

Cachaça sold in polyethylene terephthalate packaging: Determination of the physical-chemical profile, polycyclic aromatic hydrocarbons and ethyl carbamate

Cachaça comercializada em embalagem de polietileno tereftalato: Determinação do perfil físico-químico, hidrocarbonetos policíclicos aromáticos e carbamato de etila

Richard Bispo Barbosa¹, Gabriela Fontes Alvarenga², Vanuzia Rodrigues Fernandes Ferreira¹, Wilder Douglas Santiago¹, David Lee Nelson³, Maria das Graças Cardoso^{1*}

¹Universidade Federal de Lavras/UFLA, Departamento de Química/DQI, Lavras, MG, Brasil

²Universidade Federal de Lavras/UFLA, Departamento de Ciências dos Alimentos/DCA, Lavras, MG, Brasil

³Universidade Federal dos Vales do Jequitinhonha e Mucuri/UFVJM, Diamantina, MG, Brasil

*Corresponding author: mcardoso@ufla.br

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ABSTRACT

The production of cachaça in Brazil is a historical and cultural practice that has been gaining ground in the commercial sector with the marketing of its product with new labels, derivatives and different types of packaging for the beverage. Polyethylene Terephthalate (PET) packaging has been used by some producers because of its lower cost and greater durability for transport. The influence of storage on 15 samples of commercial cachaça packed in polymeric packaging was investigated. The physical-chemical quality and the presence of Polycyclic Aromatic Hydrocarbons (PAH's) in the beverages were evaluated. Of these samples, 60% contained concentrations of components that were outside the limits of the Quality Standards established by Ministry of Agriculture, Livestock and Supply (MAPA), and most of the samples contained low alcohol concentrations. The samples showed concentrations of ethyl carbamate below the limit established by MAPA (210 µg L⁻¹). All samples studied contained concentrations of PAHs higher than those found in the literature for cachaça stored in other types of packages. Contamination by PAHs can be associated with the thermal processes involved in the production of packages. Sample R12 contained the highest total concentration of the PAHs (20.90 µg L⁻¹), followed by samples R7 (17.23 µg L⁻¹) and R8 (17.61 µg L⁻¹). The mean concentration was 10.91 µg L⁻¹. Fluorene was the PAH found in the largest quantity with a concentration of 18.61 µg L⁻¹ in the R12 sample. Therefore, cachaças stored in PET packaging are unsuitable for consumption and commercialization, as they present high concentrations of contaminants.

Index terms: Beverage; contaminants; PET.

RESUMO

A produção de cachaça no Brasil é uma prática histórica e cultural que vem ganhando espaço no setor comercial com a comercialização de seu produto com novos rótulos, derivados e diferentes tipos de embalagens para a bebida. As embalagens de Polietileno Tereftalato (PET) têm sido utilizadas por alguns produtores devido ao seu menor custo e maior durabilidade para o transporte. A influência do armazenamento em 15 amostras de cachaça comercial acondicionada em embalagens poliméricas foi investigada. A qualidade físico-química e a presença de Hidrocarbonetos Policíclicos Aromáticos (HPA's) nas bebidas foram avaliadas. Destas amostras, 60% continham concentrações de componentes que estavam fora dos limites dos Padrões de Qualidade estabelecidos pelo Ministério da Agricultura, Pecuária e Abastecimento (MAPA), e a maioria das amostras continha baixas concentrações de álcool. As amostras apresentaram concentrações de carbamato de etila abaixo do limite estabelecido pelo MAPA (210 µg L⁻¹). Todas as amostras estudadas continham concentrações de HPA's superiores às encontradas na literatura para cachaça armazenada em outros tipos de embalagens. A contaminação por HPA's pode estar associada aos processos térmicos envolvidos na produção das embalagens. A amostra R12 continha a maior concentração total de HPA's (20,90 µg L⁻¹), seguida pelas amostras R7 (17,23 µg L⁻¹) and R8 (17,61 µg L⁻¹). A concentração média foi de 10,91 µg L⁻¹. O fluoreno foi o HPA encontrado em maior quantidade com concentração de 18,61 µg L⁻¹ na amostra R12. Portanto, as cachaças armazenadas em embalagens PET são impróprias para consumo e comercialização, pois apresentam altas concentrações de contaminantes.

Termos para indexação: Bebida; contaminantes; PET.

INTRODUCTION

Cachaça is a cultural symbol of Brazil; it has been produced since the colonization period. It was discovered when the slaves who work in the sugar cane mills realized that the leftover sugarcane juice fermented and gave rise to a different beverage. The news quickly reached the sugar mill owners, who improved the techniques for distilling the beverage, giving rise to cachaça. The popularization of the beverage in the country was so great that Portugal banned its production in 1659 because of competition with Portuguese wine. The ban lasted a short time and led to the Cachaça Revolt, which ended the ban by the crown in 1661 (Cardoso, 2020).

According to the Ministry of Agriculture, Livestock and Supply (MAPA), distilled sugarcane spirits is defined as the beverage produced by distilling sugarcane must. Cachaça is considered to be the Cane Spirit produced in Brazil, with an alcoholic strength of 38% v/v to 48% v/v at 20 °C, obtained by the distillation of the fermented sugarcane juice with peculiar sensory characteristics, to which up to 6 g L⁻¹ of sugar can be added (Brasil, 2005a).

