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Antioxidant Activity, Total Phenolics and Metal Contents of Ginger Powders in Hanoi, Vietnam

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Abstract

Ginger (Zingiber officinale Roscoe) is a valued food and medicine which contains biologically active substances and possesses healthpromoting properties. In the present study, the quality of ginger powders collected in Hanoi, Vietnam was investigated via determination of antioxidant activity, phenolic content and heavy metal concentrations. The results showed a good correlation between phenolic content of the fifteen ginger powders and their antioxidant activity (DPPH and ORAC scavenging capacity). However, large variation was found in the total phenolic content and antioxidant activity of the fifteen samples investigated. High levels of toxic arsenic and lead were found in sample S2 (9.05 and $11.45 \mu g/g$, respectively).

Keywords: Zingiber officinale; Ginger; Heavy metal; Total phenolic content; Antioxidant

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Introduction

Ginger is the rhizome of *Zingiber officinale* Roscoe belonging to the family Zingiberaceae. It is not only one of the most commonly used spices throughout the world, but also an important medicine in various traditional medicine systems. Various health benefits of ginger have been reported from traditional experience and scientific studies. The most well-known effect of ginger is the ability to treat nausea. Chewing ginger slices or drinking ginger tea effectively decrease symptoms of nausea in pregnant women and in patients receiving chemotherapy (Lete and Allue, 2016; Giacosa., *et al.* 2015). Ginger also improves digestive disorders, relieves gastrointestinal irritation, suppresses gastric contractions and speeds up the emptying of the stomach by 25% compared to placebo (Hu., *et al.* 2011).

The use of ginger can also relieve pain such as dysmenorrhea and arthritis pain (Daily., *et al.* 2015; Therkleson, 2014). Ginger has been widely used to treat inflammatory disorders such as osteoarthritis, rheumatoid arthritis, stomach and kidney inflammation (Kim., *et al.* 2017; Ribel-Madsen., *et al.* 2012). In cancer therapy, it not only targets cancer cells by inducing cell cycle arrest and apoptosis, but also ameliorates chemotherapy-associated side effects (Kaur., *et al.* 2016; Saxena., *et al.* 2016). Antidiabetic and anti-obesity properties of

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ginger are also other important benefits (Akash., *et al.* 2015; Wang., *et al.* 2017). In cosmetics, ginger has been used for minimizing agedrelated oxidative burden, treatment of cellulite, acne vulgaris and dandruff (Ngamdokmai., *et al.* 2017; Miglani and Manchanda, 2014; Mohanapriya., *et al.* 2013). The above health benefits are associated with the antioxidant capacity of phytochemicals in ginger such as polyphenols, essential oils and terpenoids (Sharifi-Rad., *et al.* 2017).

In Vietnam, ginger is consumed in fresh material or stored in dried powder for long-term use. However, the quality of dried ginger can be affected by drying methods and storage conditions, which can alter the phytochemical contents as well as antioxidant capacity. Trace elements are also important factors involving ginger quality. The essential metals such as Cr, Ni, Cu and Zn serve to maintain the metabolism of the human body, while the non-essential metals such as As, Cd, Pb and Hg are toxic to humans even in trace amounts (Pandotra., *et al.* 2016). Thus, the present study aims to determine the total phenolic content (TPC), antioxidant activity and heavy metals of fifteen ginger powder samples collected in Hanoi, Vietnam.

Materials and Methods

Ginger samples

Fifteen ginger powder samples were purchased from supermarkets and local markets in Hanoi during Jan–Feb 2018. All samples were dry powders, contained in plastic bags or bottles. Six samples (S1–6) from supermarkets were trademarked and were within their expiry dates, while the other nine ginger powders (S7–15) were from local markets and lacked producer names and expiry dates.

Determination of total phenolic content

The total polyphenol content was determined by the Folin-Ciocalteu method according to International Organization for Standardization (ISO) 14502-1 guidelines (ISO, 2005). The result was calculated based on the slope from serial dilution of a gallic acid standard. Results were expressed as gallic acid equivalents (GAE) mg/g of dry material.

