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Physico-chemical, microbiological, and microscopic characteristics of industrialised turmeric powder

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<u>Abstract</u>

This study aimed to evaluate the physico-chemical, microbiological, and microscopic characteristics of industrialised turmeric powder. Experiments were conducted in a completely randomised design, with four treatments represented by different turmeric brands (A, B, C, and D) and four repetition (batches), analysed in triplicate. The inhibition coefficient of DPPH, total phenolic compounds, water activity (aw), pH, titratable acidity (TA), curcumin content, colour parameters (L*, a*, b*, C*, and H*), proximate composition, minerals (Ca, Na, and K), yeast and mould count, coliform count at 45°C, and presence of Escherichia coli and *Salmonella* spp. in the four turmeric brands were evaluated. The microscopic analysis was carried out under optical microscope from the pool of replicates in each treatment. As compared to other brands, brand B exhibited better antioxidant activity and higher concentrations of curcumin and minerals, such as calcium and sodium. There were no differences in a among the brands. With regard to colour parameters, all brands showed a trend for yellow colour (b*); however, only brand A was lighter (L^*) and more saturated (C^*) than brand B. Microbiological and microscopic parameters of turmeric from all brands met with the legal requirements. However, the industrialised turmeric available in the market does not meet the quality standards based on curcumin content, indicating differences between the parameters and the presence of impurities.

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Introduction

The industrial search for natural compounds with safe active ingredients has increased in the recent years. In addition, antioxidants, which inhibit or reduce the effects of free radicals, have been widely used as additives (Denre, 2014). Several studies have reported the beneficial effects of spices including turmeric as a source of phenolic compounds with antioxidant activity and ascorbic acid (Cousins *et al.*, 2007; Singh *et al.*, 2010; Maizura *et al.*, 2011; Denre, 2014; Martinez-Correa *et al.*, 2017). Turmeric has a therapeutic potential as it shows an anti-inflammatory, anticarcinogenic, antioxidant, and antimicrobial activities (Cecilio Filho *et al.*, 2000; Prasad and Aggarwal, 2011; Salehi *et al.*, 2019) besides to its protective effects towards cardiovascular system (Li *et al.*, 2020).

Turmeric (*Curcuma longa* L.) is a natural compound, usually dehydrated and finely ground into powder, which can be consumed fresh or in dried form (Sueth-Santiago *et al.*, 2015). It contains phenolic compounds, such as curcuminoids that include curcumin (the key ingredient of turmeric with the highest content) and are present in the rhizomes (Denre, 2014; Li *et al.*, 2020). Despite this, the curcumin isolated which applied in foods presents low stability (Li *et al.*, 2020). Its bioaccessibility, bioavailability, and toxicity have been a challenge to the food processing industries (Sanidad *et al.*, 2019). In certain export markets, turmeric is assessed based on its curcumin content, which may vary (Li *et al.*, 2020). Moreover, few factors such as the harvest process, raw material quality, and light and heat sensitivity can affect the curcumin quality (Cecilio Filho *et al.*, 2000).

Antioxidant properties of turmeric are believed to be due to the presence of curcumin and carotenoids in turmeric (Cecilio Filho *et al.*, 2000; Sueth-Santiago *et al.*, 2015; Li *et al.*, 2020). Curcumin is a phenolic compound with quality antioxidant activity (Singh *et al.*, 2010). The higher curcumin or curcuminoids content in turmeric, the better the results against oxidation will be expected (Martinez-Correa *et al.*, 2017). The big problem to industries is concerning to the lack of the standardisation, quality control, and the importance to guarantee the presence of bioactive compounds in spices; which serves as an assurance criterion to the consumers (Ayer, 2017; Gad and Bouzabata, 2017). According to Srinivasan *et al.* (2016) and Ayer (2017), some problems related to the productivity of turmeric are unsatisfactory, such as the kind of genotype, multiplication, and content in curcumin, besides storage conditions or soil quality.

