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Physicochemical composition and antioxidant capacity of zucchini and green beans before and after steaming and boiling

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ABSTRACT

Different cooking methods can be used to prepare vegetables, therefore, understanding the effect of these techniques on food composition is necessary. The aim of this study was to determine the effect of cooking on the physicochemical composition of zucchini and green beans. Three treatments were considered: raw, steamed and boiled foods. For each treatment, the experiment was performed with six replicates and each sample unit was analyzed in triplicate. The analyses performed were: cooking factor, soluble solids, pH, moisture, ash, phenolic compounds and antioxidant capacity. Significant data of the treatments were analyzed by Tukey's test. The foods showed cooking factor equals to one. Both boiled zucchini and green beans showed the lowest contents of soluble solids, ash and phenolics. The moisture content of both vegetables decreased after cooking. The steamed foods presented the highest antioxidant capacity. Considering pH, the boiled zucchini presented the lowest level and both thermal processes increased the pH of the green beans. We concluded that steaming preserves or improves the composition of the foods analyzed in the present study.

Keywords: *Cucurbita moschata*, *Phaseolus vulgaris* L., vegetables, cooking, phytochemical compounds, antioxidants.

RESUMO

Composição físico-química e capacidade antioxidante de abobrinha e vagem antes e após cocção a vapor e por ebulição

Diferentes tipos de cocção podem ser empregados no preparo das hortaliças, portanto, compreender o efeito dessas técnicas na composição dos alimentos se faz necessário. O objetivo do trabalho foi obter o efeito do cozimento na composição físico-química de abobrinha e vagem. Foram considerados três tratamentos, sendo os alimentos crus, cozidos a vapor e por ebulição. Para cada tratamento, o experimento foi executado com seis repetições e cada unidade amostral analisada em triplicata. As análises realizadas foram: fator de cocção, sólidos solúveis, pH, umidade, cinzas, compostos fenólicos e capacidade antioxidante. A significância dos tratamentos foi avaliada pelo teste de Tukey. Os alimentos apresentaram fator de cocção igual a um. Ambos os vegetais cozidos por ebulição obtiveram os menores teores de sólidos solúveis, cinzas e fenólicos. A umidade na abobrinha e vagem reduziu após as cocções. Os alimentos cozidos a vapor apresentaram maior capacidade antioxidante. Em relação ao pH, a abobrinha cozida por ebulição obteve o menor teor e ambos os processamentos térmicos aumentaram o pH da vagem. Conclui-se que a cocção a vapor preserva ou melhora a composição dos alimentos analisados.

Palavras-chave: *Cucurbita moschata*, *Phaseolus vulgaris* L., verduras, culinária, compostos fitoquímicos, antioxidantes.

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Vegetables are eatable plants which show natural resources of several biological active composts, such as vitamins, minerals and antioxidants, like phenolic compounds, tocopherols and carotenoids (Cardoso *et al*. 2018; Adegbaju *et al*., 2020). The presence of these components, in a synergistic way, is fundamental for the development, reproduction and defense of plants, due to their antimicrobial and antidegradant effect (Garcia *et al*., 2019). Also, these effects are related to the consumer's health, as they can have anti-inflammatory and antioxidant action, being able to inhibit or delay damage caused by free radicals, in addition to participating in essential metabolic processes (Vizzotto *et al*., 2017).

Regular consumption of these vegetables contributes to increased longevity and quality of life, by reducing damage associated with degenerative diseases and aging, reducing the risk of developing and progressing cardiovascular disease, type 2 diabetes, some types of cancer and obesity (Locke *et al*., 2018; Basiak-Rasala *et al*., 2019; Korus, 2020).

Nevertheless, in order to enable the digestive process and maximize sensory traits, these vegetables are usually consumed after being heated, using thermal processes according to the consumer's preference, whether using moist, dry or mixed heat, which may result in changes in the physicochemical composition of these foods (Rana *et al*., 2021).

