Research, Society and Development, v. 9, n. 7, e504974409, 2020
(CC BY 4.0) | ISSN 2525-3409 | DOI: http://dx.doi.org/10.33448/rsd-v9i7.4409

Oliveira, RES, Cardoso, MG, Santiago, WD, Barbosa, RB, Alvarenga, GF & Nelson, DL. (2020). Physicochemical parameters and volatile composition of cachaça produced in the state of Paraíba, Brasil. ***Research, Society and Development,*** *9*(7):1-27. e504974409.

Parâmetros físico-quimicos e composição volátil de cachaças produzidas no estado da Paraíba, Brasil

Physicochemical parameters and volatile composition of cachaça produced in the state of Paraíba, Brasil

Parámetros fisicoquímicos y composición volátil de las cachaças producidas en el estado de Paraíba, Brasil

Recebido: 11/05/2020 | Revisado: 12/05/2020 | Aceito: 14/05/2020 | Publicado: 23/05/2020

Renan Elan da Silva Oliveira
ORCID: https://orcid.org/0000-0002-4146-2581
Federal University of Lavras, Brazil
E-mail: renanesoliveira@gmail.com
Maria das Graças Cardoso
ORCID: https://orcid.org/0000-0001-8075-1725
Federal University of Lavras, Brazil
E-mail: mcardoso@ufla.br
Wilder Douglas Santiago
ORCID: https://orcid.org/0000-0003-4941-2527
Federal University of Lavras, Brazil
E-mail: wilder.dsantiago@ufla.br
Richard Bispo Barbosa
ORCID: https://orcid.org/0000-0002-1358-0257
Federal University of Lavras, Brazil
E-mail: richardbispo2009@hotmail.com
Gabriela Fontes Alvarenga
ORCID https://orcid.org/0000-0002-2064-858X
Federal University of Lavras, Brazil
E-mail: gabrielafalvarenga@gmail.com
David Lee Nelson
ORCID: https://orcid.org/0000-0001-7435-3675
Federal University of Vales do Jequitinhonha e Mucuri, Brazil

E-mail: dlnelson@ufmg.br

Resumo

Produzida e comercializada em todo Brasil, a cachaça é um símbolo do país e para garantir a segurança dos consumidores e a qualidade da bebida esta deve atender aos padrões legais estabelecidos pela legislação brasileira. Apesar disso, ainda são comuns relatos de inadequações e a complexidade da sua composição requer estudos mais detalhados. Esse trabalho teve como objetivo avaliar a composição química de cachaças produzidas no estado da Paraíba observando a adequação aos parâmetros exigidos pela legislação e a identificação dos compostos voláteis presentes nessas bebidas. As amostras das cachaças foram coletadas no comércio local, em suas embalagens originais e encaminhadas ao Laboratório de Análise de Qualidade de Aguardente da Universidade Federal de Lavras, Minas Gerais. Os parâmetros analisados foram aqueles estabelecidos pela legislação brasileira como indicadores de qualidade. Os compostos voláteis foram identificados pela técnica de cromatografia gasosa acoplada a espectometria de massas (GC-MS). Entre as 20 amostras analisadas, 17 destas estavam fora dos padrões em pelo menos um dos parâmetros estabelecidos pela legislação. A complexidade da composição volátil foi comprovada sendo encontrados 57 compostos representados majoritariamente por ésteres (decanoato de etila e dodecanoato de etila) e álcoois (3-metil-1-butanol).

Palavras-chave: Bebidas alcoólicas e destiladas; Controle de qualidade; Legislação; Padronização.

Abstract

Cachaça is produced and marketed throughout Brazil to an extent that it has become a symbol of the country. To ensure the safety to consumers, the quality of the beverage must meet the legal standards set by Brazilian law. Nevertheless, reports of inadequacies are still common, and the complexity of its composition requires more detailed studies. This study sought to evaluate the chemical composition of cachaça produced in the state of Paraiba, determine the conformity with the parameters required by legislation, and identify volatile compounds present in these beverages. The cachaça samples were collected from the local commerce in their original packaging and sent to the Laboratório de Análise de Qualidade de Aguardente of the Universidade Federal de Lavras, in Lavras, Minas Gerais, Brazil. The parameters analyzed were those established by Brazilian legislation as indicators of quality. Volatile

compounds were identified by mass spectrometry coupled to gas chromatography (GC-MS). Among the 20 samples analyzed, 17 were irregular with respect to at least one of the parameters established by the legislation. The complexity of the volatile composition has been proven by the fact that 57 compounds represented mainly by esters (ethyl decanoate and ethyl dodecanoate) and alcohols (3-methyl-1-butanol) were found.

Keywords: Distilled alcoholic beverages; Quality control; Legislation; Standardization.

Resumen

Producida y comercializada en todo Brasil, la cachaça es un símbolo del país. Para garantizar la seguridad de los consumidores, la calidad de esta bebida debe cumplir con los estándares legales establecidos por la legislación brasileña. A pesar de eso, todavía son comunes informes de inconformidad a los estándares de calidad. Además, se requieren estudios más detallados debido a la complejidad de la composición de la cachaza. En este trabajo se buscó evaluar la composición química de las cachaças producidas en el estado de Paraíba, determinar si estaban en conformidad con los estándares exigidos por la legislación e identificar los compuestos volátiles presentes en tales bebidas. Fueron obtenidas muestras comerciales con sus empaques originales y enviadas al Laboratório de Análise de Qualidade de Aguardente de la Universidade Federal de Lavras, Minas Gerais. Se tomaron como indicadores de calidad los parámetros establecidos por la legislación brasileña. Los compuestos volátiles fueron identificados por la técnica de cromatografía gaseosa acoplada a espectrometría de masas (GC-MS). Entre las 20 muestras analizadas, 17 estaban fuera de los patrones en por lo menos uno de los parámetros establecidos por la legislación. En cuanto a la composición química, se encontraron 57 compuestos representados mayoritariamente por ésteres (decanoato de etilo e dodecanoato de etilo) y alcoholes (3-metil-1-butanol).

Palabras clave: Bebidas alcohólicas y destiladas; Control de calidad; Legislación; Estandarización.

1.Introduction

Originating from a wide range of raw materials, distilled or fermented-distilled beverages are produced and consumed worldwide since ancient times, and they have chemical and sensory characteristics peculiar to each product that depend on the raw material used. The production process basically involves the formation of compounds during fermentation and

distillation reactions, where they are separated by their boiling point temperatures. The distillation stage can be performed using different industrial types (continuous or discontinuous), and these are factors that influence the final chemical composition of the beverages.

It has been shown that the presence of different classes of volatile compounds is essential for the quality and unique characteristics of distilled beverages. A similarity can be observed in the volatile composition of these beverages, which are formed mainly by esters, alcohols and carboxylic acids, which demonstrates the chemical complexity and the importance of these compounds, mainly with regard to sensory characteristics. This conclusion is the result of a series of studies already accomplished that sought to identify the main classes of volatile compounds present in various types of distilled beverages such as whiskey (Jeleń, et al., 2019), Cognac (Awad, et al., 2017), Rum (Pino, et al., 2012), Biska (Čiča, et al., 2018), Mezcal (Vera-Guzmán, et al., 2018), Lozovača (Matijašević, et al., 2019), Raki (Darici, et al., 2019) and Cachaça (Santiago, et al., 2016).

Cachaça is the most widely consumed distilled beverage in Brazil, and the second most extensively produced alcoholic beverage. It is defined by Decree No. 6871 of June 4, 2009, as the typical and exclusive denomination for sugarcane spirits produced in Brazil, with an alcohol content of 38 to 48% v/v, at 20 ºC, obtained by the distillation of fermented must from sugarcane juice with peculiar sensory characteristics. Up to 6 g L^{-1} of sugars can be added (Brazil, 2009). Also, Normative Instruction No. 13 of June 29, 2005, of the Ministry of Agriculture, Livestock and Supply (MAPA) defines the Identity and Quality Standards (PIQs) that must be followed for marketing cachaça and distilled spirits in the national territory (Brazil, 2005a).

