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## Multivariate Analysis of the content of bioactive compounds in kale (*Brassica oleracea*)

### Análise multivariada do teor de compostos bioativos em couve (*Brassica oleracea*)

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#### RESUMO

Os antioxidantes estão associados ao retardo do envelhecimento, à prevenção do câncer e de doenças inflamatórias, porém, podem ser perdidas durante o processamento dos alimentos. Portanto, este trabalho buscou caracterizar quantitativamente o conteúdo de compostos bioativos em couve após diferentes tratamentos e explorar os dados gerados por meio da análise de componentes principais (PCA). As análises dos teores de fenólicos totais foram realizadas pelo método *Folin-Ciocalteu* e a atividade antioxidante pelos métodos DPPH e FRAP. Os gráficos dos escores mostraram que os fenólicos totais foram agrupados de acordo com a parte da couve e o solvente utilizado para a extração, enquanto o gráfico do carregamento mostrou a influência do tratamento térmico na extração. Conclui-se que a quantidade de fenólicos totais e a atividade antioxidante varia em função do solvente de extração, partes do vegetal e tratamento térmico e que isso favoreceu sua extração.

**Palavras-chave:** Atividade Antioxidante; *Brassica oleracea*; Análise multivariada; Fenólicos totais.

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#### ABSTRACT

Antioxidants are compounds associated with delayed aging and the prevention of cancer and inflammatory diseases. These substances can be lost during food processing. Therefore, this work sought to quantitatively characterize the content of bioactive compounds in kale after different treatments and to explore the data generated using principal component analysis (PCA). The analyzes of the total phenolic contents were performed by the *Folin-Ciocalteu* method and the antioxidant activity were performed by the DPPH and FRAP methods. The graphs of the scores showed that the total phenolics were grouped according to the part of the kale and the solvent used for the extraction, while the graph of the loading showed the influence of the heat treatment in the extraction. It is concluded that the amount of total phenolics contents and antioxidant activity varies depending on the extraction solvent, parts of the vegetable and heat treatment and that this favored its extraction.

**Keywords:** Antioxidant activity; *Brassica oleracea*; Multivariate analysis; Total phenolics.

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## INTRODUCTION

Vegetable consumption has increased due to its nutritional value and its therapeutic effects. The phytochemicals in these foods have antioxidant properties that may be related to delayed aging and prevention of cancer, chronic inflammatory diseases, heart and lung diseases and problems associated with aging (BRENNA; PAGLIARINI, 2011; ZHANG *et al.*, 2015).

Continuous production of free radicals during metabolic processes in the body is the cause of the development of antioxidant defense mechanisms to reduce damage to humans (EFFERTH; KOCH, 2011). Many foods contribute to these defense mechanisms. That is why it is important to know the antioxidant capacity of foods to help protect against free radicals (SEMA *et al.*, 2022).

Unlike animals which have an immune system for adaptation, defense and consequent evolution, vegetables have developed a chemical resistance biosynthesized through secondary metabolism. These substances produced by plants act on specific targets of the molecules of their predators and can perform antioxidant functions acting as therapeutics in humans, such as bioactive compounds such as phenolics (BALASUDRAM *et al.*, 2006).

Antioxidants are substances capable of inhibiting oxidation, as they have the ability to react with free radicals in the body or to chelate metal ions, preventing lipid peroxidation. Among antioxidants, non-enzymatic ones are important due to their beneficial action on the body metabolism, such as ascorbic acid, tocopherol, carotenoids and phenolic compounds (FABBRI; CROSBY, 2016). This set of compounds is divided into subgroups according to their chemical structure, such as flavonoids, phenolic acids, tannins, lignins and stilbens (ROCHA *et al.*, 2019; SASIDHARAN *et al.*, 2011).

Industrial or domestic processing can make food more attractive to the palate and increase its shelf life. However, such processing can alter the chemical composition of nutrients in food, compromising the nutritional quality of the final product. The thermal processing in vegetable foods most used at home is cooking in steam, in boiling water, in a conventional oven or microwave oven. Such processes, applied alone or associated with other processes, for example, mechanical, can lead to changes in the physical characteristics and chemical composition of foods with positive or negative effects (JIN *et al.*, 2017). In this context, vegetables are often consumed raw but there are situations

where cooking is necessary or even preferred, which may alter the antioxidant capacity of these vegetables.