The application of moist heat, despite contributing to the inactivation of antinutritional factors, formation of aromas and increased palatability of the food, even for short periods, can promote a change in the content of bioactive compounds, besides resulting in a reduction of soluble compounds, such as sugars and dietary fiber, by degradation or solubilization in an aqueous environment (Çubukçu *et al*., 2019; Arias-Rico *et al*., 2020; Rana *et al*., 2021). This heat application can also promote changes in hydrogen potential (pH) of the food, considering that this is an important parameter for the development of sensory traits and microbiological control of vegetables (Cardoso *et al*., 2018; Sousa *et al*., 2020).

In this context, the authors highlighted the importance of studies on the effects of moist heat in food preparation, considering physicochemical composition of vegetables, so that the results will allow for more guided and conscious choices regarding the best preparation methods to be used. However, the aim of this study was to evaluate the effects of moist heat on food preparation, considering physicochemical composition and antioxidant capacity of zucchini and green beans.

MATERIAL AND METHODS

Raw materials: acquisition and pre-preparation

The analyses were carried out from October 2022 to September 2023. Conventionally grown zucchini, Caipira group, (*Cucurbita moschata*) and string beans (*Phaseolus vulgaris* L.), were acquired, all in one lot, at the local retail market in Goiânia-GO. The vegetables were sanitized in running water to remove surface impurities.

Before preparing the vegetables, the authors standardized the cuts to be used: sliced zucchini and whole green beans, having only both ends removed with the aid of a stainless steel knife, portioned into 40 g fractions, with the aid of a semi-analytical balance (*Shimadzu* UX6200H).

Food preparation

Vegetables were prepared considering the regular ways of consumption (steaming and boiling). The methodology used was adapted from Brandão & Souza (2020) (Table 1).

Table 1. Conditions used in preparing vegetables. Goiânia, UFG, 2023.

Foods	Cooking methods	Time	Temperature
Zucchini (40 g) (slices)	Steaming	10 minutes	97°C
	Boiling	10 minutes	97°C
Green beans (40 g)	Steaming	15 minutes	97°C
	Boiling	20 minutes	96°C

Source: adapted from Brandão & Souza (2020).

In order to cook the vegetables, we used 1 L of water. Steamed and boiled vegetables were added when the water reached stipulated temperature (Table 1). The temperature was measured in the center of the pan using a digital infrared thermometer (*Instrutherm* TI-860). For steaming the vegetables, the authors used a stainless steel steamer with lid (Brandão & Souza, 2020).

Sample preparation and experimental design

After preparation, the vegetables were macerated using a porcelain mortar and pestle and, afterwards, they were analyzed.

The authors used a completely randomized experimental design. For each treatment (raw, steamed and boiled), the experiment was carried out with six replicates and each sample unit was evaluated in triplicate. The results of the physicochemical composition and antioxidant capacity were expressed using the mean of the eighteen determined values $\pm$ DP (standard deviation).

Cooking factor (FC)

Cooking factor was obtained by weighing the vegetables before and after being submitted to thermal procedures. The formula used was Cooking factor = weight of cooked food / weight of raw food. Calculus was performed using Excel software, version 2019.

Each treatment was carried out with six replicates, thus, the values of FC were obtained by the average of replicates ± DP. As in the other analyses, the Completely Randomized Design (CRD) was used with the experimental units.

Physicochemical composition

Total soluble solids (TSS) and hydrogen potential (pH) were determined using the methods described by IAL (2008). In order to evaluate TSS, we used a bench refractometer. In this analysis, the macerated vegetables were sieved in order to obtain the liquid part. Then, the liquids were transferred to Eppendorf microtubes and centrifuged (5000 rpm / 2 min.) in a refrigerated centrifuge. After, 3 droplets of supernatant liquid were placed on the refractometer prism and the reading taken. We measured the pH value with the aid of a pH meter with temperature compensator.

To determine moisture, the samples were dried to constant weight in a drying and sterilization oven programmed at 105°C. For the ash analysis, the crucibles with the samples were carbonized in a stove; after carbonization, the samples were calcined in an automatic muffle furnace at 550°C for 5 hours (IAL, 2008).

