Effect of Processing on Specific Phenolic Compounds of Two Market Peanuts Grown in Mexico: Possible Health Implications

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Abstract

Peanut (Arachis hypogaea L.) is a legume of high consumption worldwide. It is rich in protein and healthy fatty acids, but also contains a series of health-promoting phytochemicals, highlighting the phenolic compounds. Peanuts are not eaten raw, but first go through different treatments. This research provides information on how treatments such as roasting, frying, microwaving and germination affect the phenolic profile of peanuts and, consequently, their nutraceutical potential. In general, the treatments increased the content of gallic acid by 12-190%, p-coumaric acid (53 -197%), rutin (33 -46%), catechin (11 -301%), and epicatechin (147-841%) in Virginia variety; while in the Valencia variety increases in p-coumaric acid between 70-244%, rutin (17-68%), catechin (68-361%), and epicatechin (21-327%) were observed. Ferulic and caffeic acids were not detected. There was no effect of any of the treatments on quercetin content. Germination was the best method to increase these nutraceutical compounds. This method also induced the synthesis of resveratrol at levels higher than grapes and wine.

Keywords

Arachis hypogaea, phenolic profile, germination, roasting, microwaving

1. Introduction

The food industry has focused its attention on the development of foods that, in addition to supplying the basic nutrients for the maintenance of the organism, offer an additional benefit to health, of particular interest being those that provide antioxidant compounds [1, 2]. Peanut (Arachis hypogaea L.) is a highly consumed legume throughout the world. It is rich in protein, fiber and healthy fatty acids, but it also contains a series of beneficial phytochemicals for health, highlighting the phenolic compounds [3]. These compounds have been shown to have anti-inflammatory, cardioprotective, anticancer, antidiabetic and neuroprotective properties, among others, due to their antioxidant capacity, which prevents the damage to proteins, lipids and DNA caused by oxidative stress [4]. In addition, polyphenols exert other activities such as modulators of gene expression and cell signaling as well as the regulation of enzymatic activity [5].

Peanuts are consumed as processed foods, either directly in the shell or in the form of peanut butter, snacks and candies. Irrespective of the form of consumption, peanuts undergo a prior heat treatment, generally roasting in a conventional oven or roasting in oil, in order to reduce the microbial load, facilitate peeling, improve sensory characteristics and reduce anti-physiological factors [6]. Currently, microwave heating is also being evaluated because this method is fast, saves energy and is easy to control [7, 8]. On the other hand, it has been shown in different seeds that germination improves protein digestibility, decreases antiphysiological factors, and increases bioactive compounds, including phe-
nolic compounds [9-12].

The objective of this work was to evaluate the effect of processes such as roasting, frying, microwaves and germination on selected phenolic compounds of two commercial types of peanuts (Valencia and Virginia) grown in Mexico.

2. Materials and methods
2.1 Materials

Peanuts (*Arachis hypogaea* L.) of Valencia and Virginia varieties, obtained from different localities in Mexico, were used for this research. The Valencia peanuts were grown in the municipality of Temoc, Morelos (annual mean temperature of 19.8°C, average annual precipitation of 1,693 mm). Virginia peanuts were grown in Delicias, Chihuahua (annual average temperature of 27.7°C, mean annual precipitation of 334.2 mm). Whole pods were carefully chosen and shelled to obtain the grains. The defective grains were removed and the different treatments were applied to the healthy grains.

Pure standards of gallic acid, quercetin, p-coumaric acid, ferulic acid, caffeic acid, catechin, epicatechin, rutin, and resveratrol were purchased from Sigma-Aldrich (St. Louis, MO, USA).

2.2 Treatments

Peanuts were exposed to different types of processing (roasting, frying, microwave and germination) to consequently evaluate their effect on the content of selected phenolic compounds. Untreated dry peanuts were used as a control. Peanuts were dry roasted in a preheated convection oven at 175°C for 15 min. Another batch of peanuts was fried for 2.5 min at a ratio of 50 g of seeds in 200 mL of high oleic safflower oil preheated to 175°C. For the microwaving treatment, peanuts were heated in a microwave oven (Panasonic, model NN-6462A, Secaucus, NJ, USA), using a frequency of 2.45 GHz and a power of 450 W, for 3.5 min. After each thermal treatment, peanuts were cooled, the skin removed and the skinless seed ground.

For the germination process, peanuts were washed and disinfected by immersing them in a chlorine dioxide solution (0.25 ml of a 10% solution for each L of water) for 10 minutes. Then, they were soaked for 16 h in water at room temperature (23-25°C), drained and placed in a plastic tray with a perforated lid and on a cotton bed covered with filter paper. After three days of germination, the sprouts were harvested, oven-dried at 50°C, and pulverized. Both varieties of peanuts showed high biological activity, with germination percentages above 93%.