Kale (*Brassica oleracea* Lin. Var. *Acephala*) belongs to the Brassicaceae family and is considered one of the vegetables most cultivated and consumed by all social strata of the population in Brazil. It is rich in minerals, vitamins, fibers and antioxidants, being indicated for therapeutic purposes in the treatment of anemia, ulcers, colic, skin diseases, in addition to stimulating good intestinal peristalsis (KO; CHEEIGH; CHING, 2014).

When it comes to the compound bioactive analysis, the choice of solvent for the extraction of these compounds is important, especially when seeking to obtain one or more substances of interest (HUSSON; LE; PAGÈS, 2017). With the trend of applying green chemistry, there is an increasing search for the use of methods and techniques called ecologically correct that seek to reduce the use of organic solvents (SHARAF; ILLMAN; KOWALSKI, 1986). Water has often been used as a solvent due to its ability, under certain conditions of pressure and temperature, to acquire properties similar to organic solvents and to be non-toxic (INSTITUTO ADOLFO LUTZ, 2005).

On the other hand, it is common among researchers to study several variables simultaneously, allowing the extraction of a much larger amount of information in a single sample. In order to optimize this type of analysis, the statistical model of multivariate methods appears. They consider the multivariate relationship of the investigated variables. Thus, some algorithms were developed in order to represent the largest possible amount of information contained in a set of analytical data. Among them, the ones that stand out the most are the principal component analysis (PCA) (EMBRAPA, 2006). When the set of variables is high, the PCA is able to extract latent information and provide graphic visualization of the entire data set, in order to provide a better understanding from the investigation of the presence or absence of common groupings between the samples (EMBRAPA, 2007).

Therefore, this study aimed to evaluate the effect of different thermal processing and the influence of the type of extraction solvent on the concentration of total phenolic compounds (TP) and antioxidant activity (AA) in different parts of the kale using multivariate analysis.

## INSTRUMENTATION

The kale samples were purchased at markets in Salvador city, Bahia State, Brazil, and forwarded to the Food Technology and Chemical Analysis Laboratory from the Life Sciences Department of the Bahia State University (UNEB), properly packed in a cool box, under refrigeration.

The vegetables were initially selected, cleaned in running water and sanitized with sodium hypochlorite solution to 10ppm/15 minutes. Then they were rinsed under running water.

In order to better evaluate the chemical composition of these vegetables in their different parts, the kale leaves were subdivided into three samples: only the leaf, only the stem and the whole kale (leaf and stem).

After cutting, all samples were subjected to different thermal processing: bleaching by immersion (100 °C / 3min), steam bleaching (3 min.), braised in boiling water in the proportion 1: 1/2 (vegetable: water) / 3 min. After processing, the samples were ground in a multiprocessor and then dehydrated in a forced air circulation oven at 45° C until a humidity of around 9% was obtained. To obtain the aqueous and methanolic extracts, 0.5g of each sample and 30 mL of the respective solvent (water and methanol) were mixed, with subsequent stirring (170 rpm / 20 min) at room temperature, followed by filtration. The procedure was repeated with the residue retained in the filter for better extraction of the compounds and the resulting filtrates were mixed and stored under refrigeration.

The analyzes of total phenolic compounds (TP) were performed using the spectrophotometric method developed by Folin-Ciocalteu using the standard curve of gallic acid and the data expressed in mg / 100g (BARBA *et al.*, 2016).

The analyzes of antioxidant activity (AA) of the samples were carried out by the different DPPH and FRAP methods, according to the technical reports of the EMBRAPA (MOHANKUMAR; UTHIRA; SU, 2018; OBENG *et al.*, 2020), using the dehydrated samples, in quintuplicate.

The determination of AA by the DPPH method was carried out from the reaction between the sample extract in different dilutions (2500ppm; 1250ppm; 1000ppm) with the 0.06mM DPPH solution. The capture of the free radical DPPH promotes changes in color with a decrease in absorbance, read in a spectrophotometer at 515 nm after 2 hours.

The results were presented by the IC<sub>50</sub> values, that is, the amount of antioxidant needed to reduce the initial concentration of DPPH by 50%, the results being expressed in IC<sub>50</sub> (OBENG *et al.*, 2020).