To obtain extracts, with the aid of greaseproof paper, 2 g of the samples were weighed on an analytical balance and then transferred to falcon-type tubes and 10 mL of methanol were added. The tubes, in the absence of light, remained for one hour in an ultrasonic bath. Afterwards, the extracts were centrifuged (4000 rpm / 15 min.) in a refrigerated centrifuge and filtered with the aid of filter paper.

The total phenolic content was determined spectrophotometrically using Folin-Ciocalteu solution (Singleton & Rossi, 1965; Genovese *et al.*, 2008). Absorbance readings were performed at a wavelength of 765 nm. Using the obtained values, the concentration of total phenolics was determined by interposing values from a previously constructed gallic acid standard curve. The results were expressed in mg GAE (equivalent to gallic acid) / 100 g of food.

***In vitro* antioxidant capacity**

Antioxidant capacity was determined using spectrophotometer method through the DPPH radical (*2,2-Diphenyl-1-picryl-hidrazil*), described by Brand-Williams *et al*. (1995); Sanchez-Moreno *et al*. (1998).

This method is based on the DPPH radical capture by antioxidants, producing a decrease in absorbance at a wavelength of 517 nm. Using a standard curve previously constructed using trolox solutions (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), the results were expressed in µmol TEAC (antioxidant capacity equivalent to trolox) / 100 g of food.

Statistical analysis

Statistical analyses were performed using SYSTAT software (version 12, SYSTAT Corporation). The data were evaluated using the one-factor analyses of variance (ANOVA). The independent variable was the cooking method (raw, steaming, boiling) and the dependent variables were pH, total soluble solids (TSS), moisture, ashes, total phenolic compounds and antioxidant capacity. The significance of the independent variables was subsequently evaluated in pairwise comparisons using the Tukey test. For all the ANOVAs, the authors evaluated assumptions of independence and normality by scatter plots and histograms of residuals, respectively.

RESULTS AND DISCUSSION

Cooking factor (FC)

Evaluating the cooking factors of zucchini and green beans, for both cooking methods, the authors verified results close to 1 (Table 2), as observed in other studies which evaluated the same vegetables (Santos & Basso, 2019; Brandão & Souza, 2020).

Cooking factor equal or close to 1 shows the maintenance or a slight weight loss of the food after thermal processing (Pereira, 2017). This result was possible since moist heat cooking methods were used. These methods tend to concentrate extractive substances and hydrate food. Freitas *et al.* (2021) reported that also the chemical composition of these vegetables may have favored to maintain the weight.

Table 2. Vegetable cooking factors. Goiânia, UFG, 2023.

Foods	**Cooking methods**	
	Steam (FC) (average ± DP*)	**Boiling (FC)** (average ± DP)
Zucchini	0.96 ± 0.01	0.97 ± 0.02
Green beans	0.97 ± 0.0	1.01 ± 0.02

*DP: standard deviation.

For zucchini and green beans, which are tender vegetables, the contact of carbohydrates with steaming or boiling water can promote superficial coagulation of the food, sealing the surface of the vegetable, reducing substance losses to the cooking medium.

Total soluble solids

TSS contents were statistically similar in raw (5.2°Brix) and in steamed zucchini (5.0°Brix) (Table 3).

Table 3. Physicochemical composition and antioxidant capacity of vegetables before and after cooking. Goiânia, UFG, 2023. Goiânia, UFG, 2023.