The beverage has been commercialized in different ways using new types of packaging, labels, and lids. These changes are employed to increase the sales, as well as to reduce packaging costs. The use of glass bottles is the most traditional manner of marketing the beverage; however, the use of polymeric packaging has been an alternative because of its lower cost and durability during storage and transport. The polymeric packaging used is made of polyethylene terephthalate (PET) (Ethylene Poly terephthalate), which has greater resistance to physical shocks, and different models and sizes exist on the market. Storage, physical changes and beverage packaging have been the subject of research in different areas, such as the presence of different compounds that are considered contaminants and can be associated with migration from the plastic packaging to the beverage (Souza et al., 2022).

Polycyclic Aromatic Hydrocarbons (PAHs) formed from the degradation of organic molecules in thermal treatments are contaminants found in soil, plants, water and air. The control of the amount of these compounds is a topic of discussion in several countries because of their toxic and carcinogenic character. In thermal processes, radicals are formed from the degradation of organic structures. These radicals can react with one another to form cyclic structures that have high stability. These substances can definitively bind to DNA when in contact with the body and cause changes in cells (Lemmens et al., 2020; Singh; Agarwal, 2018). During the production of polymeric packaging, thermal processes are used to prepare the raw material and to mold the shape

of the packaging. These processes can be associated with the formation of PAHs and, consequently, the contamination of the stored beverage (Allassali et al., 2020).

This study sought to elucidate possible ways in which cachaça stored and marketed in PET packaging can be contaminated by PAHs. The discovery of contamination sources and their traceability is an important factor for ensuring that the product reaches the consumer in a safe manner and avoids exposure to unhealthy compounds.

MATERIAL AND METHODS

Cachaça samples

The samples of cachaça sold in 500 mL PET containers were acquired in the local commerce and distribution units in the Southeast region of Brazil. Two 500 mL containers were used for the analysis of each sample. Fifteen samples were obtained from different production units with different production methods (column and alembic), and they were coded from R1 to R15. The R1, R2, R14 and R15 samples were produced using the method of distillation through stainless steel columns. The remaining samples were produced by the method of distillation in copper alembics. These samples were sent to the Laboratory of Quality Analysis of Spirits of the Department of Chemistry (DQI) of the Federal University of Lavras (UFLA) in the state of Minas Gerais. All of the analyses were performed in triplicate, and the results are expressed as means ± standard deviation.

Physicochemical analysis of cachaça

The physicochemical analyses were performed according to the methods established by MAPA (Brasil, 2005b), wherein the dry extract, volatile acidity, alcohol concentration, aldehydes, esters, furfural, higher alcohols and copper were quantified in samples of cachaça. Analyses were performed with the beverage before and after the adsorption test.

Analysis of higher alcohols

The identification and quantification of higher alcohols was performed according to the method of Vilela et al. (2007) with some modifications. Analysis was performed by gas chromatography (Perkin Elmer, Clarus 580) using a flame ionization detector (FID) under the following experimental conditions: DB-WAX column (30 m x 0.25 mm id, film thickness, 0.25 µm); N₂ (White Martins, Rio de Janeiro, Brazil) was the carrier gas at a flow rate of 1.4 mL min⁻¹; injector temperature: 150 °C; detector temperature:

170 °C; volume of sample injected: 1.0 µL diluted; 1:10 split ratio. The temperature program started at 35 °C for four minutes, increased to 80 °C at a rate of 10 °C min⁻¹, held for 1 minute and then increased to 120 °C at a rate of 20 °C min⁻¹, where it was held for 1 minute. The temperature was then raised to 140 °C at a rate of 25 °C min⁻¹ and held for 30 seconds. The total run time was 13.48 min.

HPLC or chromatographic grade standards (butan-2-ol, propan-1-ol, 2-methylpropan-1-ol, butan-1-ol and 3-methylbutan-1-ol (Merck, Darmstadt, Germany) were used. The analytical curve (concentration range) was prepared from a solution with a concentration of 4 g L⁻¹ in 40% ethanol. The samples were distilled via a simple distillation process and analyzed.

Polycyclic Aromatic Hydrocarbons (PAHs)

The PAHs were identified and quantified by HPLC in a Shimadzu high efficiency liquid chromatograph equipped with two model LC-6AD high pressure pumps, a model DGU-20A3 degasser, a model CBM-20A interface and a model SIL⁻¹0AF automatic injector with sampler. The diode array detector (DAD) was used according to the method standardized by Souza et al. (2022).

The quantification of the PAHs was accomplished using an analytical curve previously constructed with the standards at concentrations that varied from 10 to 500 µg L⁻¹. For separations, an Agilent HX891706 column (4.6 x 250 mm, 5 µm) connected to an Agilent HX891706 guard column (4.6 x 12.5 mm, 5 µm) was used. Solvents for the mobile phase were water (Solvent A) and acetonitrile (Solvent B). Samples were eluted according to a gradient from 0 to 3 min (50%); 3 to 10 min (50 to 100%); 10 to 35 min (100%); 35 to 40 min (100 to 50%). The wavelength used was 254 nm, the flow rate was 0.8 mL min⁻¹, and the injected volume was 20 µL.