Through the iron reduction method, FRAP, AA was evaluated based on the capacity of the antioxidants present in the diluted sample at different concentrations (2500ppm; 1250ppm; 1000ppm) to reduce the ferric-tripyridyltriazine complex in a ferrous complex under acidic conditions. The reaction promotes a decrease in absorbance and the reading is performed on a spectrophotometer at 595nm after 30 minutes. The results were expressed in  $\mu\text{mol L}^{-1}$  ferrous sulfate/g sample (MOHANKUMAR; UTHIRA; SU, 2018).

Analyzes of TP and AA compounds were carried out on the methanolic and aqueous extracts of the kale samples (only the leaf, only the stalk and the whole kale) submitted to different heat treatments (bleaching by immersion, bleaching by steam and braising), being the of TP in quintuplicate, and those of AA in triplicate.

To better characterize the concentration of TP in the kale according to the dependent variables evaluated (parts of the kale, type of extraction solvent and heat treatment), Principal Component Analysis was used, which is an exploratory multivariate technique. It was processed with the covariance matrix of the original variables, obtaining from it the eigenvalues that built the eigenvectors. These are linear combinations of the original variables and are called main components. In addition, the results of TP and AA were also analyzed by the Tukey test at the level of 5% ( $p < 0.05$ ).

## RESULTS AND DISCUSSION

The average values (mg / 100g) of the total phenolic compounds in the whole kale, leaf and stalk, obtained in the aqueous and methanolic extracts for the Kale before (*in natura*) and after the thermal processing are shown in Table 1.

Considering the extraction solvent (aqueous and methanolic), it was observed that the medium interferes in the extraction of TP, justifying the affinity of the different compounds present in each sample for the studied extraction solvents. According to Barba *et al.* (2016), extraction with organic solvents is one of the most widespread techniques among researchers. This technique is called conventional and is applied to different matrices. However, new alternatives for the extraction of biocompounds have been

sought, such as the use of solvents that have less toxicity, such as extraction assisted by ultrasound, extraction assisted by microwave, pressurized liquids, supercritical fluids, among others (ESTEVE-TURRILLASA; PASTOR, 2016; KHAW *et al.*, 2017).

The results showed that the methanolic extraction of TP was maximum in all samples (whole kale, kale stalk and kale leaf), whether *in natura* and in all heat treatment conditions (SB, BI and B), except for whole kale that there was no significant variation ( $p < 0.05$ ) between methanolic and aqueous extraction in the braised sample (Table 1). In this case, it is suggested that the fact that the molecules of the phenolic compounds are amphipathic, foments that they bind to both the polar and non-polar molecules of the extraction solvents.

**Table 1** - Concentrations in mg / 100g (mean  $\pm$  standard deviation) of total phenolic compounds in whole kale, leaf and stalk, before and after processing, from aqueous and methanolic extracts.

Sample / Extract	<i>In natura</i>	Steam bleaching (SB)	Bleaching-immersion (BI)	Braised (B)
<b>Whole kale</b>				
Methanolic	74,43 $\pm$ 0,80 <sup>Aa</sup>	81,50 $\pm$ 0,58 <sup>Ab</sup>	73,76 $\pm$ 0,30 <sup>Aa</sup>	76,20 $\pm$ 0,71 <sup>Aa</sup>
Aqueous	29,13 $\pm$ 0,58 <sup>Ba</sup>	68,55 $\pm$ 0,83 <sup>Bb</sup>	68,51 $\pm$ 0,98 <sup>Bb</sup>	75,33 $\pm$ 0,59 <sup>Ab</sup>
<b>Leaf</b>				
Methanolic	182,52 $\pm$ 4,35 <sup>Aa</sup>	217,66 $\pm$ 1,46 <sup>Ab</sup>	145,39 $\pm$ 5,41 <sup>Ab</sup>	197,58 $\pm$ 0,67 <sup>Ab</sup>
Aqueous	125,43 $\pm$ 9,07 <sup>Ba</sup>	206,99 $\pm$ 1,84 <sup>Bb</sup>	134,23 $\pm$ 2,56 <sup>Bb</sup>	190,91 $\pm$ 0,88 <sup>Bb</sup>
<b>Stalk</b>				
Methanolic	77,32 $\pm$ 7,58 <sup>Aa</sup>	66,29 $\pm$ 6,88 <sup>Ab</sup>	68,96 $\pm$ 2,51 <sup>Ab</sup>	59,89 $\pm$ 0,14 <sup>Ab</sup>
Aqueous	22,66 $\pm$ 2,66 <sup>Ba</sup>	55,58 $\pm$ 1,70 <sup>Bb</sup>	47,24 $\pm$ 2,16 <sup>Bb</sup>	54,093 $\pm$ 1,65 <sup>Bb</sup>

\*\* Upper- and lower-case letters the same in the same column and row, respectively, for each vegetable, there is no significant difference ( $P < 0.05$ ).