A series of studies have sought to evaluate the quality of cachaça produced in Brazil. The results indicated that improvements in the production processes of cachaças, a constant evaluation of quality, and compliance with the established legal criteria are still needed (Zacaroni, et al., 2011; Bortoletto & Alcarde, 2015; Santiago, et al., 2015; Serafim, et al., 2016; Duarte, et al., 2017). Despite the efforts of various public and private agencies, the lack of standardization coupled with the informality still present in the cachaça production chain are unfavorable points in the evolution of quality and increase in the amount of this beverage exported. Similarly, the presence of contaminants, especially ethyl carbamate (EC), which has been proven to be carcinogenic, is also a barrier.

It is estimated that Brazil produces approximately 800 million liters of cachaça per year, despite having a capacity for over 1 billion liters (Ibrac, 2019). Paraíba is one of the main producing states, representing a significant market located in the Northeast of the country. Studies already carried out with cachaça from this state have shown that some physicochemical parameters were within the limits established by legislation (Lima & Nóbrega, 2004; Nóbrega, et al., 2009; Silva, et al., 2014). An up-to-date assessment of the cachaça produced in this state is necessary to ensure consumer safety and to contribute to a universal overview of the quality of distilled beverages. The objective of this research was to evaluate the chemical composition of cachaça produced in the state of Paraíba, observing the conformity to the established legal parameters, and the identification of volatile compounds present in these beverages.

2. Material and Methods

A quantitative exploratory research as oriented by Pereira et al. (2018) was performed via laboratory experiments during the period from April to June of 2019.

Samples of 20 unaged cachaças produced in the state of Paraiba were randomly collected in the local commerce, coded and sent to the Laboratório de Análise de Qualidade de Aguardente of the Universidade Federal de Lavras, in Lavras, Minas Gerais, Brazil.

That samples were stored at room temperature until the time for analysis. The physicochemical parameters established by the MAPA legislation were determined, and the volatile compounds were identified.

2.1 Analysis of the physicochemical parameters of cachaça

The analyses were performed according to the methods established by MAPA Normative Instruction No. 24 of September 08, 2005 (Brazil, 2005b), and by the use of chromatographic techniques. The parameters studied were those established by the legislation as quality indicators: the alcohol content, volatile acidity, esters, aldehydes, higher alcohols, furfural and hydroxymethylfurfural, methanol, butan-1-ol, butan-2-ol, copper, ethyl carbamate. and dry extract.

Methanol and higher alcohols were analyzed by gas chromatography equipped with a flame ionization detector (GC-FID) according to the method proposed by Santiago et al.

(2016). A DB Wax column (30 m x 0.25 mm, 0.25 μm) was used, and the separation was performed under the following conditions: split ratio (1:10), injector and detector temperatures (150 °C and 170 °C, respectively) and heating rate (1 °C min^{-1} from 55 °C to 70 °C, totaling 17 min). Helium (5.0) with a flow rate of 1.4 mL min^{-1} was used as the carrier gas. The identification of the compounds was performed by comparing the retention time of the samples against the standards, and the quantification was performed by the use of an external standard.

Furfural and HMF analyses were performed on a Shimadzu HPLC high performance liquid chromatograph equipped with two model SPD-M20A high pressure pumps, a model DGU-20A3 degasser, a model CBM-20A interface and a model SIL-10AF automatic injector. The column used for the separations was the Agilent - Zorbax Eclipse XDB-C18 (4.6 x 250 mm, 5 μm) connected to an Agilent-Zorbax Eclipse XDBC18 pre-column (4.6 x 12.5 mm, 5 μm) and the Diode Array (DAD) detector was used. The mobile phase was composed of 2% acetic acid solution in water (Solvent A) and methanol:water:acetic acid (70:28:2% v/v/v; Solvent B). The wavelength used was 280 nm, the flow rate was 0.8 mL min^{-1}, and the injected volume was 20 μL.

The method described by Santiago et al. (2017) was used for the determination of ethyl carbamate. It consists of prior derivatization of the sample and analysis by high performance liquid chromatography (HPLC). Analyses were performed on the same HPLC as that described above, but a model RF-10AXL fluorescence detector (FLD) was used.

Separations were performed using an Agilent-Zorbax Eclipse AAA column (4.6 x 150 mm, 5 μm) connected to an Agilent-Zorbax Eclipse AAA guard column (4.6 x 12.5 mm, 5 μm). Quantification of ethyl carbamate was performed using the external standard method. The excitation and emission wavelengths employed were 233 and 600 nm, respectively.

Flow rate used throughout the analysis was 0.75 mL min^{-1}, and the injected volume of samples and standard was 20 μL. Elution was performed in a gradient type system: 0 to 5 min (40-60% B); 5 to 10 min (60-70% B); 10 to 18 min (70-80% B); 18 to 19.5 min (80-90% B); 19.5 to 25 min (90-40% B); 25 to 30 min (40% B). The mobile phase was composed of 20 mmol L^{-1} sodium acetate solution (Solvent A) and acetonitrile (Solvent B).

2.2 Determination of volatile compounds

The analysis of the volatile compounds in cachaça was performed by gas chromatography coupled with mass spectrometry (GC-MS) according to the method proposed by Zacaroni et al. (2017). Volatile extractions were performed using the headspace solid phase microextraction (SPME) technique. The samples were diluted to a concentration of 10%. After dilution, 4-mL aliquots of samples were added to a 20-mL vial. Extraction was performed using a DVB/CAR/PDMS fiber (Divinylbenzene, Carboxen, and Polydimethylsiloxane) with a 30-50 µm Supelco film thickness. This fiber was previously conditioned for 1 h at 270 ºC in accordance with the specifications. The extraction temperature was 45 °C, the extraction time was 50 minutes, and the desorption time was 3 minutes.

A gas chromatograph coupled to a GC-MS QP2010 Plus mass spectrometer (Shimadzu, Japan) equipped with a AOC-5000 automatic liquid and gas injector (Shimadzu, Japan, a split/splitless injector and a 30 m × 0.25 mm; 0.25 µm SLBTM column (5% phenyl - 95% dimethylsiloxane) were used. The oven temperature was varied from 35 °C to 240 °C using a 4 °C min^{-1} heating rate. The carrier gas used was He 5.0, with a flow rate of 1.78 mL min^{-1}. The injector temperature was maintained at 270 °C and operated in 1:4 split mode. The compounds were detected by electron impact mass spectrometry at 70 eV in scan mode (29 to 600 Da). The temperature of the detector interface and the ion source remained at 240 °C and 200 °C, respectively. The filament was turned on at 1.75 min (Zacaroni, et al., 2017).

The compounds were identified by comparing the mass spectrum obtained with those of the GC-MS spectral library (Wiley 8 and FFNSC 1.2). Experimental retention indices were calculated, and they were compared with indices reported in the literature (Adams, 2017; National Institute of Standards and Technology, NIST, 2019). For comparison, similarities above 70% were considered to be identical, and the experimental indices were calculated using a homologous series of alkanes (Zacaroni, et al., 2017).

2.3 Statistical design

The completely randomized design (CRD) was used. The data obtained were normalized and subjected to analysis of variance, and the means were compared by the Kruskal-Wallis test at a 95% confidence level ($p \leq 0.05$) using the R® statistical program.

Principal component analysis (PCA) was also applied using the CHEMOFACE program (Nunes, et al., 2012) to ascertain and understand the similarity between the samples using the values found in the physicochemical and volatile compounds analyses. For the analysis of volatile compounds in PCA, the chromatographic peak areas were used.

3. Results and Discussion

3.1 Compliance with legal parameters

The physicochemical parameters of the evaluated cachaça are presented in Table 1. The alcohol content was the parameter that concentrated the largest number of samples that lied outside the limits required by the legislation. In most cases the lack of conformity was the result of a low alcohol concentration of up to 34.12% v/v, as found in sample I, or a concentration greater than the legal limit (38 - 48% v/v) in sample C. The latter can still be classified as “spirit”, a product that has a minimum and maximum alcohol concentration of 38-54% v/v, respectively (Brazil, 2005a). Problems related to inadequate alcohol content can be associated with errors in the distillation process or in the standardization of the beverage for commercialization (Cardoso, 2013).