Carotenoids are natural pigments that impart attractive colours (e.g., yellow and orange) to various foods, such as spices. In general, more than 600 types of carotenoids exist; however, only 40 are present in the diet (Kandlakunta *et al.*, 2008). Carotenoids are an antioxidant sources and provide several health benefits, including vitamin A, precursor activity, increased immunity, and decrease in chronic diseases (Kandlakunta *et al.*, 2008). Nevertheless, adding turmeric to products with different food matrices can limit its antioxidant activity. The addition of turmeric in food is common in several cultures (Sueth-Santiago *et al.*, 2015; Oliveira *et al.*, 2017); however, the authorities do not monitor the quality of the spice available in the consumer market.

Favourable climatic conditions including humidity and temperature, agricultural practices, and storage can promote turmeric contamination by filamentous fungi and mycotoxin production. Spices do not deteriorate due to fungal action; however, these microorganisms will multiply during the dehydration process, and the addition of the spice to other foods can increase the microbial load, thereby leading to food poisoning (Silva *et al.*, 2012).

It has been reported that turmeric can contain microorganisms (Silva *et al.*, 2012); and spices should be processed, distributed, and stored following the Good Manufacturing Practices to ensure the quality of the product, thereby avoiding contact with chemical, physical, and microbiological substances (Oliveira *et al.*, 2017). Nevertheless, contamination can occur during production, which may or may not be detected with the naked eye, making it necessary to evaluate samples using a microscope (Dent, 1977; Oliveira *et al.*, 2017).

Sagoo *et al.* (2009) and Oliveira *et al.* (2017) reported the presence of coliforms and *Salmonella* as microbiological contaminants, and suggested that spices subjected to heat have a higher quality. Several authors have reported that high microbial load indicates faecal contamination, caused from the lack of hygiene during the manufacturing process.

Some researchers have also demonstrated the action of curcumin, which is present in turmeric, as

an antimalarial (Martinez-Correa *et al.*, 2017). Furthermore, synergistic effect was also verified when it was added together with antibiotic against methicillin-resistant *Staphylococcus aureus* (Mun *et al.*, 2013). Antiviral and antifungal effects have also been highlighted (Moghadamtousi *et al.*, 2014).

Dehydrated spices present a potential hazard, in particular, if they are added to other foods after cooking or *in natura*, causing risk of food poisoning by pathogens. Considering the potential antioxidant activity of the spice, presence of carotenoids, calcium, and sodium content in food intake, it is possible that industrialsed turmeric does not meet the quality standards in different brands available in the consumer market.

Besides the quality of turmeric and its growing conditions, suppliers must also ensure the composition and quality of raw materials (Chatzinasiou et al., 2019). The presence of microbiological contaminants, such as filamentous fungi and mycotoxins, Salmonella, E. coli, and impurities will promote the risk of foodborne diseases, thus causing damage to the public health. In addition, consumers are seeking turmeric with high antioxidant activity whose content of curcumin had been attributed to this activity as well as medicinal properties (Forsyth et al., 2019); although this is not a parameter guaranteed by the industries, neither the therapeutic benefits that are related to the turmeric (Chatzinasiou et al., 2019). In many parts of the world, researches regarding the quality of turmeric had been discussed, especially about the addition of substances to improve the colour of the spice as well as to increase the amount of biomass produced (Prasad and Aggarwal, 2011; Moore et al., 2012; Chatzinasiou et al., 2019).

In fact, it is very important to know the quality of commercialised turmeric (Chatzinasiou *et al.*, 2019), because there is a large product available, in different brands and markets, with the same label of "turmeric", without the guarantee of quality. Moreover, this study discusses the quality of turmeric produced by the food industries, specifically phenolic compounds, and mineral contents (ashes). Thus, this study aimed to evaluate the physico-chemical, microbiological, and microscopic characteristics of industrial turmeric powder commercialised in Brazil, specifically in the central region of the country.

Materials and methods

Samples of different brands namely brand A, B, C, and D of turmeric powder were collected from September 2017 to February 2018. These samples were packed in plastic containers, stored in a dry place (temperature 28°C, 25% humidity) and protected from light until further analysis.

Physico-chemical analyses

The experiment was conducted in a completely randomised design with four treatments represented by brands (T1:A, T2:B, T3:C, and T4:D) and four repetitions (represented by the batches). All physico-chemical parameters such as a_w , pH, titratable acidity (TA), curcumin content, α -carotene, β -carotene, ashes, L*, a^* , b^* , C*, and H* were analysed in triplicate.