Source: the authors.

Leafy samples from India were analyzed for TP content and the extracts were obtained from three different solvents (water, methanol and ethanol). Curry leaves (*Murraya koenigii*) had the highest TP content, varying from 3.468,80 $\pm$ 88,03 a 5.084,53 $\pm$ 123,49  $\mu$ g de GAE/g in all extraction solvents. The Agathi (*Sesbania grandiflora*) and the fenugreek (*Trigonella foenum graecum*) showed higher TP content in the aqueous extract. The results show that the amount of TP in leafy vegetables varied between the different extraction solvents. The most likely reasons for these variations may be due to issues related to plant variety, climate, soil, in addition to the ability of these compounds to be soluble in water, in oil, or to be insoluble or attached to cell walls. Therefore, the extraction efficiency is a very important factor in the quantitative

evaluation. Analysis of bioactive compounds from food samples (MOHANKUMAR; UTHIRA; SU, 2018).

Obeng *et al.* (2020) evaluated the bioactive characteristics of four leafy vegetables from Ghana (*Solanum macrocarpo*, *Talinum fruticosum*, *Corchorus olitorius* and *Amaranthus* spp.) from methanolic extract (70%) and found TF levels that ranged from 0.10 µg GA / g to 9.81 µgGA / g, and these vegetables were considered good sources of these bioactive compounds.

The technique of extracting bioactive compounds from vegetables by solvents is also applied for industrial purposes. Gruz *et al.* (2013) compared two technological processes in the recovery of bioactive compounds from grape marc with a view to application in food. According to the authors, the increase in ethanol content favored the extraction of TP compounds, while for anthocyanins the effect was the opposite. Possibly, although anthocyanins belong to the class of phenolic compounds, other subclasses are also measured by the Folin-Ciocalteu methodology employed, and, in this case, the change in polarity by increasing the ethanol content is more favorable for most of these subclasses (GRUZ *et al.*, 2013).

Comparing the levels of TP in the different parts of the kale, a different distribution of these bioactive compounds was observed. In general, the samples of leaves and stalks of vegetables (Table 1), when analyzed separately, presented higher and lower concentrations of TP, respectively, in relation to the whole sample. This occurred in all samples submitted to different thermal processing and types of extraction solvents. The values found are not valid when compared to other vegetables (PINTO *et al.*, 2001), suggesting the full use of these vegetables in the preparations, not neglecting the stems, as normally occurs.

Arbos *et al.* (2010), investigated TP in organic and conventional vegetables and the results were: organic arugula (126,84±4,46 mg/100g), conventional arugula (90,78±2,23 mg/100g), organic lettuce (108,72±2,34 mg/100g), conventional lettuce (91,22±0,91 mg/100g), organic chicory (92,15±1,09 mg/100g) and conventional chicory (81,04±3,64 mg/100g). These data were similar to those found in this study for kale, considering, therefore, kale as an option to compose the individuals' diet plan.