2.3 Antioxidants extraction

Ground samples were extracted using 80% methanol at a solid-to-solvent ratio of 1:10 (w/v) for 8 h by magnetic stirring. The extracts were then obtained by filtration through Whatman No. 4 filter paper and stored at -20°C until analysis.

2.4 HPLC analysis

An Agilent 1200 liquid chromatograph (Santa Clara, CA, USA) equipped with a UV-Vis detector and a reversed phase C18 column (250 mm x 4.6 mm, 5 µm particle size) was used. The elution gradient consisted of a mobile phase A (water-acetonitrile-acetic in ratios 93: 5: 2 v/v/v) and a mobile phase B (water-acetonitrile-acetic 58: 40: 2) from 0 to 100% B. The flow rate was 1 mL/min, the injection volume was 20 µL (extracts at a concentration of 5 mg/mL in methanol, previously filtered through a 0.45 µm membrane), and the run time was 50 min. The absorbance was read at 280, 320 and 350 nm [13]. Calibration curves of the pure standards at different concentrations were used for the quantification of sample constituents.

2.5 Statistical analyses

Values are expressed as the mean ± SD of three replicates for all determinations. The data were analyzed by one-way ANOVA and the Tukey post-hoc test. All statistical analyses were done using SigmaPlot 13 (Systat Software Inc., San José, CA, USA). Differences were considered statistically significant if *p* < 0.05.

3. Results and discussion

Table 1 shows the amount of the individual phenolic compounds identified in each sample. Figure 1 shows an example of the chromatograms obtained at the different wavelengths. The content of caffeic acid, ferulic acid, gallic acid, resveratrol, p-coumaric acid, catechin, epicatechin, rutin, and quercetin was determined based on what was found by different researchers in different varieties of peanuts [13-15]. However, caffeic acid and ferulic acid were not detected in any of the samples analyzed in this study.
<table>
<thead>
<tr>
<th>Compounds</th>
<th>Valencia</th>
<th>Virginia</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Raw</td>
<td>Dry-roasted</td>
</tr>
<tr>
<td>Gallic acid</td>
<td>50.27±3.23b</td>
<td>49.83±1.51b</td>
</tr>
<tr>
<td>p-Coumaric acid</td>
<td>34.88±1.33a</td>
<td>59.45±4.76b</td>
</tr>
<tr>
<td>Resveratrol</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Quercetin</td>
<td>84.55±2.19a</td>
<td>84.81±1.13a</td>
</tr>
<tr>
<td>Rutin</td>
<td>39.78±0.89a</td>
<td>46.47±1.44a</td>
</tr>
<tr>
<td>Catechin</td>
<td>20.06±0.56a</td>
<td>38.32±0.99c</td>
</tr>
<tr>
<td>Epicatechin</td>
<td>300.73±12.33a</td>
<td>682.66±8.45d</td>
</tr>
</tbody>
</table>

Notes: Values are mean ± standard deviation. Different letters in the same row mean significant differences among treatments for each type of peanut (p < 0.05). ND: not detected.

Figure 1. Chromatograms of selected phenolic compounds determined by HPLC in peanut extracts. (A) gallic acid (1), catechin (2), and p-coumaric acid (3) at 280 nm; (B) epicatechin (4) and resveratrol (5), at 320 nm; (C) rutin (6) and quercetin (7) at 350 nm.
In general, there was an increase in the content of the different phenolic compounds with the thermal treatments. In raw peanuts, these compounds can be linked to cell wall components and proteins through ester bonds. Some of them are found as glycosides, where the aglycone is bound to one or more sugars, or joined together to form complex compounds. It has been shown that heat treatment can release these compounds from the matrix to which they are bound, causing an increase in the free forms in the cotyledons [6, 15]. The release of phenolic compounds could improve their bioavailability and, as a consequence, their health benefits [16]. On the other hand, the treatments were applied to seeds with testa. Flavonoids such as catechin and epicatechin, derived from the condensed tannins of the testa, may have migrated to the cotyledons. This probably happened in view of the notable increase in these compounds after the heat treatments. In the Virginia variety, catechin increased by 72, 43 and 11% with oil roasting, dry roasting and microwave, respectively, while the Valencia variety had increases of 91, 361 and 68% with the respective treatments. On the other hand, in the Virginia variety, epicatechin increased by 572, 841 and 147% with oil roasting, dry roasting and microwave, respectively, whereas in the Valencia variety, this flavonoid increased by 127, 123 and 21%, respectively.