Most samples (80%) contained volatile acidity concentrations within the required range. However, a value greater than twice the limit was observed for sample T, which reached 355.50 mg 100 mL^{-1} a.a. (anhydrous alcohol) and corroborated that of Silva et al. (2014), who found a maximum value of 324 mg 100 mL^{-1} a.a. in samples of cachaça from stills in Paraiba. The same authors observed that most of their samples did not match the limits required for this parameter. Lelis et al. (2014) evaluated cachaças produced in five Brazilian states and found average values of volatile acidity between 33.64 and 144.20 mg 100 mL^{-1} a.a. For Odello et al. (2009), Serafim et al. (2013) and Amorim et al. (2016), high levels of acidity are usually related to the contamination of must during fermentation, one of the most influential steps in the quality of the drink, resulting not only in the inadequacy of the limit, but also the low sensorial acceptance of cachaça.

Esters are the compounds with the greatest sensory impact in a distilled beverage. They stand out in the formation of fruity aromas. All the concentrations found in the cachaça for this parameter were within the limits required by the legislation. It is noteworthy that there was a significant variation between the lowest and highest values found (9.17 and 172.72 mg

100 mL^{-1} a.a.). Santiago et al. (2016) found an average ester concentration of 9.64 mg 100 mL^{-1} a.a. in the heart fraction of cachaça. Bortoletto and Alcarde (2015) observed that ester concentrations were lower than 100 mg 100 mL^{-1} a.a. in 95.9% of the 268 cachaça and spirits samples analyzed. Variations in the concentrations found for this parameter can be explained by the fact that the formation of esters occurs biochemically in the intracellular medium of yeast strains during the fermentative process or via esterification reactions during storage/aging, thus conditioning the practices exercised during the production process (Cardoso, 2013; Alcarde, 2017; Palmer, 2016).

Only one sample (K) was outside the established limit for aldehydes, with 33.88 mg 100 mL^{-1} a.a. Acetaldehyde is the main representative of these compounds, and their formation occurs in the preliminary stages of the fermentation process, mainly as a result of excessive must aeration or oxidation of higher alcohols (Cardoso, 2013; Alcarde, 2017). Some aldehydes are considered contaminants, and their isolated values should be analyzed as furfural and hydroxymethylfurfural (HMF) (Brazil, 2005a). The presence of HMF was not identified in this study. The highest furfural value, at 18.23 mg 100 mL^{-1} a.a,. was observed for sample P. This concentration was three times the maximum legal limit (5 mg 100 mL^{-1} a.a.). The pathways for the formation of furfural and HMF in cachaça are associated with the burning of sugarcane used as raw material, in which the pentoses and hexoses are degraded, or by the presence of sugar and yeast residues and overheating during distillation (Masson, et al. 2007; Cardoso, 2013; Tsakiris, et al. 2016).

Higher alcohols are composed of isopropyl, isoamyl and isobutyl alcohols, and they are the principal secondary components. All the samples were within the established limit (360 mg 100 mL^{-1} a.a.). The maximum concentration of higher alcohols found was 310.43 mg 100 mL^{-1} a.a. in the sample T. Santiago et al. (2017) observed that 155.41 mg of higher alcohols were found per 100 mL^{-1} a.a. Duarte et al. (2017) found a maximum value of 312.03 mg 100 mL^{-1} a.a. of higher alcohols in organic cachaça. One of the pathways for the formation of higher alcohols involves the degradation of amino acids during the fermentation process. Alcohols with up to five carbon atoms contribute to the characteristic floral aroma of the beverage. However, there is a substantial modification in aroma with the increase in the length of the carbon chain, which favors the emergence, when in excess, of a mixture known as “fusel oil” and results in a lower commercial value and beverage quality (Cardoso, 2013).

Secondary components are defined as the sum of volatile substances (volatile acidity, aldehydes, esters, furfural and HMF and the higher alcohols) present in cachaça, and they are

jointly responsible for the sensory characteristics of the beverage. Two samples were found to be out of the range for this parameter. The value for sample T was above the limit, with 874.98 mg 100 mL^{-1} a.a. The value was probably associated with its high values for volatile acidity, esters and higher alcohols. Some alternatives for adjusting this parameter can be used, such as filtration or redistillation, but according to Silva et al. (2013) and Zacaroni et al. (2015), such processes cause changes in the chemical composition, and, consequently, they can reduce the sensory quality of cachaça.

The dried extract corresponds to the solid material obtained after evaporation of the volatile fraction of the beverage. No limit is set for this parameter. Sample H obtained the highest value, 6.21 g L^{-1}. This value can be related to the addition of sugar during the preparation of the beverage. According to the legislation, samples with values greater than 6 g L^{-1} and less than 30 g L^{-1} are called “sweetened” (Brazil, 2005a). Although considered to be legal by Brazilian law, the addition of sugar in cachaça and spirits might be associated with the concealment of sensory defects perceived by the consumer, making it an unrecommended practice.

The results found for the presence of contaminants in the cachaça are presented in Table 2. Only the concentration of copper, an inorganic contaminant, was not within the legal limits. The concentration of copper (18.36 mg L^{-1}) in the sample T was three times the legal limit (5 mg L^{-1}).

Contamination of cachaça by copper generally occurs because the stills are built with this metal. Cardoso (2013) points out that the presence of copper promotes sensory benefits to cachaça by catalyzing reactions that decrease the presence of sulfur compounds. However, when present in excess, copper can cause serious problems for the human organism (Coffey et al., 2013). Alternatives have been proposed to reduce or eliminate this metal from cachaça after distillation, such as the use of activated carbon filters, ion exchange resins and natural adsorbents, and satisfactory results have been obtained (Lima, et al., 2009; Duarte, et al., 2014; Zacaroni, et al., 2015).

Methanol, butan-1-ol and butan-2-ol are considered by law to be contaminants in cachaça. No values above the limits established for any of the alcohols were detected. The formation of methanol occurs through the degradation of the pectin present in sugarcane during the fermentation process and subsequent distillation (Cardoso, 2013). Alcarde et al. (2010) and Santiago et al. (2016) observed that the presence of methanol is associated with beverage fractions, being more expressive in the “head” fraction. This observation reinforces

the importance of efficient disposal of these fractions during distillation. The presence of butan-1-ol and butan-2-ol in cachaça is due to contamination by the bacterium Clostridium acetobutylicum during fermentation, and their toxicities are relatively high. Thus, the necessary care with the implementation of Good Manufacturing Practices in the production unit is the main mechanism of quality control (Cardoso, 2013).

Table 1 shows physicochemical parameters of Paraiban cachaça.

Table 1. Physicochemical parameters evaluated in the cachaça samples from the state of Paraiba*