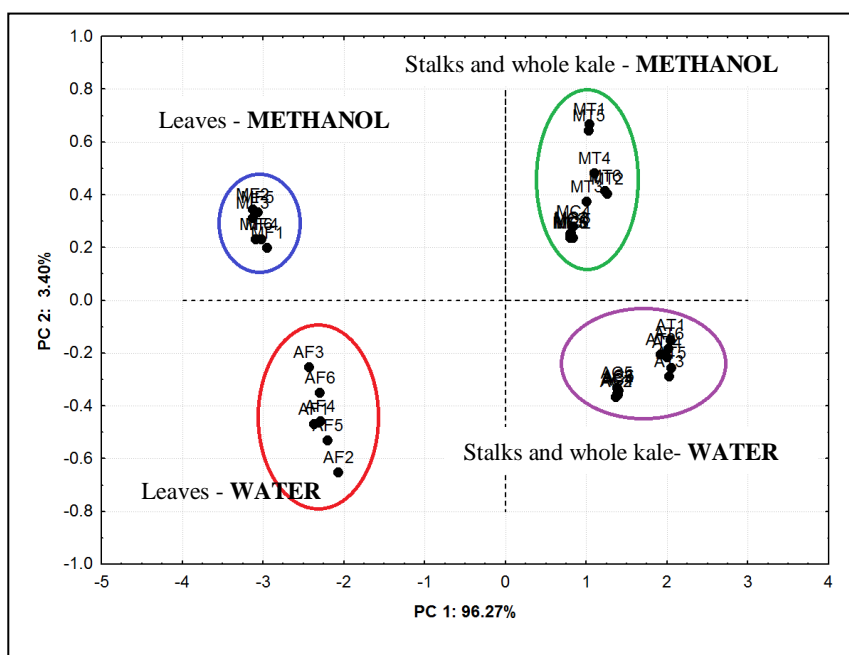
Comparing the thermal treatments (TT) applied, it was demonstrated that steam bleaching (SB) facilitated the extraction of TP, increasing their concentration in most of the samples, especially when aqueous extraction was used (Tables 1). It is suggested that

the thermal processing induces the cellular rupture of the vegetable, promoting the release of the compounds to the external environment, increasing their concentration and making them more bioavailable (NOGUEIRA *et al.*, 2003). Based on these data, SB is recommended as TT for cooking vegetables, as it has been observed that the use of other TT (BI and B) promotes direct contact between vegetables and water and, consequently, a reduction in the TP content of vegetables, possibly by leaching (GREGORY, 2010).

According to Melo *et al.* (2009) food processing can affect the content, activity and bioavailability of these compounds, since they can be degraded or leached into the cooking water. Thus, considering that according to the authors, some vegetables, such as spinach, kale, cauliflower, among others, are usually consumed after being subjected to the cooking process, the increase in temperature makes the present bioactive compounds more available, making cooking the best way to consume these vegetables. However, this processing, in the presence of water, can reduce the amount of phenolic compounds present in vegetables.

Principal component analysis (PCA) was applied to the analysis data to assess the existence of similarities between the samples and to highlight the contributions of the variables in the formation of groups. The first two main components explain 99.67% of the data variability.

**Figure 1** - Scores graph for PC1 versus PC2 obtained by treating the data obtained by determining phenolics in the kale samples.



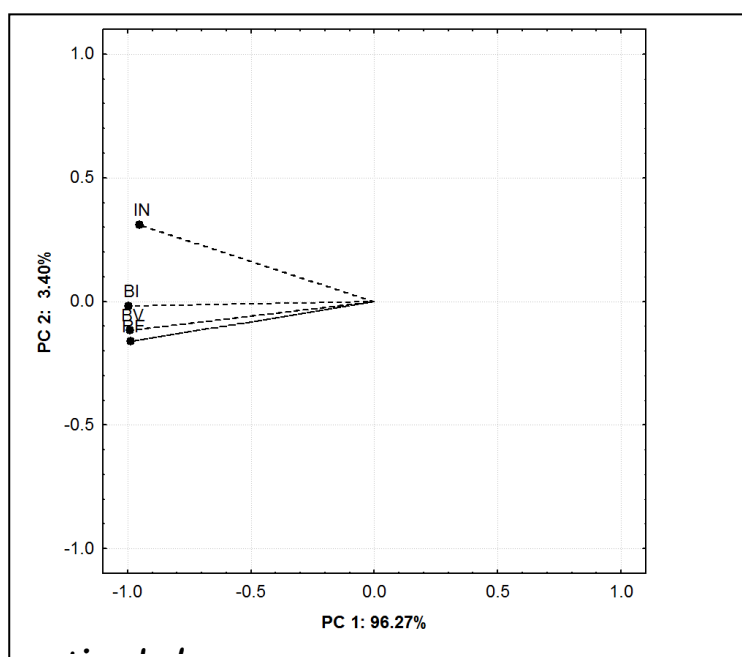
Source: the authors.



The graph of the scores (Figure 1) clearly shows the separation of four main groups: the group formed using extraction with metanol at the top: samples made up of “leaves” (left side); samples composed of “stems” and samples formed by “whole kale” (right side). The groups of extractions with water were at the bottom: samples made up of “leaves” (left side); samples composed of “stems” and “whole kale” (right side). This separation of data by group in relation to the extraction solvent and parts of the plant, possibly, is associated with the distribution of phenolic compounds in the different parts of the vegetable, as well as with their chemical structures and, consequently, their affinity with the evaluated solvents.