Antioxidant activity (DPPH) and total phenolic compounds (TPC)

The antioxidant DPPH radical scavenging activity and the total phenolic compounds (TPC) content were determined using turmeric alcohol extracts. To prepare the alcohol extract, 2.5 g of turmeric powder was weighed, and ethanol PA was added until the final volume of 25 mL in a volumetric flask. It was then allowed to stand for 1 h in the dark. Next, the contents were transferred to a 50 mL Falcon tube, centrifuged (3,310 rpm for 15 min), filtered on quantitative filter paper (7.5 μ m), and the supernatant (at a concentration of 100 mg/mL) was stored in an amber bottle until further analysis. Aliquots of the supernatant were collected and diluted in 95% ethanol to concentrations of 3.25, 5.00, 6.25, 7.50, and 8.25 mg/mL, which were all used for DPPH analysis and calculation of the IC_{50} . The highest concentration (8.25 mg/mL) was chosen to determine the CPT and estimate the range of phenolic content.

The antioxidant activity was determined via DPPH method, based on the scavenging of the DPPH radical, as proposed by Zhang and Xu (2015). For this, 0.5 of mL aliquots of each dilution were transferred to a test tube and 2.5 of mL ethanol solution of 0.06 mM DPPH was added in a light-protected environment (A₁).

To avoid interference of the solution colour during spectrophotometric readings, a solution was prepared using 5 mL of each dilution of the test solution and was homogenised in an amber volumetric flask. An aliquot of 0.5 mL of this new solution was added to 2.5 mL of ethanol PA, which was used as the blank (A_2). For the control (A_0), 0.5 mL of ethanol PA was added to 2.5 mL of the ethanol solution of 0.06 mM DPPH. The absorbance was read on a Shimadzu UV-1800 spectrophotometer at a wavelength of 517 nm, protected from light, after 30 min from the beginning of the reaction using ethanol PA as blank. The inhibition coefficient (IC₅₀) of the DPPH radical was calculated using Eq. 1:

$$IC_{50} = \frac{[A_0 - (A_1 - A_2)]}{A_0 * 100}$$
(Eq. 1)

where, A_0 = absorbance of the control in which ethanol replaced the extract; A_1 = result of the extract and DPPH radicals; and A_2 = absorbance of the blank to eliminate the colour effect of the extract. The equation of the analytical curve (percentage of inhibition by various concentrations of the extract) was used to calculate IC₅₀, replacing the value of *y* by 50 to determine the concentration of the sample that can reduce DPPH by 50%.

The TPC was determined via Folin-Ciocalteu colorimetric method. The absorbance was read in a Shimadzu UV-1800 spectrophotometer at 760 nm. To obtain the data, the analytical curve ($R^2 = 0.9975$) constructed with gallic acid was used. The results of the total phenolic compounds (TPC) were expressed in milligrams of gallic acid equivalents (GAE) in 100 g sample.

Physico-chemical parameters

The water activity (a_w) was determined by direct reading using an Aqualab 4TE (decagon), with the sample at room temperature (25°C), by following method 978.18 (AOAC, 2012). The pH was determined by potentiometry in neutralisation titrimetry with a Hanna HI2221, following method 943.02 of AOAC (2012). The potentiometric titratable acidity (TA) was determined by titration using 0.1 N sodium hydroxide until it attained pH 8.4 (IAL, 2008). The curcumin content was determined following the method described in IAL (2008). The absorbance was read at 425 nm in a Shimadzu UV-1800 spectrophotometer using ethanol PA as a blank.

The total carotenoid content of turmeric was determined by reading the absorbance of α -carotene and β -carotene at 453 and 444 nm, respectively, in a Shimadzu UV-1800 spectrophotometer following method 123/IV (IAL, 2008). The total carotenoid content was calculated using the absorption coefficients of β -carotene (=2592) and α -carotene (=2400) in petroleum ether. The measurements were carried out in triplicate. The colour parameters were determined by the CIELAB system $(L^*a^*b^*)$, where the readings were determined as L*, a*, and b*, calibrated to a blank standard (CMA-177), with illuminant D65, 10° for the standard observer and specular component excluded (SCE), using a Minolta CM-700D (Minolta-Japan) portable colorimeter. The value of Chroma (C*) and hue angle value (H*) used to indicate the saturation and hue of the colour, respectively, (AOAC, 2012) was obtained using Eqs. 2 and 3:

$$C^* = (\Delta a^{*2} + \Delta b^{*2})1/2$$
 (Eq. 2)

$$H^* = \tan -1 (b^*/a^*)$$
 (Eq. 3)