Sample	Parameters								
	Percent alcohol[1]	Volatile acidity[2]	Esters[2]	Aldehydes[2]	Furfural	HMF	Higher alcohols[2]	Secondary cpds[2]	Dry extract[3]
A	38.76±0.02(j)	**173.71**±1.75(c)	74.19±0.05(g)	4.31±0.28(q)	1.38±0.01(p)	nd	117.99±4.47(i)	371.58±6.47(e)	0.11±0.01(f)
B	47.42±0.08(b)	56.49±1.62(k)	75.80±1.02(f)	15.04±0.02(f)	3.89±0.01(i)	nd	201.14±7.74(e)	352.36±2.08(f)	0.07±0.01(f)
C	**52.56**±0.08(a)	**157.03**±0.25(d)	181.83±1.31(b)	11.66±0.19(i)	2.26±0.01(m)	nd	79.96±0.39(jk)	432.74±0.85(c)	0.12±0.00(f)
D	42.35±0.03(c)	85.48±0.07(h)	88.87±1.07(e)	24.21±0.02(b)	2.55±0.01(l)	nd	251.52±0.92(ab)	452.63±0.25(b)	0.08±0.00(f)
E	41.88±0.04(e)	100.25±3.36(f)	64.62±0.06(h)	11.44±0.28(i)	0.96±0.01(r)	nd	164.48±1.05(g)	341.75±4.08(g)	0.23±0.18(e)
F	41.20±0.01(f)	43.93±1.74(m)	48.24±1.04(i)	3.79±0.00(r)	0.77±0.00(s)	nd	86.34±2.46(j)	**183.07**±1.77(p)	0.02±0.02(g)
G	38.30±0.07(k)	69.70±1.86(j)	9.17±0.02(s)	12.94±0.26(h)	1.87±0.00(o)	nd	163.71±1.38(g)	257.39±0.72(n)	0.02±0.00(g)
H	**35.82**±0.11(o)	36.88±0.11(p)	13.49±1.26(r)	19.22±0.35(c)	nd	nd	215.65±3.00(d)	285.24±4.73(k)	6.21±0.01(a)
I	**34.12**±0.01(t)	51.63±0.02(l)	24.45±1.29(q)	8.38±0.31(l)	2.94±0.01(j)	nd	220.53±1.23(c)	307.93±0.21(i)	0.05±0.01(g)
J	39.40±0.01(h)	44.70±0.01(m)	43.46±1.12(k)	6.18±0.27(o)	4.99±0.02(g)	nd	183.34±0.72(e)	282.67±2.15(k)	0.48±0.03(d)
K	39.38±0.05(h)	76.40±1.76(i)	45.72±1.17(j)	**33.88**±0.04(a)	4.49±0.01(h)	nd	179.94±2.78(f)	340.43±3.32(g)	0.01±0.00(g)
L	42.07±0.03(d)	17.45±0.01(r)	31.31±0.02(p)	8.81±0.24(k)	**10.65**±0.03(c)	nd	213.87±0.75(d)	282.09±0.93(k)	0.01±0.01(g)
M	38.85±0.06(i)	39.68±1.95(o)	33.91±0.06(n)	14.45±0.25(g)	1.16±0.01(q)	nd	239.44±1.76(bc)	328.64±3.53(h)	0.15±0.01(e)
N	39.49±0.05(g)	27.88±1.89(q)	33.36±0.04(o)	17.16±0.02(e)	**5.70**±0.01(f)	nd	213.66±0.51(d)	297.13±1.44(j)	4.84±0.0(b)
O	**36.93**±0.11(m)	39.74±0.11(o)	40.42±0.12(l)	18.07±0.34(d)	2.83±0.01(k)	nd	162.10±3.01(g)	263.16±3.58(m)	4.39±0.03(c)
P	**35.31**±0.02(p)	149.66±0.10(e)	95.76±1.31(d)	10.20±0.61(j)	**18.23**±0.04(a)	nd	152.50±0.75(h)	426.35±1.31(d)	0.11±0.03(f)
Q	38.24±0.00(k)	92.12±0.00(g)	34.45±0.00(m)	6.92±0.27(n)	1.89±0.00(n)	nd	76.50±1.26(k)	211.88±1.54(o)	0.10±0.01(f)
R	**36.71**±0.10(n)	**217.90**±1.41(b)	104.05±3.30(c)	8.94±0.26(k)	**5.95**±0.00(e)	nd	114.14±1.76(i)	450.98±6.74(b)	0.18±0.01(e)
S	**35.12**±0.01(q)	41.80±0.01(n)	38.76±3.74(l)	7.54±0.30(m)	**6.15**±0.01(d)	nd	179.56±1.41(f)	273.81±5.47(l)	0.05±0.01(g)
T	**37.37**±0.01(l)	**355.50**±1.82(a)	192.72±0.07(a)	5.10±0.00(p)	**11.23**±0.01(b)	nd	310.43±0.66(a)	**874.98**±1.10(a)	0.23±0.01(e)
Limit**	38-48	150	200	30	5***		360	200-650	--

* Means followed by the same letters in columns do not differ by the Kruskal-Wallis test ($p \leq 0.05$); 1 - % v/v; 2 - mg 100 mL^{-1} a.a.; 3 - g L^{-1}; nd – Not detected; HMF – Hydroxymethylfurfural; **Brazil (2005a); ***Sum of furfural and hydroxymethylfurfural. Source: Autor (2020).

All the samples were within the limits for the ethyl carbamate (EC) contaminant. This substance is considered to be a compound with high carcinogenic potential, and it represents a direct risk to human health. The limits for this contaminant in cachaça were established by legislation at 210 µg L^{-1} (Lachenmeier, et al., 2010; Brazil, 2014). An evolution in the control of EC in the cachaça from Paraíba has occurred. Nobrega et al. (2009) evaluated 25 samples of cachaça from the same state and observed that more than 70% contained concentrations of EC that were outside the limit, at that time established at 150 µg L^{-1}, reaching 700 µg L^{-1} of this contaminant. This improvement was also reported by Serafim et al. (2016), who found an average value of 46.4 µg L^{-1} of EC in cachaça from this state. Although widely studied, the routes of formation of ethyl carbamate are not yet well understood.

One of the most likely ways of formation in cachaça or distilled beverages occurs through the reaction of ethanol with cyanide groups that originated from the decomposition of cyanogenic glycosides present in the raw material (Nóbrega, et al., 2011; Galinaro, et al., 2015; Gowd, et al., 2018; Cravo, et al., 2019). Table 2 shows organic and inorganic content in chachaça samples.

Table 2. Inorganic and organic contaminants evaluated in the cachaça samples from the state of Paraíba*

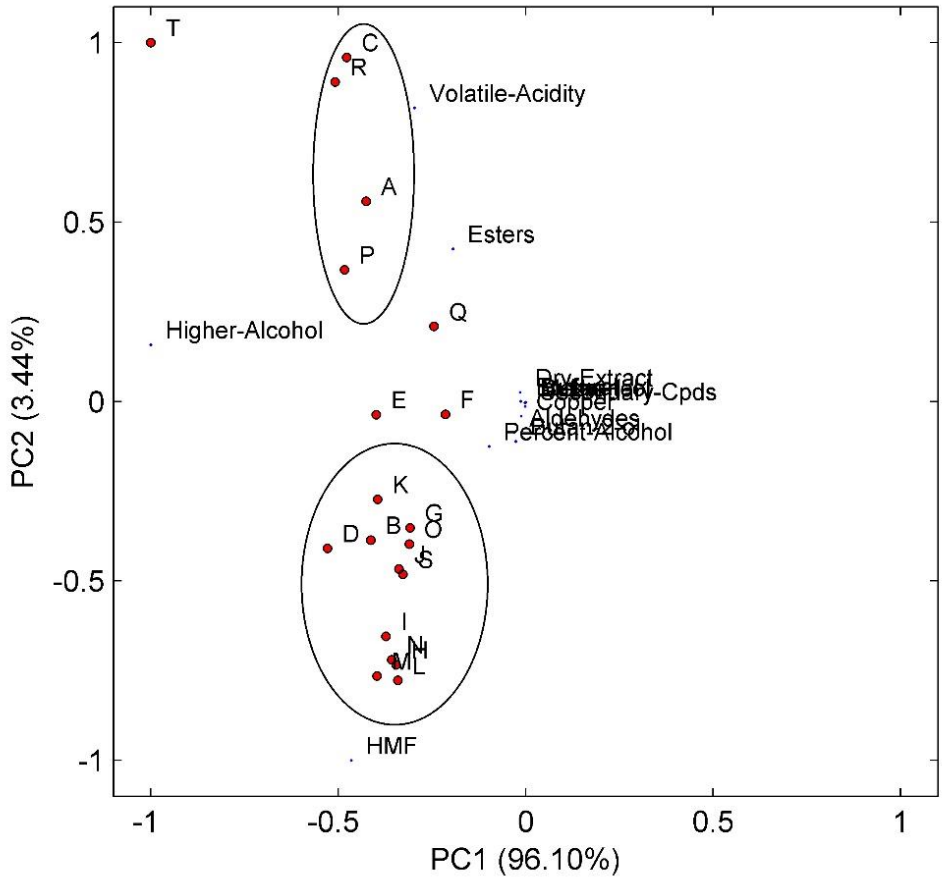
Samples	Parameters				
	Copper[1]	Methanol[2]	Butan-1-ol[2]	Butan-2-ol[2]	Ethyl carbamate
A	**5.**12 ± 0.02(e)	0.32 ± 0.01(pq)	0.81 ± 0.01(c)	0.46 ± 0.00(b)	5.89 ± 0.43(j)
B	1.30 ± 0.01(k)	0.61 ± 0.01(no)	0.75 ± 0.01(d)	nd	7.12 ± 0.09(i)
C	**7.48** ± 0.07(b)	nd	0.58 ± 0.01(g)	0.38 ± 0.01(c)	2.40 ± 0.29(l)
D	**7.45** ± 0.05(b)	0.48 ± 0.00(op)	0.62 ± 0.01(f)	nd	1.39 ± 0.20(m)
E	**7.49** ± 0.10(b)	0.88 ± 0.00(mn)	1.62 ± 0.01(a)	nd	0.48 ± 0.00(n)
F	2.77 ± 0.01(h)	2.16 ± 0.35(lm)	0.58 ± 0.02(g)	0.44 ± 0.01(b)	3.95 ± 0.06(k)
G	2.27 ± 0.04(i)	5.97 ± 0.15(ef)	0.43 ± 0.00(i)	nd	2.07 ± 0.29(l)
H	1.23 ± 0.01(k)	9.55 ± 2.23(b)	0.78 ± 0.01(c)	3.24 ± 0.11(a)	13.18 ± 0.13(f)
I	**5.64** ± 0.05(d)	9.83 ± 0.01(a)	0.64 ± 0.08(f)	nd	24.68 ± 0.46(c)
J	3.39 ± 0.17(g)	4.97 ± 0.00(kl)	0.71 ± 0.01(e)	nd	13.18 ± 0.13(f)
K	**5.16** ± 0.07(e)	7.56 ± 0.40(c)	0.29 ± 0.01(k)	nd	7.46 ± 0.32(i)
L	1.83 ± 0.02(j)	6.81 ± 0.13(d)	0.48 ± 0.01(h)	nd	29.32 ± 0.54(b)
M	1.12 ± 0.01(l)	5.04 ± 0.01(jk)	nd	nd	17.14 ± 0.42(e)
N	1.36 ± 0.01(k)	9.00 ± 0.60(b)	0.62 ± 0.01(f)	nd	31.61 ± 0.04(a)
O	1.01 ± 0.07(l)	5.30 ± 0.01(gh)	0.08 ± 0.01(l)	nd	nd
P	**6.86** ± 0.03(c)	6.14 ± 0.01(de)	0.08 ± 0.01(l)	nd	10.47 ± 0.46(g)