There were no appreciable changes in the content of quercetin (a flavonoid detected in other studies with peanuts) after treatments, although there was a discrete increase in the content of rutin, which is the quercetin rhamnoside. In contrast, Win [17] found that roasting at 160°C increased the concentration of quercetin from 104.46 to 134.41 μg/g after 30 min of treatment in peanuts of the Virginia variety from Myanmar.

A notable increase in the content of p-coumaric acid was also observed with thermal treatments. In the Virginia variety, this compound increased by 229, 174 and 53% with oil roasting, dry roasting and microwave, respectively. Craft [13] obtained similar results in samples from Virginia, Spanish and Runner varieties. In their study, the free p-coumaric acid concentration augmented from raw to dry-roasted and oil-roasted samples with a concomitant decrease in p-coumaric acid derivatives, suggesting that the heat treatments caused the liberation of phenolic acids from their parent derivatives. They found increases in free p-coumaric acid of up to 393% for roasting and 785% for frying in high-oleic Runner peanuts. Win [17] found that p-coumaric and quercetin were the phenolics that roasting increased the most under conditions of 160°C for 30-50 min.

On the other hand, germination increased the concentration of gallic acid by 234% and 191%, p-coumaric acid by 244% and 197%, rutin by 68 and 46%, catechin by 208% and 307%, and epicatechin by 297% and 806% in the Valencia and Virginia varieties, respectively. Yang [12] also found that germination increased the content of polyphenols and flavonoids and induced significant changes in the polyphenolic profile of peanuts.

It is interesting to note that resveratrol was not found in the untreated samples but was synthesized during germination, reaching a content of 19.92 μg/g in the Valencia variety (see Figure 2). This quantity is higher than that found in red grapes and red wine (maximum 8.69 and 15.35 μg/g, respectively) [18]. Wang [19] found that resveratrol concentration increased from 2.3-4.5 ppm in raw samples to 11.7-25.7 ppm after 9 days of germination, similarly to those found in this work after three days of germination. Limmongkon [20] found that the change in resveratrol concentration during germination depended on the peanut cultivar. The time in which the maximum concentration of resveratrol was reached varied from cultivar to cultivar. Likewise, Adhikari [21] found that resveratrol, total polyphenols, and flavonoid contents, as well as the antioxidant activity of peanut sprouts, significantly varied with the genotype of seeds, part of the sprout and time of germination; thus, it is important to establish the best germination conditions for maximum benefit.

Resveratrol is a phenolic compound belonging to the group of stilbenes. It was first isolated from peanuts, although it became popular as the main antioxidant in red wine [15, 19]. Its cardioprotective, antioxidant and anticarcinogenic properties are well documented [18]. In the chromatograms of Figure 2, a notable increase in the content of epicatechin and the appearance of a peak corresponding to resveratrol are observed after germinating peanuts of the Valencia variety.

Sprouts are rich in phytochemicals, which are synthesized by plants during germination as a defense mechanism against depredators, ultraviolet radiation, and other stress situations. Furthermore, the enzymes synthesized during germination could release phenolic compounds from the food matrix [22, 23].

The structure of phytochemicals influences their bioaccessibility and bioavailability and, as a consequence, their metabolic fate. For example, after ingestion, quercetin is absorbed in the upper gastrointestinal tract, whereas rutin remains intact until the distal tract [15, 16]. According to a review of 97 studies about the bioavailability of polyphenols in humans, proanthocyanidins are among the least-well-absorbed, whereas catechins are among the most well-absorbed [16]. This is important in view of the notable increase in catechin and epicatechin observed in the peanuts subjected to the different treatments.

Epicatechin has diverse biological properties such as antioxidant, anti-inflammatory, antimicrobial, antitumor, anti-diabetic, cardioprotective and neuroprotective activities [26, 27]. Besides, epicatechin improved the fatigue resistance and oxidative capacity of muscles in a mouse model, by increasing the number of capillaries and mitochondria in the muscle [28]. It has also been observed that epicatechin and quercetin act synergistically, improving the mitochondrial performance of neurons and conferring considerable protection against ischemic damage [29]. Regarding gallic acid, several beneficial effects have been reported, including antioxidant, anti-inflammatory and antineoplastic properties with therapeutic activity against gastrointestinal, neurodegenerative, metabolic and cardiovascular disorders [30].
4. Conclusion

In this study, the response to the different treatments in the phenolic profile of peanuts grown in Mexico depended on the variety of peanut. However, there were increments in the content of phenolic compounds with demonstrated beneficial effects on health, such as gallic and p-coumaric acids, catechin, epicatechin, and resveratrol. Germination was the best method to increase these nutraceutical compounds.

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References


