Evaluate the Chemical Composition of Peels and Juice of Seedless Lemon (*Citrus latifolia*) Grown in Hau Giang Province, Vietnam

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**Abstract.** In this study, the specific chemical composition of Seedless Lemon (*Citrus latifolia*) grown in Hau Giang Province, Vietnam was determined. The experiment was conducted based on the baseline methods referenced from previous studies. The results of the component survey show that, in Persian Lime, the acidity is 0.637 ± 0.08 g/L and 4.916 ± 0.03 g/L respectively with lemon peel and lemon juice (pH is 4.5 and 2 respectively). The basic components are also determined with the parameters corresponding to lemon peel and lemon juice including: Brix 2% and 8%, ash 1.780 ± 0.004% and 0.314 ± 0.002%, lipid content 2.93 ± 0.044% and 0.542 ± 0.051%. In which, the content of vitamin C in lemon juice is quite good, reaching 8.492 ± 0.28 mg/g. This research helps shape the basic parameters of local Vietnamese raw materials, thereby laying the premise for the development of related products.

1. Introduction

Lemon is a flowering plant, that belong to the Rutaceae family. The species is native to the tropical and subtropical regions of Southeast Asia [1]. Lemon was historically described as the ornamental plant in the first Islamic gardens [2]. However, concurrent applications of lemon have been extensively recognized. In food industry, lemon essential oils have been used as a natural additive as a substitute for artificial preservatives to improve shelf life and enhance the safety of products. In addition, potent antibacterial activities and the abundance of bioactive ingredients such as citric acid, ascorbic acid, minerals, flavonoids of lemon essential oils have suggested its application as a preservation agents against foodborne illnesses [3]–[5]. It has been shown that vitamin C (ascorbic acid), predominantly
found in lemon, is capable of preventing oxidative damage in the tissues of mammals both at the intracellular and extracellular level [6]. In medicinal fields, lemon juice is often used to treat scurvy, sore throat, fever, rheumatism and high blood pressure. [7].

A wide range of beneficial natural compounds could be found in lemon juice and lemon peel extract including prenyloxycomarin such as aurapten, bergamottin, inva-torin, heraclenin, and oxypeucesanin [6], [8]–[11]. Furthermore, the essential oil extracted from the outer lemon crust has been shown to contain ingredients such as citronellal and limonene, α-terpineol, geranyl acetate and linalyl [12]–[14]. As a result, lemon essential oil obtained from thin crust via traditional distillation method is widely applied as a fragrance enhancer in beverages, foods and confectionery. Moreover, it is also a fragrance agent to hide the unpleasant taste of the drug in pharmaceuticals and as a fragrance in perfumes as well as in the cosmetics industry [15], [16].

Currently, about 140 genes and 1300 species present in citrus genera have been found [3]. In Vietnam, Citrus latifolia (Persian Lime) was grown in numerous regions and has been shown to be easily adaptable to climatic and soil conditions. However, it was not until Citrus latifolia got certified by VietGAP and GlobalGAP standards that the initial output and the growing area of Citrus latifolia began to expand. Currently, Persian Lime (PL) has been widely accepted and exported to European countries. Despite the increasing popularity of the species, in-depth studies on the chemical composition of seedless lemons have remained limited [17]. Therefore, this study evaluated the chemical composition of seedless lemons grown in Hau Giang province, Vietnam. The results are expected to contribute in further developments on application of Seedless Lemon (Citrus latifolia) in manufacture of useful bioactive agents.

2. Material and method

2.1. Materials

Fruits of Seedless Lemon (Citrus latifolia) grown in Hau Giang Province, Vietnam was harvested in early March 2020. The material was transported to the Advanced Materials Laboratory, NTT Hi-Tech Institute, Nguyen Tat Thanh University. Afterwards, the material was washed in tap water, dried at room temperature, then refrigerated at 10°C to provide the basis sample for the follow experiment. Two types of materials were analyzed including lemon peels and lemon juice.

2.2. The tracking criteria [18,19]

Basic properties of the lemon material analyzed include: moisture content (AOAC 934.06), ash content (TCVN: 5253-90), content of soluble dry matter (°Brix) (using Brix level meter ATAGO (model RHB0-90-Total Meter), pH and total dissolved solid content (TSS) (using Consort multi-parameter analyzer (model C3010T)).

2.3. Protein content analysis [20]

Kjeldahl analysis was adopted to determine total nitrogen. The procedure commenced with the addition of 1 g of potassium sulfate into 1 g of solid sample to increase the boiling point of 3 ml of concentrated sulfuric acid. Following that, 0.1 g of copper sulfate and iron sulfate were added prior to chemical sampling to enhance the acid oxidation. The mixture was then allowed to stand for 6 hours at 370°C and 10 mL of the mixture was then calibrated with boric acid. The results is given as the nitrogen percent per gram of dry matter.