The loadings graph (Figure 2) shows the variables that were responsible for separating the sample groups. It is noted that the treatment "*in natura*" was responsible for separating groups of samples at the top of the score graph, that is, of the samples whose extractions were performed with ethanol. On the other hand, the treatments SB, BI and B, were responsible for separating the groups at the bottom of the score graph that has water as an extractor. The groups associated with the “leaves” matrix were located on the left side of the graph indicating that the higher levels of total phenolics are higher in these samples.

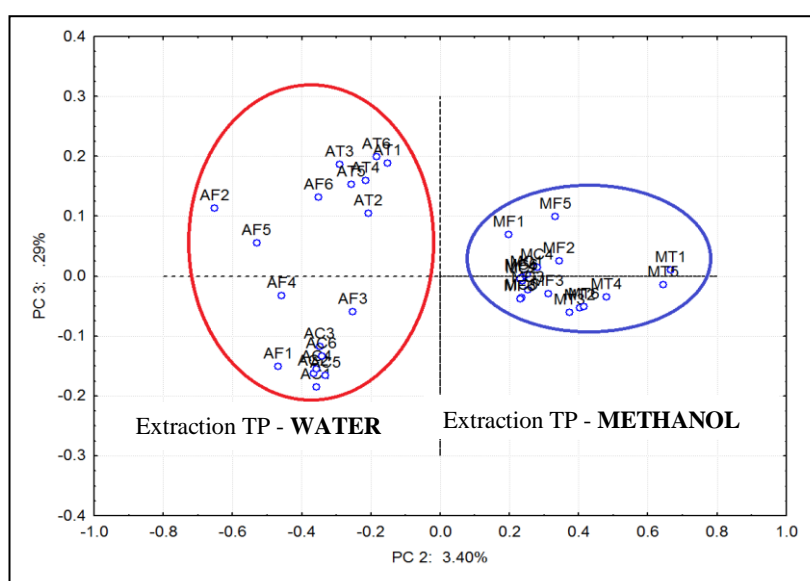
**Figure 2** - Graph of loadings for PC1 versus PC2 obtained by treating the data obtained by determining phenolics in the kale samples.



Thus, this graph shows the contribution of TT in the separation of groups, which is corroborated in Table 1, which shows that the TT used (SB, BI and B) increased the extraction of TP, influencing the chemical behavior of these substances.

The Figure 3 shows the graph of the PC2 x PC3 scores, which shows the behavior of the TP concentration data as a function of the extraction solvents and the different parts of the kale.

**Figure 3** - Graph of the PC2 x PC3 scores for PC1 versus PC2 obtained by treating the data obtained by determining phenolics in the kale samples.

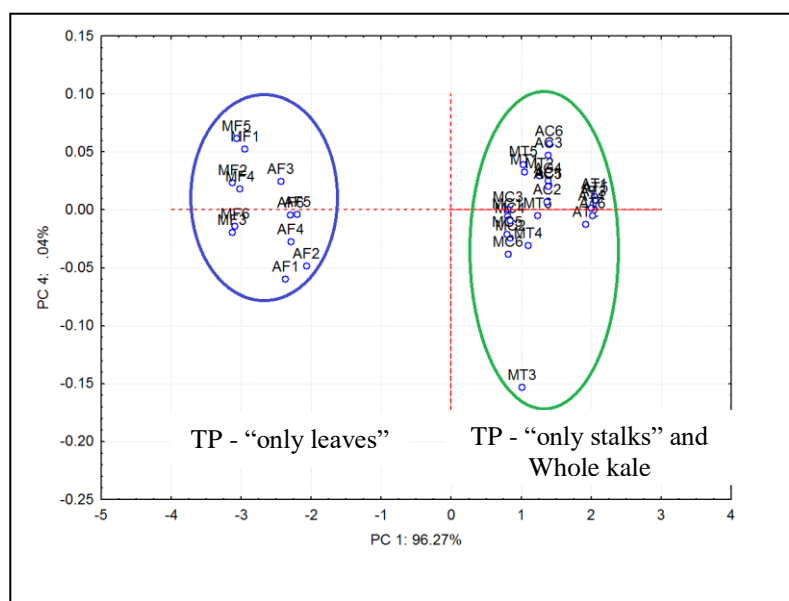


Source: the authors.