DPPH radical scavenging activity

The antioxidant activity of the ginger powders was evaluated by its scavenging capacity of the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical (Huong., et al. 2012). Briefly, the ginger powder (1 g) was extracted with methanol (5 mL x 3 times) in a sonic bath for 10 min, and the combined extracts were filtered and evaporated until dryness. The extract residue was then dissolved in methanol at different concentrations. The ginger extract solution (10 μ L) was mixed with 190 μ L of 150 μ M DPPH (Sigma-Aldrich) solution in a 96-well plate. The plate was incubated in the dark at room temperature for 30 min. Then, the absorbance of the reaction mixture was measured at 520 nm on a microplate reader. Ascorbic acid (vitamin C, Sigma-Aldrich) was used as the positive control. The scavenging capacity (SC) was determined using the following formula:

%SC = $(A_{control} - A_{sample})/A_{control} \times 100$

Where, Acontrol is the absorbance of DPPH solution without sample;

 $\mathbf{A}_{\text{sample}}$ is the absorbance of sample-treated solution.

The concentration of antioxidant able to destroy 50% of the initial DPPH (SC_{50}) was calculated from the dose-response curve of %SC vs sample concentrations.

Oxygen radical absorbance capacity (ORAC)

The oxygen radical absorbance capacity of the ginger powders was evaluated as previously described (Nguyen., *et al.* 2012). In a 96-well microplate, 25 µL ginger extract solution (as prepared in section 2.3) was mixed with 150 µL of fluorescein solution (10 nM in phosphate buffer, pH 7.4) and then incubated at 37°C for 15 min. Fluorescence was measured (Ex. 485 nm, Em. 520 nm) every 90 s to determine the background signal. After three cycles of measurement, 25 µL of an 2,2'-azobis-2-amidinopropane dihydrochloride (AAPH,

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Sigma-Aldrich) solution (240 mM in phosphate buffer) was added via an automated injector and 60 fluorescence measurements were taken over a 90 min time period. The antioxidant Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid, Sigma-Aldrich) and ascorbic acid were used as positive controls. The antioxidant activity was normalized to equivalent Trolox units to quantify the antioxidant activity of each sample.

Metal content determination

Heavy metal measurements were carried out using inductively coupled plasma-mass spectrometry (ICP-MS 7900 Agilent Technologies). Ginger powder (0.5 g) was weighed into the digestion vessel, to which was added 10 mL of 65% HNO₃ and 3 mL H₂O. The samples were then digested in a microwave oven. Digestion conditions were set at 210°C for 15 min and then constant for 15 min. After cooling to ambient temperature, the reactors were opened, and the content was quantitatively transferred into a 50 ml volumetric flask and brought to the volume with ultrapure water. All digested solutions were analyzed by ICP-MS. The operating conditions were: RF power: 1600 W; sample depth 10 mm; nebulizer gas flow rates: 0.7 L/min; auxiliary gas flow: 0.3 L/min; standard spray chamber temperature: 2°C; rinse time 20 s. Data acquisition was performed in spectrum analysis and full quant mode. Mass calibration and detector crosscalibration were performed according to the manufacturer's instructions using the prescribed solutions obtained from Agilent. Multielement standards were prepared in-house by mixing of certified, traceable, ICP grade single element standards (Sigma Aldrich) that were subsequently diluted for analysis.

Statistical analysis

Results were expressed as means \pm standard deviations (SD) of three experiments. Statistical analysis was performed using Student's t-test and p < 0.05 was considered to be significant.

Results and Discussion

Antioxidant activity and total phenolic content

2,2-Diphenyl-1-picrylhydrazyl (DPPH) is a stable organic free radical that exhibits a purple colour in solution with an absorption band at around 520 nm. It loses this absorption when it accepts an electron or a free radical species, resulting in a visually noticeable discolouration from purple to yellow. The DPPH assay is often used to evaluate the ability of antioxidants to scavenge free radicals, which are known to be a major factor in biological damage caused by oxidative stress. This assay gives reliable information concerning the antioxidant ability of the tested samples (HUANG., *et al.* 2005). The DPPH scavenging capacity of ginger powder extracts are presented in Table 1. The results show that sample S1 possessed the highest activity (SC50 = $86.4 \mu g/mL$), followed by S6 (SC50 = $88.1 \mu g/mL$). The lowest antioxidant capacity was found for sample S7 and S8 (SC50 = $256.9 \text{ and } 346.1 \mu g/mL$, respectively).

The antioxidant capacity of ginger powder extracts were also evaluated by an ORAC assay. Similar to the case of DPPH scavenging capacity, samples S1 and S6 exhibited strongest antioxidant activity with the ORAC values of 0.82–0.83, which were comparable with those of Trolox (ORAC value of 1). S7 and S8 were the least effective antioxidants.