A stock solution with a concentration of 1000 µg L⁻¹ was prepared containing the analytically pure PAH standards (Merck and Sigma-Aldrich) naphthalene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo(a) anthracene, acephenanthylene and benzo(a) pyrene. Dilutions varying from 10 to 500 µg L⁻¹ were made in ethyl acetate for the construction of the analytical curve.

The sample was extracted by the Solid Phase Extraction (SPE) technique using an Agilent Sampli Q12 collector Manifold, in which samples were passed through 500 mg/6 mL cartridges (ODS-5 Octadecyl; 18% EC - Whatman). Each cartridge was activated with 5.0 mL of dichloromethane, 5.0 mL of methanol and 5.0 mL of Milli-Q water. After conditioning, aliquots of a solution containing

50.0 mL of the sample and 10 ml of acetonitrile were passed through the cartridge. Ethyl acetate was used to elute the analytes. The extract was concentrated on a Buchi R-14 rotavapor at 50 °C, and the final volume was completed to 1.0 mL with ethyl acetate (Machado et al., 2014).

Ethyl carbamate

Ethyl carbamate was quantified by HPLC according to the method proposed by Anjos et al. (2011) using the previously described chromatograph equipped with a model RF-10AXL fluorescence detector (DFL) and an Agilent-Zorbax Eclipse a.a.A column (4.6 x 150 mm, 5 µm) connected to an Agilent-Zorbax Eclipse a.a.A pre-column (4.6 x 12.5 mm, 5 µm). External standardization was used to quantify the contaminant. A stock solution of 10.0 mg L⁻¹ was prepared from ethyl N-xanthyl carbamate (CNXE) in ethyl acetate. In the construction of the analytical curve, eight points in the range of 15.0 - 210.0 µg L⁻¹ were used. The excitation and emission wavelengths employed were 233 and 600 nm, respectively. The samples were derivatized by adding 4 mL of cachaça sample to an amber flask, followed by the addition of 0.8 mL of a propanol solution of 0.02 mol L⁻¹ 9-xanthidrol. After gentle stirring, 0.4 ml of 1.5 mol L⁻¹ hydrochloric acid was added, and the mixture was stirred for one minute. The reaction mixture was kept at rest for 60 minutes and filtered through a 0.45 µm polyethylene membrane (Millipore).

Furfural and hydroxymethylfurfural

The quantification of furfural and hydroxymethylfurfural was performed by HPLC using a diode array detector (DAD) according to the method of Rodrigues et al. (2020), with minor modifications. The determination of furfural and hydroxymethylfurfural was achieved using an analytical curve previously constructed with the respective standards at concentrations that varied from 0.1 to 25 mg L⁻¹.

The separation was achieved on an Agilent-Zorbax Eclipse XDB-C18 column (4.6 x 250 mm, 5 µm) connected to an Agilent-Zorbax Eclipse XDBC18 (4.6 x 12.5 mm, 5 µm) pre-column. A diode array detector was employed. The mobile phase was composed of 2% acetic acid in water (Solvent A) and methanol: water: acetic acid (70:28:2% v/v/v) (Solvent B). The gradient varied from 0 to 25 min (0-40%B); 25 to 40 min (40 to 55% B); 40 to 43 min (55-60% B); 43 to 50 min (60-100%); 50 to 55 min (100-0% B); 55 to 60 min (0% B). The wavelength employed was 280 nm, the flow rate was 0.8 mL min⁻¹, and the volume injected was 20 µL. Samples and standards were filtered through a 0.45 µm polyethylene membrane (Millipore) and injected directly into the chromatographic system.

Injections were performed in triplicate. The limits of detection (LD) and quantification (LQ) of the chromatographic analyses were calculated according to Ribani et al. (2004) using the parameters obtained from the analytical curves.

RESULTS AND DISCUSSION

The results for the quantification of methanol and higher alcohols in the cachaça samples sold in PET packaging are described in Table 1. The only sample that contained methanol concentrations greater than the detection limit was the R3 sample (6.70 mg 100 mL⁻¹ a.a.); however, the concentration was within the legal limit. Methanol is considered to be an undesirable contaminant in the beverage because of its toxicity (Cardoso, 2020). It can be formed by the degradation of pectin present in the cellular structure of sugarcane through the boiling of plant material during the distillation process. Its formation can be avoided through proper filtration of the sugarcane juice and must before distillation.

Butan-1-ol and butan-2-ol are considered to be contaminants, and their legal limits are 3.0 and 10 mg 100 mL⁻¹ a.a., respectively. All the samples contained

concentrations of butan-1-ol below the legal limit; however, samples R3 (24.48 mg 100 mL⁻¹ a.a.) and R5 (19.53 mg 100 mL⁻¹ a.a.) contained concentrations of butan-2-ol superior to that established by the legislation. High levels of these two alcohols can be associated with contamination of sugarcane or must by butyl bacteria, and this contamination occurs as a result of inadequate cleaning of equipment or the transportation (dragging) and storage of sugarcane in direct contact with the soil (Cardoso, 2020).

The sum of the concentrations of higher alcohols was within the established limits. Sample R3 contained the lowest concentration of higher alcohols (72.83 mg 100 mL⁻¹ a.a.). Despite being a result within legal limits, higher alcohols are desirable in the beverage. They contribute to the formation of the “flavour” of the beverage. Low levels of higher alcohols can be associated with the fermentation process; the type of yeast or stress to which the yeast is submitted during the process can lead to a smaller yield of secondary compounds (Bortoletto; Silvello; Alcarde, 2018).