Analysis of the centesimal and mineral composition

The moisture content was analysed via gravimetric method in an oven at 105°C for 24 h (method 012/IV); the ashes by incineration in a muffle furnace at 550°C (method 018/IV); the lipids by the Soxhlet extraction (method 032/IV); the total carbohydrates by the difference indicated in the formula provided in the methods proposed by IAL (2008); and the proteins by determining the total nitrogen content using the Kjeldahl nitrogen distilling apparatus with a factor of 6.25 to convert nitrogen into protein (method 928.08) of AOAC (2012). For analysing minerals (calcium, sodium, and potassium), the treatments were subjected to wet digestion following the method 393/IV of IAL (2008). The calcium content was determined using the titrimetry of complexation method with indicator black of eriochrom T, according Baccan et al. (2001). The sodium content was determined using ion-selective electrodes in the LAQUAtwin-Horiba equipment, calibrated with the standard of 150 mg/L solution (Standard solution: model Y022L).

To determine the potassium content, a Digmed DM-62 flame photometer was used, with an aspiration rate of 1.9 mL/min and repeatability in a standard deviation of 0.04. The analytical curve was traced at concentrations of 0 - 50 mg/L of K ($R^2 = 0.9833$). The methodology was performed following the method 956.01 of AOAC (2012).

$$y = 0.1185x + 0.7057$$
 (Eq. 4)

Where, y = absorbance of KCl (Potassium Chloride), and x = concentration of KCl.

Microbiological and microscopic analyses

The presence of moulds, yeasts, coliforms at 45°C, as well as that of *E. coli* and *Salmonella* spp. was determined in 10^{-1} dilutions. Briefly, 25 g of each sample was transferred into vials containing 225 mL of peptone water (0.1%). The microorganisms were inoculated on plates with culture-specific chromogenic enzyme substrates (Compact Dry-Idexx®) with surface plating and direct counting. Here, 1.0 mL aliquots of the dilutions were inoculated on specific plates to detect mould and yeast, coliforms at 45°C, and *E. coli*.

Mould and yeast plates were incubated at 30°C for 5 d and the plates for detecting *E. coli* and coliforms at 45°C were incubated at 45°C for 24 h. For analysing *Salmonella* spp., the sample was kept in peptone water in a sealed vial for approximately 16 - 18 h at 37°C. Furthermore, 0.1 mL of the sample was inoculated into a test tube containing 10 mL of

Rappaport Vassiliadis broth, and the tubes were incubated at 41°C for 24 h. Simultaneously, a 0.1 mL aliquot of the sample was added to 1 mL of autoclaved distilled water and plated. The plates were incubated at 44°C for 24 h and the results were expressed in colony forming units (CFU/g).

The microscopic analysis of the organisms was carried out under an optical microscope (Kasvi Red-220), binocular, and LED illumination (3.4 V, 700 mA, 2.38 W, 60 Hz). The sample were analysed under achromatic objectives at $40 \times$ and $100 \times$, from the pool of the batches of each treatment. Briefly, 10 g of the sample was added to 100 mL of 2.5% sodium hypochlorite until the sample was cleared. Filtration was carried out under vacuum on filter paper and the material was then washed with distilled water. Small amount of the material was used to prepare slides with 2% glycerinated water for observing foreign materials. The result was expressed as mean values after visualising the foreign materials in the sample. To study the presence of starch, Lugol's solution was used. The results were compared to RDC No. 12 (Brazil, 2001) and RDC No. 14 (Brazil, 2014) for foreign materials, and CNNPA No. 12 (Brazil, 1978) for the presence of starch.

Statistical data analysis

All data, except TPC and DPPH as well as proximate, microbiological, and microscopic composition, were subjected to the Shapiro-Wilks test and, when normality between the data was verified (p > 0.05), the mean values of the treatments were analysed by ANOVA. In case of any difference between the treatments, a Scott-Knott mean test (p < 0.05) was applied. The means that did not present normality were subjected to the Kruskal-Wallis test. The data were analysed with the ASSISTAT 7.7 software and the level of significance was set at 5% probability of difference between means for all analyses.