2.4. Analysis of total fat content [21]

Differences between mass determined before and after Soxhlet extraction was used as the basis to determine total fat content. The process began with the addition of 5 g of dry sample in a tube with diethyl ether solvent. The heating lasted 48 hours at 60°C. The volume of the solution was then recorded after the extraction had been completed and the flask had been dried completely. The result is given as mg lipid per gram of dry matter.
2.5. Determination of carbohydrate content [22]
Carbohydrate content was determined by the following formula:
Carbohydrates (%) = 100 - (Moisture (%) + Ash level (%) + Protein content (%) + Lipid content (%)).

2.6. Determination of total fiber content [23]
Lignin, hemicellulose and ash were removed from the material with H\(_2\)SO\(_4\) 1.25%, NaOH 1.25% and ethanol 90%. The material was then dried to constant mass. Afterwards, the first mass \((m_1)\) was weighed and recorded. The amount of cellulose in the sample was calculated by following formula \((X: \text{percent of total fiber in the sample} \, \text{(%), sample weight (g)}\):

\[
\%X = \frac{m_1 - m_2}{m} \times 100
\]

2.7. Determination of acidity [24]
The prepared samples were mixed, weighed 10 g to the nearest 0.001 g, transferred into a 250 ml conical flask with distilled water, added water to about 150 ml and heated on a water-bath at 80°C for 15 minutes. The mixture was then quickly mixed, then transferred into 250 ml volumetric flask, mixed well with water, and allowed to settle. The sample was filtered, and the filtrate was transferred into the beaker. 25 ml of the filtrate was transferred into a 100 ml conical flask. Afterwards, 3 drops of 0.1% phenolphthalein was added and titrated with 0.1 N NaOH to a solid pink color for 30 seconds. The results were titrated recorded 3 times.

2.8. Determination of total ascorbic acid content [25]
A previously described DCPIP titration method was employed to determine total ascorbic acid content. The method relies on the formation of dehydroascorbic acid and the colorless lenco derivative of Vitamin C oxidation with 2,6 dichlorophenolindophenol. Excessive reaction of 3-4 drops of DCPIP leads to optimization of the pH in the bottle as a light pink solution in 30 seconds.

2.9. Determination of reducing sugar content [26]
Dinitrosalicylic acid (DNSA) was used to measure reducing sugar content. One milliliter of alcohol was extracted from the sample, which was then made up to 10 ml with distilled water. Then, a test tube containing one mL of the mixture and 0.5 ml of DNSA was maintained in a boiling water bath for 5 minutes. After being cooled, the tubes were made up to 20 ml. The sample was measured at a wavelength of 540 nm. The result was given as a percentage of reducing sugar per 100 gr of fresh sugar.

3. Result and discussion
Table 1 presents the chemical properties of lemon peel juice and lemon juice from Persian lemon. Based on Table 1, there is a difference between the chemical composition of lemon peel and lemon juice. Firstly, the acidity of lemon juice is about 7 times higher than that of the lemon peel solution. This is related to the higher pH value of the lemon peel solution \((\text{pH} = 4.51)\) compared to the lemon juice solution \((\text{pH} = 2.076)\). This result is similar to the study of Kokate et al. (2008) in which the acidity in lemon juice was about 5% and the pH value changed from 2 to 3 [27].

The Brix value represents the concentration of dissolved solids (%) in the solution. A lemon juice solution has a brix of 8, indicating that every 100g of lemon juice contains 8 g of dissolved solids and 92 g of water. Similarly, lemon peel juice has a brix of 2 (2 g of dissolved solids and 98 g of water). The moisture in lemon peel is about 60.96%. Compared to previous study of Mohammed et al. (2013), the moisture content obtained higher [28]. This is explained by the climate and soil conditions leading to different moisture content of the material. The moisture of lemon peel is in the humidity range of about 0.2 - 84.33%, which is in accordance with the research of García-Perez et al (2008).
Besides, the content of vitamin C (mg) in lemon juice is about 1.4 times higher than in lemon peel. The value of vitamin C in lemon juice (8.392 mg / g) is higher than that in a previous investigation of Nguyen Bao Ve and Le Thanh Phong (2011) [29]. Scientific studies have shown a significant antioxidant effect of *C. latifolia* fruit extracts, suggesting their use in anti-ageing cosmetics. In particular, vitamin C from *C. latifolia* is used as an ingredient in dermatological cosmetics. Its external use increases collagen production, which makes the skin smoother and more tense. It is also used in anti-aging products to reduce shallow wrinkles, and as a synergistic antioxidant in combination with vitamin E [30].