The results of the physicochemical analyses can be seen in Table 2. Of the 15 samples under study, only five were within the legal limits for the ethanol concentration.

Table 1: Results for concentrations of methanol and higher alcohols in cachaça samples.

	Methanol mg 100 mL ⁻¹ a.a.	Butan-1-ol mg 100 mL ⁻¹ a.a.	Butan-2-ol mg 100 mL ⁻¹ a.a.	Propan-1-ol mg 100 mL ⁻¹ a.a.	2-Methyl-propanol mg 100 mL ⁻¹ a.a.	3-Methyl-butan-1-ol mg 100 mL ⁻¹ a.a.	Sum of higher alcohols mg 100 mL ⁻¹ a.a.
R1	<LD	0.61 ± 0.07 ^c	<LD	53.28 ± 0.34 ^d	68.45 ± 0.22 ^b	154.21 ± 0.70 ^b	275.93 ± 1.26 ^c
R2	<LD	0.36 ± 0.02 ^e	<LD	56.06 ± 0.18 ^c	64.95 ± 0.99 ^c	167.06 ± 0.65 ^b	288.07 ± 1.81 ^b
R3	6.70 ± 0.25 ^a	0.05 ± 0.01 ^g	24.48 ± 0.50^a	44.11 ± 0.72 ^e	19.57 ± 0.16 ^j	9.16 ± 0.35 ^g	72.83 ± 0.90 ^m
R4	<LD	0.85 ± 0.01 ^a	<LD	44.44 ± 0.13 ^e	53.20 ± 0.22 ^f	159.72 ± 1.00 ^b	257.36 ± 1.35 ^f
R5	<LD	0.38 ± 0.05 ^e	19.53 ± 0.23^b	98.61 ± 0.10 ^a	53.35 ± 0.47 ^f	116.21 ± 0.57 ^d	268.17 ± 0.94 ^d
R6	<LD	0.64 ± 0.06 ^d	<LD	33.18 ± 0.02 ⁱ	68.45 ± 0.22 ⁱ	93.62 ± 0.84 ^e	152.68 ± 0.88 ⁱ
R7	<LD	0.53 ± 0.24 ^e	2.55 ± 0.05 ^e	42.11 ± 0.07 ^f	64.95 ± 0.99 ^h	58.83 ± 0.44 ^f	137.99 ± 0.56 ^k
R8	<LD	0.11 ± 0.18 ^f	<LD	68.69 ± 0.63 ^b	19.57 ± 0.16 ^f	143.63 ± 0.49 ^c	264.37 ± 0.22 ^e
R9	<LD	0.70 ± 0.01 ^b	8.82 ± 0.15 ^c	44.16 ± 1.16 ^e	53.20 ± 0.22 ^a	198.28 ± 0.02 ^a	321.86 ± 0.13 ^a
R10	<LD	0.86 ± 0.01 ^a	<LD	54.13 ± 0.25 ^d	53.35 ± 0.47 ^h	123.88 ± 0.90 ^d	215.05 ± 1.51 ^h
R11	<LD	0.07 ± 0.01 ^g	2.40 ± 0.17 ^e	39.83 ± 0.68 ^g	25.88 ± 0.06 ^h	54.69 ± 0.42 ^e	130.33 ± 0.45 ^k
R12	<LD	0.09 ± 0.01 ^g	3.34 ± 0.01 ^d	44.37 ± 0.11 ^e	37.05 ± 0.19 ^h	59.03 ± 0.02 ^e	140.51 ± 0.13 ^l
R13	<LD	0.43 ± 0.02 ^e	<LD	36.76 ± 0.85 ^h	52.06 ± 0.07 ^e	154.39 ± 0.01 ^b	247.27 ± 1.23 ^g
R14	<LD	0.32 ± 0.02 ^e	<LD	41.92 ± 0.65 ^f	79.43 ± 1.05 ^g	93.56 ± 0.29 ^e	178.96 ± 1.61 ⁱ
R15	<LD	0.56 ± 0.02 ^d	<LD	54.64 ± 0.14 ^d	37.05 ± 0.36 ^d	155.42 ± 1.65 ^b	269.36 ± 1.31 ^d
Legislation	20.00	3.00	10.00	-	-	-	360.00

<LD: lower than limit of detection; LD for methanol: 1.48 mg 100 mL⁻¹ a.a.; LD for butan-2-ol: 0.19 mg 100 mL⁻¹ a.a. Means followed by the same letter in the columns are not significantly different by the Scott-Knott test ($\alpha = 5\%$); Values in bold are higher than the legal limit.

Table 2: Results of the physicochemical analysis and quantification of ethyl carbamate in the cachaça samples stored in PET and glass containers.