The antioxidant activity of plant extracts containing phenolic components is due to their capacity to be donors of hydrogen atoms or electrons and to capture the free radicals (Gonzalez., *et al.* 2003). Thus, the total phenolic content of ginger powders was investigated. The levels of phenolic constituents in ginger powders were determined using the Folin-Ciocalteu reagent and are presented in Table 1. The average amount of total phenolics was 9.24 mg GAE/g, but there was large variation in the total phenolic content (TPC) of the samples investigated. The maximum TPC was found for sample S1 (16.87 mg GAE/g), followed by S6 (14.37 mg GAE/g). Sample S8 contained the lowest amount of phenolic components (2.29 mg GAE/g). The Folin-Ciocalteu assay to determine total phenolic concentrations is based on an electron transfer mechanism, and typically has a high degree of linear correlation with DPPH and ORAC antioxidant capacities (Dudonne., *et al.* 2009). In fact, we found a good correlation between phenolic content of the fifteen ginger powders and their antioxidant activity.

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Samples	DPPH, SC ₅₀ (µg/mL)	ORAC (Trolox equivalent)	TPC (mg GAE/g)	
S1	86.4 ± 12.61	0.82 ± 0.03	16.87 ± 2.72	
S2	205.7 ± 15.96	0.34 ± 0.01	10.33 ± 1.38	
S3	110.6 ± 9.64	0.55 ± 0.02	7.77 ± 0.56	
S4	157.4 ± 22.52	0.35 ± 0.02	7.85 ± 0.51	
S5	168.3 ± 14.76	0.46 ± 0.03	9.53 ± 0.74	
S6	88.1 ± 10.12	0.83 ± 0.04	14.37 ± 0.95	
S7	256.9 ± 21.68	0.21 ± 0.01	4.96 ± 0.37	
S8	346.1 ± 41.54	0.17 ± 0.01	2.29 ± 0.35	
S9	268.2 ±16.21	0.42 ± 0.01	4.17 ± 0.26	
S10	147.6 ± 9.55	0.58 ± 0.02	8.05 ± 0.52	
S11	133.9 ± 8.47	0.71 ± 0.03	12.61 ± 0.79	
S12	215.2 ± 16.35	0.40 ± 0.01	3.84 ± 0.16	
S13	231.8 ± 13.84	0.49 ± 0.02	5.15 ± 0.23	
S14	120.4 ± 7.92	0.74 ± 0.03	13.50 ± 0.75	
S15	194.6 ± 11.60	0.38 ± 0.01	6.71 ± 0.24	
Ascorbic acid	22.4 ± 1.52	0.52 ± 0.03	-	

Table 1: Antioxidant activity and total phenolic content of ginger powders collected in Hanoi,

 Vietnam (Values are mean of triplicate measurements ± standard deviation).

Ginger powders are exposed to various conditions including cultivation, processing, packing and storage. Any improper condition may result in instability of the phytochemical composition and consequently impact product quality. Since many phenolic substances are sensitive to heat and sunlight, the large variation in the total phenolic content among the ginger powders investigated might be due to the drying process (Ajayi, *et al.* 2017).

Normally, ginger is sliced and dried under direct sunlight, in a solar tunnel drying system or an oven. Several factories or farms dry crop products by using conventional ovens heated by fossil energy or wood burning. The heating temperature is not well controlled in those kinds of ovens, and therefore chemical decomposition will occur when overheating. A previous study reported that a temperature of 60°C was considered optimum for drying ginger (JAYASHREE., *et al.* 2014). Direct sun-drying is very popular in Vietnam due to its simple and low-cost advantages. The unknown-source samples S7–9 purchased from local markets may have been home-made products, which are usually prepared by simple methods such as sun-drying. Long-exposure time under a scorching sun may have caused the decrease of TPC in S7 and S8.

Metal concentration analysis

Table 2 illustrates the concentrations of Cr, Mn, Ni, Cu, Zn, As, Cd, Hg and Pb in fifteen ginger powders analyzed, expressed in μ g/g. Among nine trace metals investigated, Mn (29.03–357.86 μ g/g) was the most accumulated metal followed by Zn (15.39–29.99 μ g/g) and Cu (1.08–11.30 μ g/g). The Cd and Hg elements were also detected at low concentrations in all tested ginger samples. The average values of the metal concentrations in the fifteen ginger powders were decreased as Mn>Zn>Cu>Ni>Pb>As>Cr>Cd>Hg.