Results and discussion

Physico-chemical analyses

The ethanol extract of treatments B and D presented better values for inhibiting 50% of the generated radicals (IC₅₀) and total phenolic compounds (TPC; mg GAE/g) as listed in Table 1.

Turmeric contains curcuminoids, which are responsible for the potent antioxidant action (Cousins *et al.*, 2007) and curcumin can increase the antioxidant activity, neutralise free radicals, and reduce lipid peroxidation.

DPPH is preferably used to analyse the

IC ₅₀	TPC (mg GAE/g)	
73.142 ± 0.002	2.262 ± 0.001	
7.915 ± 0.002	15.684 ± 0.009	
25.825 ± 0.004	5.470 ± 0.001	
8.799 ± 0.004	14.357 ± 0.002	
	73.142 ± 0.002 7.915 ± 0.002 25.825 ± 0.004	

Table 1. Means \pm standard deviation of the inhibition coefficient (IC₅₀) and total phenolic compounds (TPC) obtained from the ethanol extracts of turmeric powder.

antioxidant activity and is based on the determination of the antioxidant scavenger activity *in vitro*. DPPH results in a violet solution when dissolved in solvents such as ethanol and methanol, and this colour is reduced in the presence of curcumin. The use of DPPH provides an easy and quick approach to evaluate the antioxidant properties of curcumin (Cousins *et al.*, 2007).

Various brands of turmeric exhibited antioxidant properties, mainly due to the hydroxyl and phenolic groups of curcumin, a substance commonly found in turmeric (Sueth-Santiago *et al.*, 2015). The results indicate that the free radical scavenging activity, observed via DPPH analysis, can presumably be attributed to a high content of phenols with larger reducing capacity. Denre (2014) obtained an IC₅₀ of 5.99 mg/mL for the extract of the turmeric rhizomes marketed in India, which is similar to the values obtained for the IC₅₀ activity of treatments B and D; whereas, Cousins *et al.* (2007) and Hirun *et al.* (2014) obtained an IC₅₀ of 9.74 mg/mL and IC₅₀ between 23.41 and 59.56 mg/mL, respectively. Growing system, field conditions, postharvest processing, raw quality, drying process (Cousins *et al.*, 2007; Singh *et al.*, 2010) are conditions that have negative influence in the final product. In fact, any temperature could make some changes or affect the activity and/or degradation of the phenolic compound. Generally, the rhizomes of turmeric are fresh when harvested, and then, they are submitted to drying process. After that, they are grounded and sold at the markets, or proceed to industries for processing or add in some other product such as ingredient of curry or seasoning.

Drying process differs among industries or among producers of the turmeric, and unfortunately, there was no standard method with focus on bioactive compounds protection. According to the researchers, those quality can be affected by the temperature and time that the turmeric rhizomes are dried and stored (Park *et al.*, 2019; Surendhar *et al.*, 2019; Zuniega and Esguerra, 2019).

Treatment B, represented by brand B, presented a higher acidic value when compared statistically with the other brands analysed (p < 0.05; Table 2). In contrast, the brands (treatments) with the lowest acidic values were A and C when compared statistically with the other brands analysed (p > 0.05; Table 2). These brands also revealed the least values for IC₅₀ and TPC. It was observed that brand B has a higher curcumin content (p < 0.05) as compared to brand A. The presence of curcumin interferes with the pH since it has three acidic hydrogens that can be