	Ethanol conc. % v/v ethanol at 20 °C	Aldehydes mg 100 mL ⁻¹ a.a.	Furfural mg 100 mL ⁻¹ a.a.	Volatile acidity mg 100 mL ⁻¹ a.a.	Copper mg L ⁻¹	Esters mg 100 mL ⁻¹ a.a.	Dry extract g L ⁻¹	Ethyl carbamate µg L ⁻¹
R1	39.23 ± 0.06 ^b	18.58 ± 0.03 ^d	<LD	22.34 ± 0.03 ^f	0.13 ± 0.01 ^g	21.85 ± 0.03 ^d	15.89 ± 0.05 ^b	57.20 ± 0.88 ^c
R2	39.44 ± 0.05 ^b	14.84 ± 0.30 ^f	<LD	29.63 ± 0.03 ^d	0.31 ± 0.01 ^f	21.73 ± 0.02 ^d	0.88 ± 0.01 ^g	77.80 ± 0.24 ^a
R3	35.52 ± 0.04^e	6.69 ± 0.16 ^k	<LD	18.51 ± 2.08 ^g	0.19 ± 0.01 ^g	18.10 ± 1.23 ^e	0.10 ± 0.01 ^h	18.31 ± 0.10 ^g
R4	38.53 ± 0.05 ^d	10.89 ± 0.01 ^j	<LD	26.54 ± 0.03 ^e	0.80 ± 0.02 ^e	26.70 ± 0.03 ^c	0.04 ± 0.01 ^h	8.31 ± 0.40 ^j
R5	36.45 ± 0.02^e	26.82 ± 0.14 ^a	1.97 ± 0.05 ^b	262.55 ± 2.11^a	4.03 ± 0.01 ^a	119.93 ± 0.05 ^a	1.17 ± 0.01 ^g	<LD
R6	38.30 ± 0.05 ^d	13.84 ± 0.02 ^h	1.77 ± 0.10 ^c	70.57 ± 2.00 ^b	2.90 ± 0.04 ^b	45.87 ± 1.06 ^b	2.26 ± 0.01 ^f	<LD
R7	25.73 ± 0.03^j	17.17 ± 0.02 ^e	5.08 ± 0.07^a	45.42 ± 0.05 ^c	1.07 ± 0.01 ^d	29.98 ± 0.03 ^c	3.57 ± 0.01 ^d	37.24 ± 0.13 ^f
R8	27.88 ± 0.13^j	11.88 ± 0.06 ^j	<LD	68.12 ± 0.32 ^b	0.31 ± 0.01 ^f	29.20 ± 1.40 ^c	0.07 ± 0.01 ^h	<LQ
R9	32.01 ± 0.12^f	22.43 ± 0.26 ^b	<LD	70.75 ± 2.55 ^b	2.26 ± 0.15 ^c	33.47 ± 1.46 ^c	0.02 ± 0.01 ^h	51.57 ± 1.46 ^d
R10	31.99 ± 0.01^f	11.05 ± 0.01 ^j	<LD	27.40 ± 0.01 ^e	0.08 ± 0.02 ^h	26.79 ± 0.01 ^c	0.07 ± 0.02 ^h	11.59 ± 0.53 ^j
R11	27.64 ± 0.11ⁱ	14.38 ± 0.06 ^g	<LD	31.71 ± 0.13 ^d	1.00 ± 0.01 ^d	29.45 ± 1.43 ^c	0.08 ± 0.01 ^h	37.43 ± 0.13 ^f
R12	29.24 ± 0.04^h	13.60 ± 0.02 ^h	<LD	24.98 ± 0.03 ^e	1.11 ± 0.07 ^d	30.77 ± 1.42 ^c	2.62 ± 0.01 ^e	47.53 ± 0.20 ^e
R13	40.00 ± 0.21 ^a	14.36 ± 0.08 ^g	<LD	27.40 ± 1.97 ^e	0.39 ± 0.01 ^f	27.85 ± 0.15 ^c	28.68 ± 0.09 ^a	13.2 ± 0.30 ^h
R14	31.07 ± 0.04^g	7.11 ± 0.01 ^k	<LD	23.51 ± 0.03 ^f	0.13 ± 0.01 ^g	30.34 ± 2.71 ^c	0.14 ± 0.01 ^h	6.73 ± 0.60 ^k
R15	38.94 ± 0.02 ^c	21.27 ± 0.27 ^c	0.61 ± 0.01 ^d	23.51 ± 0.03 ^e	0.18 ± 0.02 ^g	24.21 ± 2.21 ^d	14.76 ± 0.38 ^c	61.44 ± 0.22 ^b
Legislation	38-48	30,00	5,00	150,00	5,00	200,00	-	210,00

ND: Not detected. <LD: lower than limit of detection; LD for ethyl carbamate: 1.71 µg L⁻¹; LQ for ethyl carbamate: 5.69 µg L⁻¹; LD for furfural: 0.017 mg 100 mL⁻¹ a.a.; Means followed by the same letter in the columns are not significantly different by the Scott-Knott test (α = 5%); Values in bold are higher than the legal limit.

Low values for the alcohol concentration can be associated with different factors, such as failure during the production process, incorrect cutting of fractions, storage, among others. The dilution of beverages to an alcoholic strength inferior to the legal limit is an illegal practice. It increases the yield of the product and reduces the cost of the final product. However, the product delivered to the consumer is of a low quality that does not match the label on the beverage (Bortoletto; Silvello; Alcarde, 2018).

Samples R7, R8 and R11 had the lowest ethanol concentrations observed in this study, and these values were about 10 - 12% below the concentration required by the legislation. Despite the possible losses during the production process and storage of the beverage, the values are much lower than others reported in the literature.