Q	**5.66** ± 0.06(d)	5.12 ± 0.00(ij)	0.05 ± 0.00(l)	nd	7.70 ± 0.76(i)
R	4.40 ± 0.06(f)	5.34 ± 0.01(fg)	0.04 ± 0.01(m)	nd	8.48 ± 0.19(h)
S	3.45 ± 0.17(g)	9.00 ± 0.04(b)	1.44 ± 0.01(b)	nd	24.03 ± 0.52(d)
T	**18.36** ± 0.40(a)	5.23 ± 0.01(hi)	0.37 ± 0.00(j)	nd	10.63 ± 0.04(g)
Limit**	5	20	3	10	210

* Means followed by the same letters in columns do not differ by the Kruskal-Wallis test ($p \leq 0,05$); 1 - mg L^{-1}; 2 - mg 100 mL^{-1} a.a.; 3 - µg L^{-1}; nd - Not detected; **Brazil (2005a). Source: Autor (2020).

Main component analysis was applied, and it was possible to describe 99.54% of the data. The first component described 96.10% of the data (Figure 1).

Figure 1. PC1 x PC2 biplot graph of loadings and scores regarding the physicochemical composition of cachaça from the state of Paraiba.



Source: Autor (2020).

Two distinct groups were observed because of the physicochemical characteristics of the cachaça. The groupings were mainly represented by volatile acidity, esters and HMF values.

Volatile acidity was the main differentiating parameter for group A. Sample T behaved as an outlier, probably associated with the high values found for higher alcohols. The other compounds showed similarities for all the samples.

3.2 Volatile compounds present in cachaça

The volatile compounds found in the cachaça samples are presented in Table 3. We found 57 different compounds, most of them belonging to the classes of esters and alcohols. Some compounds were similar for most samples. Ethyl acetate, ethyl octanoate, ethyl decanoate, ethyl dodecanoate, ethyl tetradecanoate and ethyl hexadecanoate were the most abundant esters. Some authors have identified the principal ester present in cachaça samples to be ethyl acetate (Nascimento, et al., 2009; Serafim & Franco 2015). However, the highest percentages found in this study were for ethyl decanoate and ethyl dodecanoate, in agreement with the results obtained by Zacaroni et al. (2017).

The results also corroborate one of the first studies of the composition of volatile compounds in cachaça produced in the state of Paraiba (Nóbrega, 2003). The authors concluded that the principal compounds of the volatile fraction of cachaça are esters, represented mainly by ethyl decanoate and ethyl octanoate, which are responsible for important sensory characteristics of the beverage.

Formation of ethyl esters is usually associated with the enzymatic fermentation process involving the synthesis or degradation of fatty acids and reactions between alcohols and acids. Some factors might modify the production of these compounds by yeast, especially the strain of yeast, the chemical composition of the medium and the fermentation conditions (aeration, hydrostatic pressure and temperature) (Saerens, et al., 2008; Alves, et al., 2010).

In fact, a number of variables can affect the intracellular environment of yeast, interfere with the production of volatile compounds and cause a change in the sensory profile of the beverage. Serafim and Franco (2015) evaluated cachaça produced by different yeasts (selected, wild and mixed) and types of distillation (column and still) and proved that the volatile composition is influenced by the type of production, highlighting ethyl acetate as the principal ester.

Amorim et al. (2016) found that the main ester produced in fermented cachaça with a pure culture of *S. cerevisae* and an inoculum mix was ethyl hexanoate, which was also a principal ester found in this study. Franitza et al. (2016) observed that the production steps substantially modify the concentrations of volatile compounds in rum, and fermentation is mainly responsible for the formation of esters and alcohols.

Among the alcohols found in this study, 3-methyl-1-butanol (isoamyl alcohol) stands out, although 2-methyl-1-butanol, phenethyl alcohol and nerolidol were also present in most of the samples. The majority presence of isoamyl alcohol corroborates the results found by Serafim and Franco (2015) and Zacaroni et al. (2017).

That alcohol is formed via secondary yeast metabolism by the degradation of sugars and amino acids (Nobrega, 2003). Nerolidol, a sesquiterpene, is also an alcohol that is constantly present in cachaça and other distilled beverages (Santiago, et al., 2016; Lopes, et al., 2019).

This terpene originates via the secondary metabolism of plants, and, although not produced during the beverage production stages, it is probably derived from the raw material and carried through during distillation. It contributes to the sensory characteristics related to floral and fruity aromas.

The profile of the volatile compounds in cachaça corresponded to that found in other distilled beverages. Pino et al. (2012) used CG-MS/CG-FID techniques to identify and quantify 116 volatile compounds in aged rum, of which 37 were esters, represented mainly by ethyl acetate, and 26 were alcohols, represented mainly by isoamyl alcohol. Čiča et al. (2018) evaluated the volatile composition of Biska, a distilled beverage produced in Croatia, by the SPME-CG-MS technique and found 166 compounds, including 53 esters and 32 alcohols.

Vera-Guzmán et al. (2018) evaluated Mezcal samples produced in Mexico from *A. angustifolia* and *A. potatorum*. They found 84 volatile compounds by the liquid-liquid extraction technique and identified them by GC-MS. The main volatile compounds were alcohols, esters and organic acids, as well as some phenols, ketones, furans, naphthalene and others.

Other unique compounds were identified in the samples, proving the complexity of the beverage. However, unlike the chemical composition of the aforementioned beverages, cachaça contained a series of compounds that can probably characterize it.

The biggest differences were concentrated in the presence of aromatic hydrocarbons. Some of these compounds had been identified in cachaça by Santiago et al. (2016), and they

can be indicated as a probable defect because these compounds are associated with unwanted sensory characteristics.