D (Brand			
Parameter	Α	В	С	D
$a_{ m w}$	$0.50\pm0.04^{\rm a}$	$0.53\pm0.03^{\text{a}}$	$0.53\pm0.04^{\rm a}$	$0.52\pm0.05^{\text{a}}$
pН	$6.25\pm0.11^{\text{a}}$	$6.34\pm0.09^{\text{a}}$	6.15 ± 0.10^{b}	$6.06\pm0.06^{\rm b}$
TA	$5.25\pm1.36^{\rm c}$	8.89 ± 0.46^{a}	$4.78\pm0.99^{\text{c}}$	7.07 ± 0.73^{b}
Curcumin (%)	$3.44\pm0.01^{\rm B}$	$4.12\pm0.02^{\rm A}$	$\begin{array}{c} 3.50 \pm \\ 0.04^{\mathrm{AB}} \end{array}$	$3.98\pm0.05^{\rm AB}$
α -carotene (µg/g)	$0.77\pm0.14^{\rm a}$	0.70 ± 0.12^{a}	$0.56\pm0.04^{\rm a}$	$0.77\pm0.18^{\rm a}$
β -carotene (μ g/g)	$0.76\pm0.20^{\rm A}$	$0.56\pm0.08^{\rm AB}$	$0.50\pm0.03^{\rm B}$	0.57 ± 0.07^{AB}
Ash (%)	$1.67\pm0.66^{\rm d}$	$6.49\pm0.58^{\text{a}}$	$3.02\pm0.35^{\text{c}}$	4.89 ± 0.24^{b}

Table 2. Means \pm standard deviation of the physico-chemical analyses of the treatments (represented by brands).

Identical lowercase letters in the same row do not differ significantly by the Scott-Knott test at a 5% significance level; identical uppercase letters in the same row do not differ significantly by the Kruskal-Wallis test at a 5% significance level. $a_w =$ water activity; TA = total titratable acidity (expressed in mL of NaOH/100 g).

released in the medium, making it more acidic (Sueth-Santiago *et al.*, 2015), which can be accounted for the differences in IC₅₀, TPC, curcumin content, and acidity between the analysed brands as well as the relationship between the antioxidant activity and curcumin content.

The relationship between the TPC content and IC_{50} indicated similar results for different brands (higher contents found in B, D, C, and A, respectively). Moreover, Maizura *et al.* (2011) reported similar results in a study revealing that higher total phenolic content in the extracts resulted in enhanced antioxidant activity.

As reported by several authors, a difference was observed in the content of total phenolic compounds in turmeric, lower value reported by Denre (2014). The difference between TPC values can be attributed to several factors such as turmeric exposure to the sun, soil, drying method, variety, climate of the region where it was grown, and storage temperature (Cousins *et al.*, 2007).

The quality of the raw material and controlling the conditions and types of cultivation would impact the quality of the raw material, in turn leading to an impact on the market value of the turmeric and postharvest processing (Cecilio Filho *et al.*, 2000). These can impact the market value of the rhizomes, thus requiring a standardisation among the turmeric available in the market. It is recommended to standardise the quality of the turmeric available in the consumer's market via inspections carried out by inspection agencies, to ensure the presence of turmeric with the bioactive potential demonstrated by several authors.

The water activity (a_{m}) and α -carotene parameters did not reveal any differences (p > 0.05) between the analysed brands (Table 2). Brands A and B presented statistically equal and higher pH values as compared to the other brands (p < 0.05). The pH values of brands A and B were 6.06 to 6.34, respectively. However, the titratable acidity values of brands A and B were 4.78 to 8.89, respectively, and brand B presented higher acidity values. The pH and the acidity are inversely correlated. Nevertheless, the presence of organic acids can cause a buffering effect during the storage, or promote the development of microorganisms, thus interfering with the pH value and acidity, and leading to a misinterpretation between the pH value and the acidity of the product. The control of the pH and water activity guarantees the quality of the product during the validity period as they interfere in the development of deteriorating and pathogenic microorganisms. Foods with an a_w value lower than 0.6 are considered safe as they are less prone to the deterioration caused by bacteria, filamentous fungi, and chemical and enzymatic reactions (Oliveira *et al.*, 2017).

Regarding β -carotene (Table 2), brand A presented a higher amount (0.76, p < 0.05) as compared to brand C (0.50). The maturation stage of the rhizome can interfere with the carotenoid content (Kandlakunta *et al*, 2008; Singh *et al.*, 2010). The difference in the quantity of carotenoids is influenced by the following factors: processing, exposure of turmeric to high temperatures or sunlight, and the type of storage, which may cause unsaturation and degradation of pigments, variety of species, and the place and type of cultivations (Cecilio Filho *et al.*, 2000).

For the ash content (Table 2), resolution No. 12 of the National Commission on Norms and Standards for Foods (CNNPA) (Brazil, 1978) establishes a maximum limit of 7.5%. Therefore, all brands comply with the present legislation. Significant difference was observed between all brands, and brand B presented the highest ash content (6.49). This is due to the high levels of minerals, such as K, P, and Ca in the turmeric composition, resulting in a high ash content ranging from 2 to 9%, as observed by Cecilio Filho *et al.* (2000). For consumer satisfaction, it should be ensured that the turmeric available in the market is free of contaminants for the final product.