Santiago, Cardoso and Nelson (2017) determined the concentrations of higher alcohols, phenolic compounds, color intensity and dry extract in cachaça stored and aged in different types of wood and mentioned that the beverage can be stored in wooden barrels or stainless steel drums before commercialization. For the authors, the sealing of these containers and control of the temperature in the storage

room must be performed correctly to avoid the evaporation of volatile compounds.

During the aging and storage of beverages, a decrease in the alcohol concentration is common. It can be corrected with the use of a freshly distilled white cachaça or by adding a beverage with a higher alcohol content during aging to compensate for the loss during storage.

Packaging can influence the loss of volatile compounds during storage, and the type of sealing, material, exposure and transport conditions are extremely important variables in this process. However, Holanda et al. (2015) stored cachaça in glass and plastic containers for 120 days, and observed that there were no differences in the ethanol concentrations of cachaça stored in the PET or glass containers. These results do not corroborate those found in this work, where several variations occurred.

The aldehyde concentration in the beverage was within the legal limits for all the samples. Acetaldehyde is the principal compound in this class. It is formed during the fermentation of the beverage, where it is reduced to ethanol. Sample R5 had the highest concentration of this compound (26.82 mg 100 mL⁻¹ a.a.), whereas sample R14 contained the lowest concentration.

In large quantities, it is undesirable because it is associated with sensory damage, and it is responsible for the hangover after drinking the beverage (Cardoso, 2020).

Furfural and hydroxymethylfurfural are two aldehydes that are controlled separately and are considered to be undesirable contaminants in the beverage because of their toxicity. Its presence can be associated with different factors, including the burning of sugarcane before harvesting, aging of the beverage in charred barrels, or the distillation of the fermented must without the total consumption of sugar by the yeasts (Bortoletto; Silvello; Alcarde, 2018; Masson et al., 2007). Glucose and fructose undergo a dehydration reaction when exposed to thermal processes, a water molecule is eliminated, and furfural and hydroxymethylfurfural, respectively, are formed (Alcarde, 2017).

The volatile acidity in most of the samples was within the established limit of 150.0 mg 100 mL⁻¹ with the exception of sample R5, which contained a 262.55 mg 100 mL⁻¹. The volatile acidity arises during the fermentation process, and it contributes to the aroma and flavor of the beverage at low levels. To control the volatile acidity in cachaça, the still must be well sanitized, and the yeasts that ferment the must should be homogeneous so as to avoid contamination by bacteria during fermentation. In addition, other factors such as must management and control of time and temperature during fermentation are essential to prevent high levels of volatile acidity in the cachaça (Cardoso, 2020).

The concentrations of copper were within the legal limits. This metal is a contaminant in the beverage and comes from the distillation apparatus. It is found in beverages produced in copper stills. Sample R5 had the highest concentration of the contaminant, 4.03 mg L⁻¹. The copper from copper stills can result in the complexation of dimethyl sulfide (DMS). As a consequence, the removal of this sulfur compound improves the sensory quality of the beverage. In adequate concentrations and with adequate cleaning of the stills, copper can be an alternative that helps improve the final quality of the distillate (Bortoletto; Silvello; Alcarde, 2018).

Esters are compounds responsible for the composition of the "flavour" of the beverage. In ideal concentrations, they provide fruity and floral aromas that contribute to the sensory quality of the beverage (Cardoso, 2020). In the samples analyzed, all the concentrations found were within the established limits, and sample R5 contained the highest concentration (119.93 mg 100 mL⁻¹).

Esters can be associated with the fermentation and production process Ribeiro et al. (2017) studied the influence of must treatment and the use of native and selected yeast. There was a difference in the total contents of these compounds in cachaças produced with different yeasts. The levels found were higher in the cachaça fermented with the native yeast, for which the concentrations ranged from 32.75 to 37.16 mg 100

mL⁻¹. These results that corroborate most of the samples in this work, with the exception of sample R5, which contained the highest concentration among the samples.

The results obtained for the concentrations of dry extract were low, ranging from 28.68 mg 100 mL⁻¹ for the R13 sample, 15.89 mg 100 mL⁻¹ for the R1 sample, to 14.76 mg 100 mL⁻¹ for R15. After production, the beverage can be sweetened by adding sugar, which in turn dissolves in the beverage and is detected in the dry extract analysis (Brasil, 2005a). Oliveira et al. (2020) studied cachaça produced in the state of Paraíba and found values of up to 6.21 g L⁻¹ of dry extract in the cachaça samples. The authors associated the high values of dry extract with the fact that the samples had been sweetened. The results found in this work corroborate those of the authors because the highest values for dry extract were found in the sweetened cachaça samples.

The ethyl carbamate concentrations of the samples analyzed in this work are within the legal limit (Table 2). The highest concentrations were found in samples distilled through stainless steel columns: R1 (57.20 µg L⁻¹), R2 (77.80 µg L⁻¹) and R15 (61.44 µg L⁻¹). The presence of these contaminants can be associated with the distillation system used in the production of the beverages. Rodrigues et al. (2020) studied organic contaminants in cachaça produced in copper stills and stainless steel columns and observed higher ethyl carbamate concentrations in beverages produced by the fractional distillation through stainless steel columns. Values ranging from <LQ to 245.31 µg L⁻¹ were found. Their results corroborate those found in this work, where the highest values found were those obtained with beverages distilled through a stainless steel column. Other factors can be associated with the presence of ethyl carbamate in other samples, such as those studied by Cravo et al. (2019). The authors studied the presence of cyanogenic glycosides in different sugarcane varieties and observed a correlation of the compound dhurrin, a cyanogenic glycoside, with the levels of ethyl carbamate found in the cachaça samples.