Their presence in cachaça can be attributed to contamination during the production process or from packaging. Nevertheless, there is evidence that the formation of aromatic compounds in post-fermented tea might stem from microbiological action involving gallic acid methylation (Lv et al., 2012).

Table 3. Volatile compounds identified in the cachaça samples from the state of Paraiba.

	Compound	IR_{Exp}	RL_{it}	N	Area (m)	Descriptor
Ester						
1	Ethyl acetate	-	606	14	6.38	Pineapple[a,d]
2	Ethyl butanoate	803	802	1	0.36	Pineapple[d]
3	Isoamyl acetate	876	876	11	0.37	Banana[a,c]
4	Ethyl hexanoate	999	997	15	0.47	Fruity[a], sweet[c]
5	Ethyl benzoate	1170	1169	6	0.79	Chamomile, floral[a], Fruity[a]
6	Diethyl succinate	1179	1176	6	0.35	Wine[a], fruity[a], floral[c], Sweet[c]
7	Methyl salicylate	1191	1190	13	1.77	Peppermint[a]
8	Ethyl octanoate	1196	1196	19	5.48	Fruity[a,b,c,d], sweet[b,c]
9	2-Phenethyl acetate	1254	1260	4	1.10	Floral[a,b,c,d], honey[a]
10	Ethyl salicylate	1268	1266	2	0.62	Minty[a]
11	Ethyl nonanoate	1273	-	5	0.37	Fruity[b,c,d]
12	Menthyl acetate	1290	1294	1	0.57	-
13	Isobutyl benzoate	1328	1327	1	0.44	-
14	Ethyl Decanoate	1394	1395	20	15.13	Grape[a,b,c], fruity[b,c,d]
15	Ethyl 9-hexadecenoate	1541	-	2	0.99	-
16	Ethyl dodecanoate	1593	1594	20	14.21	Fruity[b,c,d], sweet[b,c]
17	Isopentyl tetradecanoate	1647	-	5	0.82	-
18	Ethyl tetradecanoate	-	1795	18	4.14	-
19	Farnesyl acetate	-	-	1	1.48	-
20	Diisobutyl phthalate	-	-	12	2.89	-
21	Ethyl hexadecanoate	-	1902	20	6.95	Waxy[a], fruity[b,c,d]
22	Ethyl oleate	-	-	4	0.30	Fatty[d]
23	Ethyl linoleate	-	2159	4	0.47	Fatty[d]
Alcohol						
24	Isobutyl alcohol	-	-	20	1.98	Malty[d]
25	3-Methyl-1-butanol	719	723	20	21.93	Whisky[a], malty[a,d],

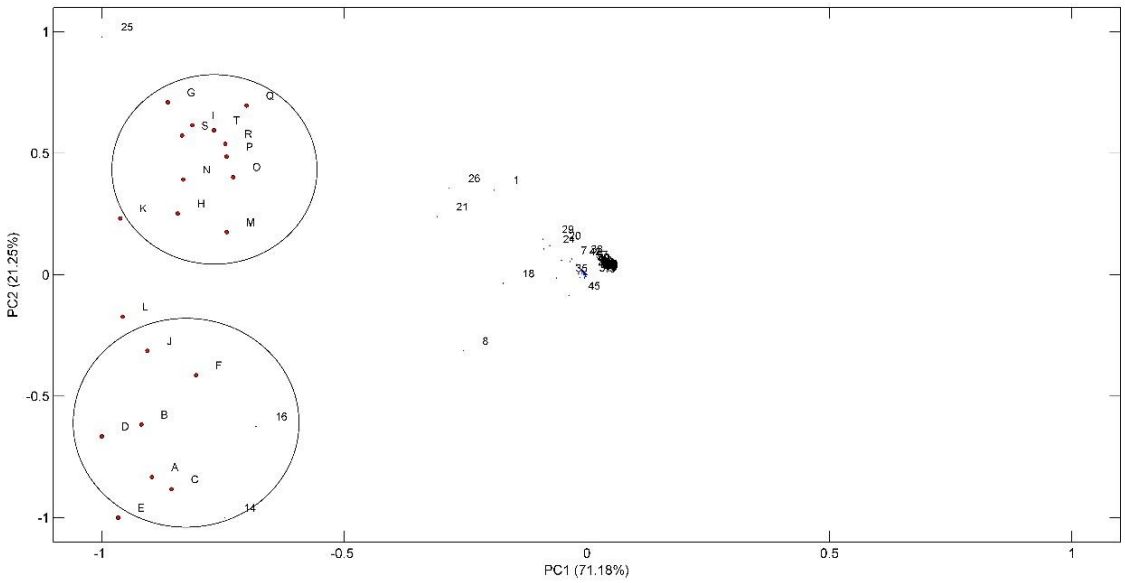
						Fruity[b], [b]
26	2-Methyl-1-butanol	720	724	18	6.98	Malty[a], fruity[b], sweet[b]
27	2,6-Dimethylheptan-4-ol	1056	-	1	1.04	-
28	Octanol	1073	1063	4	0.51	Fruity[b,c,d]; sweet[b]
29	Phenethyl alcohol	1113	1125	13	3.11	Honey[b], floral[b]
30	Nonanol	1156	1154	1	0.17	Fatty[a], green[a], Rasberry[b], floral[b]
31	Menthol	1161	1167	5	1.22	Peppermint[a]
32	4-Ethylphenol	1165	1169	1	0.87	-
33	4-Ethylguaiacol	1273	1287	1	0.42	Spicy[a], clove[a]
34	Dactylol	1560	-	1	1.1	-
35	Nerolidol	1561	1561	16	1.71	Woody[a], floral[a,b], Waxy[a], apple[b,c], rose[b,c]
36	2,3-Dihydrofarnesol	1687	1688	4	0.76	-
37	Farnesol <2Z,6Z>	1690	1698	1	0.33	Lemony [b], floral [b], honey[b]
Acids						
38	Acetic acid	-	600	11	1.33	Sour[a]
39	Caprylic acid	1175	1167	3	1.73	Sweaty[a], cheesy[a], rotten fruit[b], fatty[b], rancid[b]
40	Decanoic acid	1369	1364	6	1.19	Fatty[a,b,c], rancid[a,c],
41	Dodecanoic acid	1564	1565	1	1.20	Metalic[b,c], fatty[b]
Aromatic hydrocarbon						
42	Methylbenzene	762	773	13	1.24	Painty[a]
43	Ethenylbenzene	890	893	7	0.24	Balsamic[a], gasoline[a]
44	Trimethylbenzene	992	-	6	0.57	-
45	Trimethyl-tetrahydronaphthalene	1189	-	6	2.52	-
Aldehyde						
46	Furfural	831	828	5	0.66	Bread[a], almond[a,d], sweet[a,d]
47	Nonanal	1105	1100	3	0.32	Fatty[a], cítric[a,b], soapy[b]
48	Phellandral	1258	-	1	0.93	-
Ketone						
49	Menthone	1155	1148	1	2.79	Fresh[a], green[a]
50	5-Methyl-2-(1-methylethyl)-cyclohexanone	1178	-	6	1.35	-
51	β-Damascenone	1380	1386	7	0.26	Floral[d], apple[a], rose[a], honey[a]
Alkene						
52	Limonene	1028	1024	3	0.47	Lemony[a], orange[a], fruity[d],
53	2,2,6,7-tetramethylbicyclo[4.3.0]nona-4,7,9(1)-triene	1210	-	6	0.22	-
54	(E)-β-Farnesene	1453	1454	4	0.30	-

Eter						
55	5-Isopropyl-3,3-dimethyl-2-methylene-2,3-dihydrofuran	1273	-	2	0.75	-
Acetal						
56	1,1-Diethoxyethane	705	711	1	0.09	Fruity[b]
Alkyl chloride						
57	2-Chloro-2-methylpropane	738	-	1	10.54	-

RI_{exp} – Experimental retention index; RI_{lit} – Literature retention index; N – number of samples in which the compound was identified; (m) – mean percentage of area; a – Flavornet (2019); b - Lopes et al. (2019); c – Costa et al. (2018); d – Niu et al. (2017). Source: Autor (2020).

Through principal component analysis applied to volatile compounds, 92.43% of the compounds could be described, where 71.18% of the total variance was described by the first principal component (Figure 2).