The analysed brands revealed differences in the turmeric colour (p < 0.05), except for parameter b*. The L* component ranged from 44.10 to 61.90 and brand B is darker than brand A (Table 3).

The coordinate a* (Table 3) ranged from 16.46 to 25.38, and brand C presented a lower value as compared to that of the other brands (p < 0.05). The values obtained in this study are similar to those observed by Hirun *et al.* (2014), although the turmeric powder manufacturers of the analysed brands have not specified the drying or processing method used.

For the coordinate b* (Table 3), a small variation from 60.40 to 67.45 was observed, with no statistically significant difference, and with a trend towards yellow, as expected for turmeric. Turmeric contains on average 1.8 to 5.4% of pigments responsible for the yellow colour (Cecílio Filho *et al.*, 2000). This is due to the presence of curcuminoids, which are responsible for the pigmentation of reddish-yellow rhizomes (Cousins *et al.*, 2007), and thus are used for food colouring and the main ingredient in curry (Forsyth *et al.*, 2019).

The coordinate C* (Table 3) presented values between 64.58 and 69.45, and brands A and C had the most vivid colours as they are more saturated, while B and D being the faintest (p < 0.05).

The hue values (H*) ranged from 68.31 to

Brand	L*	a*	b*	C*	H*
А	$61.90\pm5.78^{\rm A}$	$25.38\pm3.76^{\rm a}$	64.48 ± 6.35^{a}	$69.45\pm5.11^{\text{a}}$	$68.31\pm4.27^{\rm B}$
В	$44.10\pm2.07^{\rm B}$	$22.27 \pm 1.03^{\text{a}}$	60.62 ± 2.43^{a}	64.58 ± 2.62^{b}	$69.83\pm0.29^{\rm AB}$
С	$48.98\pm1.26^{\rm AB}$	$16.46\pm0.78^{\text{b}}$	67.45 ± 0.76^{a}	$69.42\pm0.82^{\text{a}}$	$76.28\pm0.62^{\rm A}$
D	$46.56\pm3.44^{\mathrm{AB}}$	$23.68 \pm 1.10^{\text{a}}$	$60.40\pm2.98^{\text{a}}$	64.94 ± 3.23^{b}	$68.69\pm0.12^{\rm AB}$

Table 3. Mean \pm standard deviation of the colour parameters in the analysed turmeric brands.

Identical lowercase letters in the same column do not differ significantly by the Scott-Knott test at a 5% significance level; identical uppercase letters in the same column do not differ significantly by the Kruskal-Wallis test at a 5% significance level; ns = not significant (p > 0.05).

76.28 (Table 3), and the colour is estimated to be in the first quadrant, more precisely between orange and yellow (Ramos and Gomide, 2017). Difference was observed (p < 0.05) between brands C and A, with C depicting a yellow hue (76.28) and A depicting an orange hue (68.31), which supports the use of turmeric for food colouring.

The ash content varied from 1.98 to 7.03%, and brand B presented higher values (Table 4).

The moisture ranged from 10.58 to 13.00% in the analysed brands, with brand C presenting lower values (Table 4). The brands presented lipid contents (Table 4) between 0.42 and 1.88%, which is lower than the values obtained by Kuttigounder *et al.* (2011).

The moisture and lipid content values were lower as compared to those observed by Kuttigounder *et al.* (2011). Protein content (Table 4) differed slightly between the brands, that is, 5.28 and 7.30%. The total carbohydrate contents of the brands (Table 4) were similar to the values obtained by Kuttigounder *et al.* (2011). The proximate composition varies based on the variety, agricultural practices, rhizome planting location, use of agricultural pesticides and rhizome maturity, conditions of cultivation and processing (Cecilio Filho *et al.*, 2000; Hirun *et al.*, 2014), thus reinforcing the need to monitor and standardise the types of cultivation and processes.

Brand B presented higher calcium (237.85 mg/100 g) and sodium (180.21 mg/100 g) contents as compared to the other analysed brands (p < 0.05; Table 5).