The results obtained in the identification and quantification of 10 Polycyclic Aromatic Hydrocarbons (PAHs) can be seen in Table 3. All the samples contained PAHs, and the lowest concentrations (0.731 and 0.741 µg L⁻¹) were found in samples R4 and R13, respectively. Sample R12 had the highest concentration of all the PAHs (20.90 µg L⁻¹), followed by samples R7 (17.23 µg L⁻¹), R8 (17.61 µg L⁻¹) and R9 (17.02 µg L⁻¹). The mean value found for all the samples in this study was 10.91 µg L⁻¹.

PAHs with up to three rings were found in higher concentrations in all the samples, and anthracene was not detected in any sample. Acenaphylene was detected in all the samples. Fluorene was found in large concentrations in the samples with the highest levels of PAHs, R7 (15.98 µg L⁻¹) followed by R12 (18.61 µg L⁻¹).

Table 3: Concentration of PAHs ($\mu\text{g L}^{-1}$) in samples of cachaça sold in PET packaging.

											Sum of the PAHs
R1	0.26 ± 0.01^g	1.34 ± 0.01^e	<LQ	0.60 ± 0.01^c	<LD	<LQ	3.25 ± 0.02^a	0.86 ± 0.01^b	2.84 ± 0.06^a	<LD	9.15 ^h
R2	<LQ	7.51 ± 0.01^b	0.84 ± 0.01^i	1.56 ± 0.11^b	<LD	<LD	1.74 ± 0.02^b	0.67 ± 0.02^c	0.98 ± 0.01^h	<LD	13.31 ^e
R3	<LD	0.87 ± 0.01^s	3.09 ± 0.02^h	0.54 ± 0.01^d	<LD	<LD	<LQ	<LD	0.71 ± 0.01^j	<LD	5.21 ^k
R4	0.09 ± 0.01^h	<LD	<LQ	0.12 ± 0.01^f	<LD	<LQ	0.52 ± 0.01^d	<LD	<LQ	<LD	0.73 ^l
R5	<LD	5.39 ± 0.11^c	7.39 ± 0.04^c	<LD	<LD	<LQ	<LD	0.53 ± 0.02^d	0.97 ± 0.01^h	<LD	14.29 ^d
R6	<LD	7.70 ± 0.13^a	3.57 ± 0.05^g	<LD	<LD	<LQ	<LD	<LD	1.47 ± 0.04^e	<LD	12.75 ^f
R7	<LD	<LD	15.98 ± 0.11^b	<LD	<LD	<LD	<LQ	<LD	1.24 ± 0.03^f	<LD	17.23 ^c
R8	2.97 ± 0.05^b	1.12 ± 0.04^f	6.48 ± 0.05^d	<LQ	<LD	<LQ	0.37 ± 0.01^e	2.47 ± 0.03^a	2.21 ± 0.01^d	1.99 ± 0.04^a	17.61 ^b
R9	4.72 ± 0.04^a	2.35 ± 0.08^d	0.65 ± 0.02^j	0.49 ± 0.02^e	<LD	1.12 ± 0.03^b	0.87 ± 0.01^c	2.63 ± 0.01^a	2.49 ± 0.02^b	1.71 ± 0.02^b	17.02 ^c
R10	0.25 ± 0.02^f	0.57 ± 0.09^h	5.53 ± 0.06^e	1.78 ± 0.01^a	<LD	0.26 ± 0.01^d	<LD	<LD	1.07 ± 0.01^s	1.46 ± 0.01^c	10.91 ^g
R11	0.37 ± 0.01^e	0.29 ± 0.03^j	4.55 ± 0.03^f	<LD	<LD	<LQ	<LQ	<LD	0.43 ± 0.22^n	0.62 ± 0.01^e	6.26 ^l
R12	0.25 ± 0.01^f	<LD	18.61 ± 0.04^a	<LD	<LD	<LQ	<LD	<LD	0.86 ± 0.01^i	1.17 ± 0.01^d	20.90 ^a
R13	<LQ	<LD	<LD	<LD	<LD	<LD	0.27 ± 0.02^f	<LD	0.47 ± 0.01^n	<LD	0.74 ^l
R14	0.68 ± 0.01^d	0.53 ± 0.02^l	3.11 ± 0.05^h	<LD	<LD	<LQ	<LQ	<LD	0.97 ± 0.01^h	<LD	5.28 ^k
R15	1.24 ± 0.02^c	<LQ	<LD	<LD	<LD	1.68 ± 0.04^a	3.01 ± 0.03^a	0.69 ± 0.02^c	0.43 ± 0.01^h	<LD	7.05 ^l
LD	0.06	0.08	0.19	0.11	0.16	0.07	0.08	0.13	0.09	0.17	-
LQ	0.21	0.27	0.62	0.36	0.54	0.24	0.27	0.45	0.29	0.57	-

<LD: lower than the limit of detection; <LQ: lower than the limit of quantification; ND: not detected. Means followed by the same letter in the columns are not significantly different by the Scott-Knott test ($\alpha = 5\%$). The highest values are in bold.

