen quenchers) and able to exhibit reduction effect (Sugunabai et al., 2015). For instance, quercetin-3, 5-diglucoside and cyanidin-3-sophoroside-5-glucoside exhibit reducing properties by breaking the free radical chain and donate a hydrogen atom to the specific compound (Gordon, 1990; Duh et al., 1999). Previous study also stated that the reducing activity of C. asiatica was higher than the reducing activity in ascorbic acid, but lower than BHT (Suzanna, 2014).

In this study, 60% herbal noodle had the highest TFC value than other noodles. There was a significant difference (p<0.05) observed between all samples. This may be due to the presence of flavonoid compounds that exhibit the antioxidant properties in some vegetables and fruits (Hertog et al., 1992; Wang et al., 1999). In previous studies, they found that flavons and flavonons components in C. asiatica exist in free form of 5-OH such as quercetin and rutin (Fatmawati, 2005). Besides, other components such as luteolin, apigenin, and kaempferol also found in some parts of C. asiatica (Zainol et al., 2009).

For the determination of TPC, there was a significant difference (p<0.05) observed between all samples and the TPC value increased gradually as the concentration of herbal incorporated in noodles increased. This might be due to the presence of phenolic components such as tocopherol and flavonoids. This finding was in an agreement with the previous study which stated that phenolic compounds that derived naturally have the potentials in exhibiting antioxidant properties. However, it was slightly decreased of TPC value in 60% herbal noodle in this study as the high temperature during the parboiling process might disrupt the phenolic compound interaction. In fact, there are few factors that may induce the degradation of antioxidants such as oxygen, water and heat treatment that are being applied especially during the food processing which contributes to the oxidative degradation of some components of antioxidants in the sample (Fares et al., 2010).

From the nutritional composition analysis, herbal noodles had slightly lower value compared to control noodle. According to Uma Maheswari (2012), tannin has
the ability in reducing protein content by decreasing the digestibility and palatability of the protein. Firstly, tannin disrupts the protein by binding to it and form tannin protein complexes, resulting protein availability to be decreased (Gopalan et al., 2007). Since complexes formed are insoluble, thus, it decreases the digestibility of protein (Bello et al., 2008). Secondly, tannin disrupts iron by binding to it and causes irreversible changes in the absorption of iron. Therefore, it can be said that the consumption of herbal noodles provides benefits for the dietary management of inherited metabolic disorders, liver failure and it may be introduced for medical purposes (Uma Maheswari, 2012). Meanwhile, the results of carbohydrate and moisture contents were in agreement with the previous study by Gull et al. (2016). Carbohydrate content may be affected by the incorporation of grains such as chickpea flour or soybean flour, seeds and dietary fibres as they provide benefits to hyperglycemia by reducing the glycemic index of products (Li et al., 2014). Both noodles (control and 60% herbal) had 67.45% and 67.69% moisture content, respectively. This might be due to the ingredient used in the noodles such as wheat flour. The starch content in flour is a hygroscopic substance that can absorb more water into the noodles (Rehman and Shah, 1999).

4. Conclusion
In conclusion, the development of herbal noodle has been accepted in our community due to health consciousness and its nutritional value. Herbal noodle enriched with C. asiatica also contains high antioxidant and phytochemical contents which can be used in functional food applications.

Conflict of Interest
The authors declare no conflict of interest.

Acknowledgments
The researcher would like to thank Universiti Tun Hussein Onn Malaysia (UTHM) for providing research grants (H277) and research facilities.

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