Hossain and Ishimine (2005) found higher Ca and K contents in powdered rhizomes cultivated in grey soils in Japan, since this type contained the

Table 4. Mean \pm standard deviation of the proximate composition of the analysed turmeric brands based on dry weight (%).

Brand	Moisture (%)	Ash (%)	Lipid (%)	Protein (%)	Carbohydrate (%)
А	12.02 ± 0.06	1.98 ± 0.02	0.42 ± 0.04	7.30 ± 0.18	78.27 ± 0.07
В	12.58 ± 0.01	7.03 ± 0.11	0.47 ± 0.06	7.17 ± 0.12	72.73 ± 0.07
С	10.58 ± 0.05	3.32 ± 0.05	1.88 ± 0.08	5.78 ± 0.13	78.43 ± 0.08
D	13.00 ± 0.64	5.13 ± 0.10	0.55 ± 0.07	5.28 ± 0.10	76.03 ± 0.08

Table 5. Means \pm standard deviation of the mineral contents of the analysed turmeric brands.

Parameter	Brand			
(mg/100 g)	Α	В	С	D
Calcium	$161.31 \pm 9.77^{\circ}$	237.85 ± 18.93^{a}	$138.54\pm10.4^{\rm d}$	190.40 ± 9.99^{b}
Sodium	$53.96 \pm 16.55^{\circ}$	180.21 ± 16.33^{a}	$88.75\pm30.01^{\text{b}}$	$116.67 \pm 13.05^{\text{b}}$
Potassium	$49.25\pm26.25^{\rm A}$	$85.89\pm14.63^{\rm A}$	$69.82\pm60.77^{\rm A}$	$49.95\pm5.78^{\rm A}$

Identical lowercase letters in the same row do not differ significantly by the Scott-Knott test at a 5% significance level; identical uppercase letters in the same row do not differ significantly by the Kruskal-Wallis test at a 5% significance level; ns = not significant (p > 0.05).

highest amount of K and Ca. The highest sodium content was obtained via cultivation in red soil. The brands revealed no differences (p < 0.05) in the potassium contents. Mane *et al.* (2018) found lower values of calcium and sodium in their studies on turmeric, which corroborates the influence of cultivation conditions on the quality of turmeric. Nevertheless, the studies reported that the differences between mineral availability and its absorption could be due to the amount of minerals or interaction between them.

Microbiological and microscopic analyses

No coliforms were observed at 45°C (CF), with absence of *E. coli* (EC) and *Salmonella* spp. (SL) in the analysed brands. Nevertheless, moulds and yeasts were present in brands A (3.6×10^2 CFU/g) and C (1.10×10^1 CFU/g). The absence of these bacteria indicates that the turmeric brands (brands A, B, C, and D) complied with the microbiological standards in foods established by RDC No. 12/2001 (Brazil, 2001). Although moulds and yeasts were present in brands A and C, both brands complied with the legislation that establishes 5×10^3 CFU/g of sample as a reference value (Brazil, 1998).

Favourable environmental conditions of humidity, temperature, and storage can favour the contamination of spices by insects and fungi which could results in food poisoning (Silva *et al.*, 2012). Therefore, monitoring should be carried out by the competent authorities in order to avoid contamination with the mycotoxins.

The microscopic analysis revealed the presence of impurities, such as dead mites, animal hair, fragments, and insect eggs, all in brand A, and were absent in the other analysed brands. Nevertheless, tissue fragment was observed in brands C and D. Despite the impurities observed in the analysed brands, all brands complied with the legislation (Brazil, 2014). Nevertheless, it is known that fragments of insects pose a risk to the consumer's health, as they indicate failures in good manufacturing practices. No starch was observed in the analysed brands.

Knowing the functional properties of turmeric and the consumption habits of the population, it is recommended for the competent agencies to thoroughly inspect the products, and thereby ensure that the marketed product meets the quality standards, assuring the attributes of the spice.

Conclusion

Commercially available turmeric does not have a standardised quality. All brands presented good microbiological quality, and complied with the stipulated legislation. Although the brands complied with the limits established by the legislation regarding the presence of impurities, the industry still indicates failures in Good Manufacturing Practices in the production chain and needs to improve the production, transport, and storage to ensure a contamination-free product.

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