There is a clear formation of two groups: the group of samples whose TP compounds were extracted with water and the group of samples whose TP were extracted with methanol. Within these groups, there is also a greater tendency for separation between the stem and leaf samples, mainly in the leaf samples in the water extraction group. These clusters corroborate with the data in Table 1, which shows that there was a greater extraction of TP in the sample “only leaf”. In table 1, in the extraction with methanol, the best extraction in the sample composed of “leaf” is also very evident, whose data were presented further from the group on the right side of Figure 3.

The TP concentration data as a function of the extraction solvents and parts of the vegetable (kale) are represented in the graph of the scores for PC1 X PC4 (Figure 4).

**Figure 4** - Graph of PC1 x PC4 scores for PC1 versus PC2 obtained by treating the data obtained by determining phenolics in the kale samples.



Source: the authors.

Therefore, the formation of two well-defined groups is observed. The group formed by the samples composed of “leaves” and the group formed by the other parts of the kale (stem and whole kale). Within these groups there are also trends of separation according to the solvent used (water or methanol). It is observed that, when the TP are extracted in the “leaves only” group, there is a visible separation of the TP, when using methanol or water as the extraction solvent. According to Table 1, the sample formed by “leaf” was the one that presented the highest concentration of TP when compared to the other samples (“stalk” and whole kale), even considering the two extraction solvents, although methanol has favored better extraction.

Regarding the AA of vegetables, the results obtained for the different parts of the kale by the DPPH and FRAP methods are shown in Table 2.

**Table 2** - Average values of antioxidant activity by DPPH and FRAP methods in kale samples.

Samples	DPPH (IC50)	FRAP ( $\mu\text{M}$ ferrous sulphate/g sample)
Whole kale	517,99 $\pm$ 3,36 <sup>A</sup>	0,0047 $\pm$ 0,00 <sup>A</sup>
leaf	388,16 $\pm$ 3,45 <sup>B</sup>	0,0028 $\pm$ 0,00 <sup>B</sup>
Stalk	640,21 $\pm$ 4,26 <sup>BC</sup>	0,0028 $\pm$ 0,00 <sup>B</sup>

\* For equal letters in the same column there is no significant difference (P < 0.05).

Source: the authors.

Primary antioxidants, such as TP compounds, act by disrupting the chain reaction by donating H to free radicals, especially to LOO •. The efficiency of these antioxidants is related, for the most part, to their ability to donate H to free radicals (PRIOR; WU; SCHAICH, 2005).

For the determination of the antioxidant activity of a substance, there is no universal method for its determination in food samples, since these are complex matrices, with several bioactive compounds with specific chemical characteristics. The DPPH • inhibition and ferric ion reducing power (FRAP) assays were selected to evaluate the antioxidant activity of the samples, since these methods are simple, relatively fast, act by two complementary mechanisms of action and can provide valuable information on the type of antioxidants present in the samples, including their mechanism of action (OLIVEIRA, 2015).

The FRAP method is based on the reduction, in an acidic medium, of a ferric complex of TPTZ (2,4,6-tripyridyl-s-triazine) to a ferrous complex, with a strong dark blue color (APAK *et al.*, 2007). In this method, an electron transfer mechanism occurs (BENZIE; STRAIN, 1996). One of the limitations of this technique is that it only assesses the sample's ability to reduce ferric ions and not its ability to neutralize free radicals or other antioxidant species. In turn, the DPPH test • measures the ability of antioxidant substances present in the samples to neutralize DPPH• radicals (2,2-diphenyldifenil-1-picrylhydrazylpicrilhidrazilo). In methanolic solution, DPPH • has a strong purple color, with a maximum absorbance between 515 and 520 nm. Upon contact with certain antioxidant compounds in the sample, they will neutralize the radical, donating a hydrogen atom and converting it into a colorless yellow compound. The loss of purple color can be monitored over time, by spectrophotometry, and is correlated with the anti-radical capacity of the tested sample (APAK *et al.*, 2007; PRIOR; WU; SCHAICH, 2005; TIRZITIS; BARTOSZ, 2010).