Manganese was found at low concentrations (29.03 μ g/g) in sample S8. The ginger S3 contained the highest levels of Mn (357.16 μ g/g). Manganese is an essential element for many living organisms, including humans. Adverse health effects such as Parkinson-like syndrome can be caused by overexposure of Mn (Avila., *et al.* 2013). It has a recommended daily intake of 2 mg/day, based on the upper range value of manganese intake of 11 mg/day (Nebguide, 2015).

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Samples	Cr	Mn	Ni	Cu	Zn	As	Cd	Hg	Pb
S1	0.78	164.82	3.03	7.40	26.38	3.56	0.11	0.08	2.90
S2	4.96	303.86	6.88	11.30	53.31	9.05	0.21	0.06	11.45
S3	0.47	357.16	3.38	4.88	19.63	0.32	0.15	0.06	0.33
S4	0.42	303.53	3.18	4.62	15.39	0.27	0.13	0.06	0.40
S5	0.93	124.08	3.23	8.45	29.99	0.83	0.19	0.02	0.79
S6	1.58	278.63	4.52	6.04	26.76	1.82	0.32	0.03	2.91
S7	1.20	82.61	4.40	10.80	41.13	0.21	0.05	0.04	0.90
S8	0.32	29.03	2.29	2.92	15.76	0.39	0.04	0.04	0.43
S9	0.28	54.1	1.82	1.08	20.4	0.24	0.08	0.03	0.61
S10	0.53	76.8	3.14	2.85	33.7	0.66	0.05	0.03	0.75
S11	0.62	86.1	2.09	3.10	19.8	0.85	0.10	0.02	1.06
S12	0.24	108.2	1.93	2.04	22.6	0.27	0.06	0.04	0.95
S13	0.48	92.7	3.07	2.66	19.3	0.61	0.11	0.06	0.62
S14	0.95	168.5	2.48	1.26	33.1	0.55	0.14	0.05	0.46
S15	0.84	67.4	1.50	2.17	28.6	0.40	0.08	0.04	0.75
Average	0.97	153.17	3.13	4.77	27.06	1.34	0.12	0.04	1.69

Table 2: Metal concentrations $(\mu g/g)$ in ginger powders collected in Hanoi, Vietnam.

The toxic heavy metals such as As, Pb, Cd, Hg are known to be hazardous to humans. Inorganic arsenic is a poison that increases the risk of cancer. According to World Health Organization guidelines (WHO, 2010), the arsenic benchmark dose lower confidence limit for a 0.5% increase in the incidence of lung cancer in humans (BMDL0.5) was 3.0 μ g/kg body weight per day. The arsenic content of the tested samples ranged from 0.21 to 9.05 μ g/g. The mean value was 2.06 μ g/g and the highest as concentration was found in sample S2 (9.05 μ g/g), followed by S1 (3.56 μ g/g) and S6 (1.82 μ g/g). Thus, the consumption of such gingers poses a serious health risk. Similar to the as level, the highest content of Pb in the fifteen ginger powders was found in S2 (11.45 μ g/g), followed by S1 (2.90 μ g/g) and S6 (2.91 μ g/g). Exposure to high levels of lead causes brain and kidney damage, paralysis and gastrointestinal symptoms. Children are particularly sensitive to lead toxicity, and even relatively low levels of lead exposure probably induce irreversible neurological damage. A provisional tolerable weekly intake of 25 μ g/kg body weight has been recommended for lead (EC, 2004). Soil and underground water are the main sources of heavy metals that contaminate plants. The high levels of as and Pb in several ginger samples may have been due to them being sourced from polluted areas.

Conclusion

In this work, fifteen ginger powders collected in Hanoi, Vietnam were investigated for their quality in terms of antioxidant capacity, total phenolic content and heavy metal concentrations. To the best of our knowledge, this is the first time that the quality of ginger powders in Vietnam has been reported. The results show that antioxidant capacity was directly proportional to total phenolic content. Significant differences existed in total phenolic content among the samples investigated. For trace elements, toxic metals as and Pb were found in high concentrations in sample S2. From the above results, it can be concluded that the quality of ginger in Hanoi, Vietnam is not well controlled and may pose a high health risk to consumers.

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Conflict of interest

We declare that we have no conflict of interest.

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