### Table 1. Chemical properties of lemon peel and lemon juice

| No | Evaluation criteria     | Lemon peel          | Lemon juice          |
|----|-------------------------|---------------------|----------------------|
| 1  | Humidity (%)            | 60.960 ± 1.34       |                      |
| 2  | Acid (g/l)              | 0.637 ± 0.08        | 4.916 ± 0.03         |
| 3  | pH                      | 4.510 ± 0.01        | 2.076 ± 0.021        |
| 4  | Protein (%)             | 3.59 ± 0.19         | 26.475 ± 0.643       |
| 5  | Brix degree (%)         | 2                   | 8                    |
| 6  | Vitamin C (mg/g)        | 5.787 ± 1.492       | 8.392 ± 0.288        |
| 7  | Lipid (%)               | 2.93 ± 0.044        | 0.542 ± 0.051        |
| 8  | Carbohydrate (%)        | 30.74               | 64.4                 |
| 9  | Ash (%)                 | 1.780 ± 0.004       | 0.314 ± 0.002        |
| 10 | Total fiber (%)         | 4.78                | 3.35                 |
| 11 | Reducing sugar (mg/ml)  | 0.633 ± 0.058       | 1.866 ± 0.305        |

For lipid content, the results showed that Lemon juice and lemon peel solution both contained relatively low lipid content (<5%). The lipid content in lemon peel (2.93%) is higher than that in lemon juice (0.5%). The lipid content in lemon juice is 0.5%, which is higher when compared with the result of Mohanapriya et al (2013), at 0.3% [31]. The lipid content in lemon peel is lower than that of Janati et al. (2012) with 4.98% [32]. In theory, the difference in lipid content of different citrus varieties allows different species to be distinguished, and their fatty acid profiles have been proposed as a chemical marker to identify the source. The base of the cultivar has been bred. Therefore, by quantifying the lipid content, this will be an initial step in the classification and identification of the varieties of lemons grown in Hau Giang province, Vietnam in the future.

The protein content of lemon peel (3.59%) was lower than that of Janati et al. (2012) where the protein of lemon peel was found to be around 9.42% [32]. The protein content measured in lemon juice (26.47%) was lower than that of Klavons et al. (1985) with a result of 29.8% [33]. The role of protein in the organoleptic properties of foods, apart from a source of amino acid, is accentuated in a previous study [1]. Lemon juice has about 7.37 times higher protein content in comparison with that in the peel. Thus, as compared to other varieties tested in previous studies, the present Vietnamese variety of lemon has a lower content of protein. Meanwhile, the total ash is a fraction consisting of mineral salts left over after heating the sample at high temperature (600 °C). The ash content of lemon peel (1.780%) was lower than that published by Mohammed et al. (2013), at 2.04%, [28] and is consistent with the range from 1.7 - 3.5%, which was described for different citrus species [34]. The ash content in the juice is very low (0.31%). Carbohydrates or glucides are organic compounds consisting of monosaccharides, their derivatives or condensation products. Carbohydrates are the remaining organic matter after subtracting moisture, ash, protein and fat. The carbohydrate content in peel and lemon juice are 30.74% and 64.43%, respectively.

Since health benefits of crude fiber are mostly related to peristaltic movement of the intestinal tract, the low consumption or absence of fiber may be associated with constipation risk and thus colon diseases.
(piles, cancer and appendicitis) [1]. Fiber also promotes bacteria growth in the rumen, in turn playing important role in ruminant digestion [35]. Fiber consists of carbohydrate molecules (monosaccharides or polysaccharides) and has two main components: soluble fiber (SF) and insoluble fiber (IF). Total fiber (TDF) is the sum of SF and IF. TDF in lemon juice (3.35%) was higher than that of Mohanapriya et al. (2013) with a result of 2.8% [36]. In lemon peel, the total fiber obtained was 4.78%, which is much lower than the figure found in Janati et al. (2012), at 15.18% [32]. The results showed that the crude fiber content in lemon peel had higher value than the two grains, maize and soybean oilcake, with values of 2.7 and 3.3, and 7%, respectively [36, 37]. Therefore, with the value of protein reported in this study, lemon peel may be suitable for use as animal feed.

The final ingredient in Persian lemon, “reducing sugar”, is the sugar containing aldehyde group (-CHO) or ketone (-CO) such as glucose, fructose, arabinose, maltose, and lactose. The analysis results show that the content of reducing sugar in lemon juice is 1.7 times higher than that of lemon peel.

4. Conclusion
The benefits of plants and herbal medicines are widely recognized for many years. This study presented a compositional analysis on Seedless Lemon (Citrus latifolia) Grown in Hau Giang Province, Vietnam, an abundant source of plant potentially applicable in many fields. It was found that Persian lime have a good content of vitamin C, crude fiber, lipid and other components. Therefore, this is the basis to confirm that Persian Lime has the potential to be exploited for use in food, medicine as well as in cosmetics.

Acknowledgment:
This work was supported by grants from Science and Technology Department of Hau Giang Province, Viet Nam.

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