Analyses	**Zucchini**			**Green Beans**		
	Raw	**Steamed**	**Boiled**	**Raw**	**Steamed**	**Boiled**
	Average ± *DP			**Average ± DP**		
Total soluble solids (°Brix)	5.2±0.0^{a}	5.0 ± 0.3^{a}	3.6 ± 0.4^{b}	5.6 ± 0.3^{b}	6.0 ± 0.3^{a}	5.33 ± 0.2^{c}
pH	6.1 ± 0.1^{a}	5.8 ± 0.3^{a}	5.3 ± 0.7^{b}	5.5 ± 0.2^{c}	6.1 ± 0.1^{a}	5.91 ± 0.2^{b}
Moisture (%)	95.1 ± 1.2^{a}	94.1 ± 2^{b}	93.7 ± 0.6^{c}	93.2 ± 0.5^{a}	91.5 ± 0.9^{c}	92.5 ± 0.5^{b}
Ash, wet basis (%)	0.39 ± 0.1	0.44 ± 0.1	0.4 ± 0	0.78 ± 0	0.77 ± 0.1	0.65 ± 0
Ash, dry basis (%)	8.3 ± 2.4^{a}	7.8 ± 1.8^{b}	6.5 ± 1^{c}	11.5 ± 0.9^{a}	9.1 ± 0.8^{b}	8.7 ± 0.7^{c}
Total phenolics (mg GAE**/100 g)	16.5 ± 5.2^{a}	17.2 ± 2.0^{a}	11.15 ± 1.3^{b}	23.7 ± 1.3^{a}	22.3 ± 2.2^{b}	18.4 ± 0.9^{c}
Antioxidant capacity (µmol TEAC***/100 g)	54.8 ± 5.1^{b}	67.7 ± 7.6^{a}	59.35 ± 7.8^{b}	118.2 ± 7.6^{b}	128.2 ± 10.9^{a}	108.78 ± 4.3^{c}

TSS, pH, total phenolics and antioxidant capacity = Different superscript letters between the preparation methods, considering the same food, indicate statistically significant difference ($p<0,01$). Moisture and ash = Different superscript letters between the preparation methods, considering the same food, a statistically significant difference was noticed ($p<0,05$). *DP: standard deviation. **GAE: Gallic acid equivalent. ***TEAC: Antioxidant capacity equivalent totrolox.

However, evaluating green beans, the authors observed significant statistical difference in TSS content according to the cooking method used. We observed that steamed green beans showed the highest TSS content (6.0°Brix), followed by raw green beans (5.6°Brix) and boiled green beans (5.33°Brix), respectively (Table 3). Therefore, according to this study, we can conclude that steaming preserves or increases TSS content in these evaluated vegetables whereas boiling reduces.

Zor*et al*. (2022) also reported lower TSS contents in vegetables boiled under atmospheric pressure, such as potatoes, carrots, black carrots, beets, sweet-potatoes, celery and turnip. Armesto *et al*. (2017) also observed that boiling provided a decrease from 52.5 to 55.2% in contents in kale. These losses are related to thermal degradation and diffusion of water-soluble compounds during heating. In this sense, the methods which do not use water or use only a little quantity of water for thermal procedures show fewer TSS losses, since most losses occur through the diffusion of soluble compounds in water (Diamante *et al.*, 2019).

Considering the increase in TSS content verified in steamed green beans, Reis *et al*. (2015) also identified an increase when evaluating broccoli and cauliflower submitted to different cooking methods (boiling, steaming, microwave cooking and *sous vide*). According to these authors, this result may be related to the plant tissue destruction by the cooking process, which makes these compounds more available.

Hydrogen potential (pH)

Raw (6.1) and steamed zucchini (5.8) obtained similar pH and significantly higher content than boiled zucchini (5.3) (Table 3). This result may have happened due to the release of organic acids from the food matrix or the formation of new compounds during thermal treatment (Chiavaro *et al*., 2012; Armesto *et al.*, 2017).

On the other hand, thermal processing of green beans promoted an increase in pH in both cooking methods, with values of 5.5, 6.1 and 5.91 in raw, steamed and boiled green beans, respectively (Table 3). Therefore, in green beans, cooking methods increased the pH. This result may have been caused by damage to the cells during cooking, which can cause the release of organic acids and the destruction of some thermal unstable acids (Armesto *et al.*, 2017). The degradation of organic acids during cooking induces the proton release (Arkoub-Djermoune *et al.*, 2016).

Given the above, pH of the vegetable showed different behavior in boiling. According to Diamante *et al.* (2019), different effects of cooking can occur due to the differences in the food matrix and in the chemical properties of the compounds.