Figure 2. PC1 x PC2 biplot graph of the loadings and scores for the volatile compounds in the cachaças from the state of Paraiba.



Source: Autor (2020).

Two groupings were identified by the analysis, where group A was characterized by the large percentage of compound 25 (3-methyl-1-butanol). Group B was distinguished by the presence of compound 16 (ethyl dodecanoate).

These results correspond to the high percentages of area that were attributed to these compounds when submitted to GC-MS analysis.

The principal component analysis allowed us to infer that these are the principal compounds for the characterization and differentiation between the samples.

4. Final Considerations

Most of the samples analyzed did not meet the Identity and Quality Standards for cachaça according to the current legislation. Main inadequacies found in this study were related to alcohol and copper concentrations.

Nonconformities are related to the inefficiency or nonexistence of the application of Good Manufacturing Practices by the producers.

The complexity of volatile compounds in the cachaça matrix has been demonstrated, and esters and alcohols are the principal compounds.

The cachaças produced in the state of Paraíba require improvements in the production process to adapt to the established norms. Further studies should be performed to assess the quality and ensure the continued supply of a safe product for consumers.

Acknowledgements

Authors thank the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPQ), Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG), and the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for their financial support and scholarship.

References

Adams, RP. (2017). *Identification of essential oil components by gas chromatography/mass spectrometry*. Gruver: Texensis Publishing.

Alcarde, AR. (2017). *Cachaça: ciência, tecnologia e arte*. São Paulo: Edgard Blücher.

Alcarde, AR., Souza, PA., & Belluco, AES. (2010). Volatilization kinetics of secondary compounds from sugarcane spirits during double distillation in rectifying still. *Scientia Agricola*, 67(3), 280-286.

Alves, J. A., Lima, L. C. O., Dias, D. R., Nunes, C. A. & Schwan, R. F. (2010). Effects of spontaneous and inoculated fermentation on the volatile profile of lychee (Litchi chinensis Sonn) fermented beverages. *International Journal of Food Science & Technology*, 45(11), 2358-2365.

Amorim, J. C., Schwan, R. F. & Duarte, W. F. (2016). Sugar cane spirit (cachaça): Effects of mixed inoculum of yeasts on the sensory and chemical characteristics. *Food Research International*, 85(1), 76-83.

Awad, P., Athès, V., Decloux, M. E., Ferrari, G., Snakkers, G., Raguenaud, P. & Giampaoli, P. (2017). Evolution of Volatile Compounds during the Distillation of Cognac Spirit. *Journal of Agricultural and Food Chemistry*, 65(35), 7736-7748.

Bortoletto, A. M., & Alcarde, A. R. (2015). Assessment of chemical quality of Brazilian sugar cane spirits and cachaças. *Food Control*, 54(1), 1-6.

Brazil, (2014). *Ministério da Agricultura, Pecuária e Abastecimento* (MAPA). Instrução Normativa n. 28, de 08 de agosto de 2014.

Brazil. (2009). *Ministério da Agricultura, Pecuária e Abastecimento*. Decreto nº 6871, de 04 de junho de 2009.

Brazil. (2005a). *Ministério da Agricultura, Pecuária e Abastecimento*. Instrução Normativa n. 13, de 29 de junho de 2005.

Brazil. (2005b). *Ministério da Agricultura, Pecuária e Abastecimento*. Instrução Normativa n. 24, de 08 de setembro de 2005.

Cardoso, M. G. (2013). *Produção de aguardente de cana*. Lavras: Editora UFLA.

Čiča, K.H., Rupert, M., Koczoń, P., Derewiaka, D., Gajdoš-Kljusurić, J., Petravić-Tominac, V., ... & Stanzer, D. (2018). Characterisation of flavour compounds in Biska - a herbal spirit produced with mistletoe. *Journal of the Institute of Brewing*, 125(1), 143-154.

Coffey, A.J., Durkie, M., Hague, S., McLay, K., Emmerson, J., Lo, C., Klaffke, S., … & Bandmann, O. (2013). A genetic study of Wilson's disease in the United Kingdom. *Brain*, 136(5), 1476-1487.

Costa, G. P., Nicolli, K. P., Welke, J. E., Manfroi, V. & Zini, C. A. (2018). Volatile profile of sparkling wines produced with the addition of mannoproteins or lees before second fermentation performed with free and immobilized yeasts. *Journal of the Brazilian Chemical Society*, 29(9), 1866-1875.

Cravo, F. D., Santiago, W. D., Lunguinho, A. S., Barbosa, R. B., Oliveira, R. E. S., Alvarenga, G.F., Santos, S.D., … & Cardoso, M. G. (2019). Composition of cachaças produced from five varieties of sugarcane and the correlation of the presence of dhurrin in the cane with that of ethyl carbamate in the product. *American Journal of Plant Sciences*, 10(2), 339-350.

Darici, M., Bergama, D. & Cabaroglu, T. (2019). Effect of triple pot still distillation on the volatile compositions during the Rakı production. *Journal of Food Processing and Preservation*, 43(6), 1-11.

Duarte, F. C., Cardoso, M. G., Santiago, W. D., Machado, A. M. R. & Nelson, D. L. (2017). Brazilian organic sugarcane spirits: physicochemical and chromatographic profile. *Revista Ciência Agronômica*, 48(2), 288-295.

Duarte, F.C., Cardoso, M.G., Magriotis, Z.M., Santiago, W.D., Mendonça, J.G.P. & Rodrigues. L.M.A. (2014). Removal of copper in cachaças using clays. *Ciência e Agrotecnologia*, 38(4), 382-389.

FLAVORNET. *Flavornet and human odor space*. Available at: http://www.flavornet.org/flavornet.html Accessed 15 August 2019.

Franitza, L., Granvogl, M. & Schieberle, P. (2016). Influence of the production process on the key aroma compounds of rum: from molasses to the spirit. *Journal of Agricultural and Food Chemistry*, 64(47), 9041-9053.

Galinaro, C. A., Ohe, T. H. K., Silva, A. C. H., Silva, S. C. and Franco, D. W. (2015). Cyanate as an active precursor of ethyl carbamate formation in sugar cane spirit. *Journal of Agricultural Food Chemistry*, 63(33), 7415-7420.

Gowd, V., Su, H., Karlovsky, P. & Chen, W. (2018). Ethyl carbamate: an emerging food and environmental toxicant. *Food Chemistry*, 248(1), 312-321.

IBRAC. *Instituto Brasileiro da Cachaça*. Available at: http://www.ibrac.net/index.php/servicos/estatisticas/mercado-interno Accessed 20 August 2019.

Jeleń, H. H., Majcher, M. & Szwengiel, A. (2019). Key odorants in peated malt whisky and its differentiation from other whisky types using profiling of flavor and volatile compounds. *LWT - Food Science and Technology*, 107(1), 56-63.

Lachenmeier, D. W., Lima, M. C. P., Nóbrega, I. C. C., Pereira, J. A. P., Kerr-Corrêa, F., Kanteres, F. & Rehm, J. (2010). Cancer risk assessment of ethyl carbamate in alcoholic beverages from Brazil with special consideration to the spirits cachaça and tiquira. *BMC Câncer*, 10(266), 1-15.

Lelis, V.G., Chaves, J.B.P., Dutra, M.B.L., Reis, R.S. and Lessa, J.B. (2014). Ocorrência de carbamato de etila em cachaças de alambique e em aguardentes industriais. *Revista Ceres*, 61(4), 467-474.

Lima, A. J. B., Cardoso, M. G., Guimarães, L. G. L., Lima, J. M. & Nelson, D. L. (2009). Efeito de substâncias empregadas para remoção de cobre sobre o teor decompostos secundários da cachaça. *Química Nova*, 32(4), 845-848.

Lima, A. K. S., & Nóbrega, I. C. C. (2004). Avaliação de parâmetros de qualidade em aguardentes de cana produzidas no Estado da Paraíba. *Boletim CEPPA*, 22(1), 79-103.

Lopes, A. C. A., Genisheva, Z., Nunes, J. A. R. & Duarte, W. F. (2019). Production and characterization of a new sweet sorghum distilled beverage. *Sugar Tech*, 21(6), p. 966-975.