The values found for the DPPH method correspond to the amount of the sample needed to reduce the initial radical concentration (IC<sub>50</sub>) by 50% and is expressed in g of sample / g of DPPH. Thus, the lower the IC<sub>50</sub> value, the smaller the sample quantity used to reduce the DPPH radical and the greater its antioxidant activity (COSTA *et al.*, 2014). Thus, the leaf analyzed separately showed greater antioxidant activity (AA), followed by whole kale and stalk (Table 2), corroborating the results of the TP concentration (Table 1).

Leafy vegetables from Ghana (*Solanum macrocarpo*, *Talinum fruticosum*, *Corchorus olitorius* e *Amaranthus* spp) submitted to AA analysis by the DPPH method and the values found ranged from 1.4% to 70.3% for DPPH activity (OBENG *et al.*, 2020).

Some factors can interfere with the behavior of TP and flavonoid compounds during processing, such as whether they are in free form or conjugated (GUTIÉRREZ-URIBE; ROMO-LOPEZ; SERNA-SALDÍVAR, 2011) or the transformation of them into other compounds (DOGRA; AHUJA; SREENIVASULU, 2013; LIMMONGKON; JANHOM; AMTHONG, 2017), interfering in your concentration and, consequently, in your AA. This, in turn, is related to the chemical structures of bioactive compounds, number and position of hydroxyl groups, glycosylation, among others (AGUILERA *et al.*, 2011).

Arbos *et al.* (2010) determined the AA of vegetables by the DPPH method in organic lettuce extract at a concentration of 0,1 mg.mL<sup>-1</sup> and found 23.1 ± 0.20 of DPPH inhibition (IC<sub>50</sub>), a lower value when compared to those found in this study.

Some studies show that the values found are expressed as a percentage of inhibition. Tiveron (2010), found values (%) of DPPH (1: 5 dilution) for some vegetables, such as: pumpkin (39.7), leek (21), carrot (22), cucumber (12.5). The values found in percentage of inhibition (%) for eggplant in dilution 1: 4 was 31.56 and for gilo 30.74. These values are lower than those found in pumpkin by Tiveron (2010), but they are higher than those found in leeks, carrots and cucumbers.

For the FRAP test, the higher the value obtained, the greater the AA. Thus, for this method, the whole kale showed a higher AA in relation to the leaf and the stalk (Table 2). The AA data obtained by the FRAP method ranged from 0.0028 to 0.0047 μM ferrous sulfate / g sample) and were lower when compared with the data from the study by Tiveron (2010) for other vegetables, which obtained for: the cucumber (0.016 μM ferrous sulfate / g); pumpkin (0.02 μM ferrous sulfate / g), leeks (0.01 μM ferrous sulfate / g) and carrots (0.01 μM ferrous sulfate / g).

Assessing the AA of vegetables subjected to heat treatment, Melo *et al.* (2009) observed that the cooking method employed may not alter, increase or reduce the content of antioxidants present in the vegetable, which is in accordance with the present study, suggesting that heat treatment by different methods may alter the TP content in the vegetables in the sample. and, consequently, AA.

The antioxidant capacity was directly related to the levels of TP obtained by the DPPH method, that is, the highest concentration of TP and AA were found in the kale leaf (Tables 1 and 2), the same not occurring for the FRAP method, where the highest AA was for whole kale. Possibly, the presence of other AA compounds in the sample, the principle of action of the methods used, the difference in the composition of the phenolic compounds in the different parts of this vegetable, as well as its physiology, among others, have interfered in the antioxidant capacity of these samples.

## CONCLUSION

From the data obtained, it is concluded that the concentration of total phenolic compounds and antioxidant activity varied according to the extraction solvent, type of heat treatment used and parts of the vegetables. In the kale, the sample containing “only the leaf” showed the highest amount followed by the whole kale. However, the sample composed of “stalk” showed a significant amount of these bioactive compounds, suggesting the consumption of whole kale, not discarding this part of the vegetable. Among the thermal treatments applied, steam bleaching provided the best extraction of total phenolic compounds. In the study of multivariate comparison, the graphs of the scores showed that the total phenolics were grouped according to the part of the cabbage and the solvent used for extraction, while the graph of the loading showed the influence of heat treatment on the extraction of phenolic compounds with respect to in natura kale for all its parts (stalks, leaves and whole kale) using water as the extraction solvent. Therefore, it is essential to know the changes in chemical compounds that occur in food since its preparation, not only for scientific research, but also for the consumer, who can make decisions about how to prepare their food so that they are healthier.

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