Moisture and ashes

The authors verified that moisture content in zucchini and green beans decreased significantly after thermal procedures: greater decrease when boiling, in zucchini, and steaming, in green beans (Table 3). Paula *et al.* (2019) also observed a reduction in moisture content in Cabotiá pumpkin considering the three cooking methods evaluated in this study (steaming, microwave cooking and airfryer); however, these authors highlighted that in the airfryer the reduction is greater, since the food is dehydrated during the process.

Diamante *et al.* (2019), evaluating physicochemical characteristics of cauliflower *in natura* and after thermal procedures, verified, in general, a moisture loss of inflorescences when submitted to steaming and microwave cooking; the boiled samples showed a slight increase, though. This result was similar to the one obtained by Llorent-Martínez *et al.* (2020) with broccoli before (87.2%) and after steaming (85.5%) and boiling (92.1%). Cardoso *et al*. (2018) observed that the increase in moisture content in broccoli was higher in boiling and under pressure cooking when compared with microwave cooking.

Armesto *et al*. (2016), studying the effect of different cooking processes on the quality parameters of kale, observed that all cooking methods used (boiling, steaming, under pressure and microwave cooking) increased significantly the moisture content in the vegetable. Evaluating the effect of cooking methods on Chinese cabbage, Chin *et al*. (2022) observed an increase in moisture content of the samples in all methods evaluated (boiling and steaming), which can be justified by the fact that heat is responsible for breaking the middle lamella of plant cell, leading to water absorption, and consequently starch gelatinization.

Given the above, the authors verified that almost all foods mentioned belong to the Brassicaceae family (cauliflower, broccoli and kale). This result shows that, mainly, boiling cooking method provides an increase in moisture content of brassicas, differing from the observed in zucchini and green beans. According to Mathlouthi (2001), in addition to the nature of the evaluated samples, the technique used and the loss of other volatile compounds may interfere with the divergence observed between the effect of thermal processing on different vegetables.

Evaluating the ash contents (dry base), the authors noticed a decrease in these contents after thermal processing, considering that the most significant reduction was noticed in boiled vegetables. Thus, this study reinforces the information that steaming is the most effective cooking method to retain fixed mineral residue content (Table 3). Mansour *et al*. (2015) found similar results to the ones in this study: all cooking methods (steaming, boiling and microwave cooking) reduced ash contents in broccoli and cauliflower, the highest reduction was verified in boiling, though.

In addition to ratifying the reduction of ash content in boiled vegetables, Monalisa *et al*. (2020), studying pumpkin, reported that as boiling time increases, ash contents decrease. The pumpkin which is boiled for 10-20 min shows 0.89 and 0.82% of ashes, respectively.

Corzo-Ríos *et al*. (2020) verified a reduction in ash content in nine varieties of autoclaved beans (120°C/15 lb/in^2). Garcia-Herrera *et al*. (2020) verified a reduction in ash content in all boiled wild vegetables. According to these authors, this is due to a high solubility of minerals in water. Moreover, Bernhardt & Schlich (2006) noticed a significant reduction in mineral content in boiled red bell pepper. According to the authors, these losses are not caused by the destruction of the mineral content, but by leaching into the cooking water. The justifications listed are consistent with the dissolving characteristic of moist heat-cooking methods.

Total phenolics

In relation to total phenolics content, the authors verified that the contents were similar in raw (16.5 mg GAE/100 g) and steamed zucchini (17.2 mg GAE/100 g). We observed a reduction in total phenolics content in boiled zucchini, though (11.15 mg GAE/100 g) (Table 3). Piepiórka-Stepuk *et al*. (2023) also reported significant losses of phenolics contents in pumpkins submitted to thermal processing, showing loss up to 55% of phenolics contents when boiling cooking. The decrease in these compounds can occur due to the fact that they are water soluble, so that higher loss rates can be observed in boiled foods.