Lv, H., Zhong, Q., Lin, Z., Wang, L., Tan, J., & Guo, L. (2012). Aroma characterisation of Pu-erh tea using headspace-solid phase microextraction combined with GC/MS and GC–olfactometry. *Food Chemistry*, 130(4), 1074-1081.

Masson, J., Cardoso, M. G., Vilela, F. J., Pimentel, F. A., Morais, A. R. & Anjos, J. P. (2007). Parâmetros físico-químicos e cromatográficos de aguardentes de cana queimada e não queimada. *Ciência e Agrotecnologia*, 31(6), 1805-1810.

Matijašević, S., Popović-Djordjević, J., Ristić, R., Ćirković, D., Ćirković, B. & Popović, T. (2019). Volatile aroma compounds of brandy 'Lozovača′ produced from Muscat table grapevine cultivars (Vitis vinifera L.). *Molecules*, 24(13), 1-15.

Nascimento, E. S. P., Cardoso, D. R. & Franco, D. W. (2009). Comparação de técnicas de determinação de ésteres em cachaça. *Química Nova*, 32(9), 2323-2327.

NIST. *National Institute of Standards and Technology*. Chemistry WebBook. NIST Standard Reference Database Number 69. Available at: https://webbook.nist.gov/chemistry/ Accessed 10 September 2019.

Niu, Y., Yao, Z., Xiao, Q., Xiao, Z., Ma, N. & Zhu, J. (2017). Characterization of the key aroma compounds in different light aroma type Chinese liquors by GC-olfactometry, GC-FPD, quantitative measurements, and aroma recombination. *Food Chemistry*, 233(1), 204-215.

Nóbrega, I. C. C. (2003). Análise dos compostos voláteis da aguardente de cana por concentração dinâmica do "headspace" e cromatografia gasosa-espectrometria de massas. *Ciência e Tecnologia de Alimentos*, 23(2), 210-216.

Nóbrega, I. C. C., Pereira, J. A. P., Paiva, J. E. & Lachenmeier, D. W. (2009). Ethyl carbamate in pot still cachaças (Brazilian sugar cane spirits): Influence of distillation and storage conditions. *Food Chemistry*, 117(4), 693-697.

Nóbrega, I. C. C., Pereira, J. A. P., Paiva, J. E. and Lachenmeier, D. W. (2011). Ethyl carbamate in cachaça (Brazilian sugarcane spirit): Extended survey confirms simple mitigation approaches in pot still distillation. *Food Chemistry*, 127(1), 1243-1247.

Nunes, C. A., Freitas, M. P., Pinheiro, A. C. M. & Bastos, S. C. (2012). Chemoface: a novel free user-friendly interface for chemometrics. *Journal of the Brazilian Chemical Society*, 23(11), 2003-2010.

Odello, L., Braceschi, G. P., Seixas, F. R. F., Silva, A. A., Galinaro, C. A. & Franco, D. W. (2009). Avaliação sensorial de cachaça. *Química Nova*, 32(7), 1839-1844.

Palmer, G. H. (2016). Beverages, Distilled. *Encyclopedia of Food Grains*, 3(1), 193-205.

Pino, J.A., Tolle, S., Gök, R. & Winterhalter, P. (2012). Characterisation of odour-active compounds in aged rum. *Food Chemistry*, 132(3), 1436-1441.

Saerens, S. M. G., Delvaux, F., Verstrepen, K. J., Dijck, P. V., Thevelein, J. M. & Delvaux, F.R. (2007). Parameters Affecting Ethyl Ester Production by Saccharomyces cerevisiae during Fermentation. *Applied and Environmental Microbiology*, 74(2), 454-461.

Santiago, W. D., Cardoso, M. G., Zacaroni, L. M., Rodrigues, L. M. A., Duarte, F. C., Ribeiro, C. F. S. & Nelson, D. L. (2015). Multivariate analysis for the characterization of physico-chemical profiles of cachaça produced in copper stills over a period of six years in Minas Gerais state. *Journal of the Institute of Brewing*, 121(2), 244-250.

Santiago, W. D., Cardoso, M. G., Santiago, J. A., Teixeira, M. L., Barbosa, R. B., Zacaroni, L. M., … and Nelson, D. L. (2016). Physicochemical profile and determination of volatile compounds in cachaça stored in new oak (*Quercus sp*.), amburana (*Amburana cearensis*),

jatoba (*Hymenaeae carbouril*), balsam (*Myroxylon peruiferum*) and peroba (*Paratecoma peroba*) casks by SPME-GC-MS. *Journal of The Institute of Brewing*, 122(4), 624-634.

Santiago, W. D., Cardoso, M. G., Lunguinho, A. S., Barbosa, R. B., Cravo, F. D., Gonçalves, G. S. & Nelson, D. L. (2017). Determination of ethyl carbamate in cachaça stored in newly made oak, amburana, jatobá, balsamo and peroba vats and in glass containers. *Journal of The Institute of Brewing*, 123(4), 572-578.

Serafim, F. A. T., Seixas, F. R. F., Silva, A. A., Galinaro, C. A., Nascimento, E. S. P., Buchviser, S. F., ... and Franco, D. W. (2013). Correlation between Chemical Composition and Sensory Properties of Brazilian Sugarcane Spirits (Cachaças). *Journal of the Brazilian Chemical Society*, 24(6), 973-982.

Serafim, F. A. T., & Franco, D. W. (2015). Chemical traceability of industrial and natural yeasts used in the production of Brazilian sugarcane spirits. *Journal of Food Composition and Analysis*, 38(1), 98-105.

Serafim, F. A. T., Pereira-filho, E. R. & Franco, D. W. 2016. Chemical data as markers of the geographical origins of sugarcane spirits. *Food Chemistry*, 196(1), 196-203.

Silva, F. A., Vendruscolo, F., Carvalho, W. R., Soares Júnior, M. S., Pinheiro, M. V. M. & Caliari, M. (2013). Influence of the number of distillations on the composition of organic sugarcane spirit. *Journal of the Institute of Brewing*, 119(3), 133-138.

Silva, M. J., Moreira, R. T., Coutinho, E. P., Cruz, G. R. B. & Sousa, J. R. T. (2014). Características físico-químicas e sensoriais de cachaças de alambiques produzidas na microrregião do Brejo Paraibano, PB. *Revista Brasileira de Produtos Agroindustriais*, 16(4), 445-451.

Tsakiris, A., Kallithraka, S. & Kourkoutas, Y. (2016). Brandy and Cognac: Manufacture and chemical composition. *Encyclopedia of Food and Health*, 462-468.

Vera-Guzmán, A. M., Guzmán-Gerónimo, R. I., López, M. G. & Chávez-Servi, J. L. (2018). Volatile compound profiles in mezcal spirits as influenced by agave species and production processes. *Beverages*, 4(1), 1-10.

Zacaroni, L. M., Cardoso, M. G., Saczk, A. A., Santiago, W. D., Anjos, J. P., Masson, J., ... & Nelson, D. L. (2011). Caracterização e quantificação de contaminantes em aguardentes de cana. *Química Nova*, 34(2), 320-324.

Zacaroni, L. M., Magriotis, Z. M., Cardoso, M. G., Santiago, W. D., Mendonça, J. G., Vieira, S. S. & Nelson, D. L. (2015). Natural clay and commercial activated charcoal: Properties and application for the removal of copper from cachaça. *Food Control*, 47(1), 536-544.

Zacaroni, L. M., Sales, P. F., Cardoso, M. G., Santiago, W. D. and Nelson, D. L. (2017). Response surface optimization of SPME extraction conditions for the analysis of volatile compounds in Brazilian sugar cane spirits by HS-SPME-GC-MS. *Journal of The Institute of Brewing*, 123(2), 226-231.

Porcentagem de contribuição de cada autor no manuscrito

Renan Elan da Silva Oliveira – 30%

Maria das Graças Cardoso - 20%

Wilder Douglas Santiago - 20%

Richard Bispo Barbosa - 10%

Gabriela Fontes Alvarenga – 10%

David Lee Nelson – 10%