For green beans, the authors verified that the raw vegetable obtained the highest total phenolic content (23.7 mg GAE/100 g), followed by steamed green beans (22.3 mg GAE/100 g) and boiled green beans (18.4 mg GAE/100 g) (Table 3). Kapusta-Duch *et al.* (2019), studying purple cauliflower, verified similar results related to a significant reduction in total phenolics compounds after any thermal treatment, showing losses of 16.9% in total phenolics compounds for steamed and 64.2% for immersion cooking cauliflower.

Given the above, the authors reinforce that boiling cooking stands out as the method which shows the highest reduction in total phenolic content for both vegetables. Silva *et al.* (2019), evaluating the effect of different cooking methods (steaming, boiling, microwave cooking and *sous vide*) in pumpkins, obtained similar results, verifying the lowest concentrations of total phenolics using the boiling method, with a reduction of 64.94% in total phenolic contents.

Despite damage to the plant's cell wall and softening of its pulp lead to the release of phenolic substances bound to the cell walls and insoluble fractions of the plant tissue, making them available for reaction with the Folin-Ciocalteu reagent, the time the vegetables remain in hot water can lead to the degradation of the compounds, in addition to facilitating the migration of water from the cell with the nutrients dissolved in it, causing losses through leaching process (Buratti *et al*., 2020; Chin *et al*., 2022; Piepiórka-Stepuk *et al*., 2023).

Antioxidant capacity

Steamed zucchini (67.7 µmol TEAC/100 g) showed higher antioxidant capacity when compared with raw and boiled zucchini (54.8 and 59.35 µmol TEAC/100 g, respectively) (Table 3).

Steamed green beans also showed higher antioxidant capacity (128.2 µmol TEAC/100 g) when compared with raw green beans (118.2 µmol TEAC/100 g). In line with the data of the present work, López-Hernández *et al*. (2022), studying broccoli, reported a greater antioxidant capacity of the food when it was steamed for 10 minutes, observing a 15% increase in content. The authors justified the results by the release of antioxidants, previously bounded to the cell wall, or even by the hydrolysis of esterified compounds and inactivation of oxidative enzymes. The same authors also observed a reduction in 60% of antioxidant capacity when the food was boiled for 10 minutes. This result is similar to the one presented in this study, since boiled green beans showed the lowest antioxidant capacity (108.78 µmol TEAC/100 g) (Table 3).

Guillén *et al.* (2017), studying the effect of cooking on carrots and artichokes, also verified a significant loss of antioxidant capacity for both boiled vegetables. This decrease in antioxidant capacity was of 83 and 90.8%, respectively.

Using FRAP method in African pumpkin (*Momordica balsamina*) and pumpkin leaves (*Cucurbita moschata*), Mashiane *et al*. (2021) verified losses up to 90.96% of the antioxidant content after cooking samples in boiling water. The authors reported that losses can be associated with different factors, like the instability of antioxidant compounds, such as vitamin C, which degrades when exposed to heat, and the loss of antioxidant compounds to the cooking water through leaching (Armesto *et al*., 2017; Mashiane *et al*., 2021).

Chin *et al*. (2022), evaluating the effect of cooking on boiled and steamed Chinese kale, noticed a significant increase in the food antioxidant capacity in both cooking methods. According to these authors, the use of heat can lead to denaturation of proteins and softening of the plant cell wall, providing an increase in the bioavailability of bioactive compounds that contributes to increasing antioxidant capacity. Thus, this information confirms that the effect of cooking method is related to peculiarities of food matrices, as well as processing conditions.

Given the above, we highlight that cooking vegetables shows benefits related to food consumption. According to the heat used and food preparation system chosen significant differences can be observed in food composition. We conclude that steaming preserves or improves the composition of the food, besides being a more effective method in maintaining phenolic compounds and increasing antioxidant capacity.

Contribution of each author to the paper

-Luiza M Gomes, Tâmera N V Almeida, Alana M R Freitas, Carolina B de Castro: The students were responsible for the acquisition, pre-preparation and preparation of food, laboratory analysis, interpretation and discussion of data, in addition to writing the paper. -Rodrigo B M Cavalcante: Professor responsible for the conception and writing of the project, guidance and training of students with the analysis methodologies used, interpretation and discussion of data and writing the paper.

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