Research carried out by Wenzl and Zelinnova (2019) showed that PAHs of lower molar mass are the most common in food contamination; their formation occurs at lower temperatures, and they are more easily absorbed by the food matrix. These data corroborate those found in this work, where it was observed that the greatest contamination by compounds with up to three rings were found in the R12 sample, representing about 90% of the total concentration of the contaminants in the beverage.

Other studies on the quantification of PAHs in cachaça have obtained similar results, Silva et al. (2018) found contaminants in the 20 samples of alembic cachaça analyzed. Benzo(a)pyrene was present in 19 of the samples analyzed, a result that partially corroborates those of this work because the compound was detected in only five samples.

Galinaro, Cardoso and Franco (2007) and Riachi et al. (2014) studied cachaça produced with burnt and unburned sugarcane and observed a higher concentration of PAHs in cachaça produced with burnt sugarcane. The increase was associated with the thermal process that played a role in the degradation of organic molecules of residual agricultural pesticides, which is one of the pathways for formation of PAHs.

The quality of the raw material used for the production of cachaça can contribute to the presence of PAHs in the final drink. The inappropriate practice of burning sugarcane during harvesting can result in the formation of PAHs in sugarcane, contributing to the presence of these contaminants in products produced from this raw material (Singh et al., 2016). Tfouni and Toledo (2007) analyzed commercial sugars and found that the concentration of PAHs in the final product depended on the amount of burned sugarcane being processed, and PAHs were detected in 57% of the samples at concentrations ranging from not detected to $1.35 \mu\text{g kg}^{-1}$. Silva et al. (2011) measured PAHs in rapadura produced in different regions of Brazil, and the contents of the sum of PAHs in the samples ranged from 4.03 to $0.50 \mu\text{g kg}^{-1}$. The authors associated the presence of these contaminants with the burning of sugarcane used as the raw material.

The emergence of PAHs is associated with thermal combustion or pyrolysis processes by some authors. According to Purcaro, Moret and Conte (2016), the ideal temperature for formation of these compounds is between 500 and $750 \text{ }^\circ\text{C}$. The breakdown of organic compounds into unstable and small radicals occurs. These radicals recombine through successive reactions to form cyclic and structures stabilized through resonance of electron pairs. During the manufacturing process, PET packaging undergoes thermal processes in material synthesis and

polymer molding so that it obtains the desired shape. These processes can be associated with the formation of PAHs and other contaminants found in polymeric packaging for food purposes (Souza et al., 2022)

Machado et al. (2014) investigated the storage of cachaça in high-density polyethylene drums and glass containers. The beverage samples stored in glass bottles were contaminated only by PAHs of up to three rings, whereas high concentrations of PAHs with more than three rings were found in the samples stored in polyethylene drums. Among the compounds with high molar mass, fluoranthene and pyrene were the most frequently found, whereas benzo(a)anthracene, acephenanthylene and benzo(a)pyrene were almost not detected in the samples. The contamination of cachaça by PAHs from high-density polyethylene packaging can be associated with the production of the packaging at high temperatures, which can cause the formation of these compounds.

When he evaluated five brands of cachaça stored in PET packaging for different periods of time, Souza et al. (2022) observed that the types of packaging interfered in the concentration of these contaminants. Samples stored for a longer time in PET packaging contained higher concentrations of these contaminants in all the brands of cachaça analyzed. The highest total concentration of PAH's in the packages at the times analyzed was found in the Ap1_{8m} sample ($17.916 \mu\text{g L}^{-1}$), a result that corroborates those found in this work.

The toxic and carcinogenic effect of PAHs is associated with the number of condensed aromatic rings present in their structure. High molecular weight PAHs are more toxic and potential carcinogens. The carcinogenic effect of individual PAHs can be obtained from comparison with benzo(a)pyrene using a conversion factor called Toxicity Equivalence Factor (TEF). Based on the TEF, FAO/WHO concluded that 13 PAHs are clearly carcinogenic and genotoxic. Among the PAHs evaluated in this study, benzo(a)pyrene is considered carcinogenic to humans, naphthalene and benzo(a)anthracene are considered possibly carcinogenic to humans. These three PAHs deserve special attention and must have their concentrations and intake controlled (Silva et al., 2011; Codex Alimentarius Comition - CAC, 2005; World Health Organization - Who, 2005).

No limits are defined for the presence of PAHs in foods and beverages by Brazilian legislation. The only limitations found were for the levels of benzo(a)pyrene in specific products such as ice, water and olive oil, which range from 0.03 to $2.00 \mu\text{g Kg}^{-1}$; According to European regulation No. 835/2011, which determines the maximum levels of PAHs in some foods, the maximum levels must be safe and represent the lowest possible values (EC, 2011; Garcia et

al., 2014). The total sum of PAH's suggested by the German Society for Fat Science is $25 \mu\text{g L}^{-1}$ for smoked foods (Da Silva et al., 2021; Galinaro; Franco, 2009). In this work, no sample contained concentrations higher than those suggested by this legislation. However, the lack of specific legislation for cachaça can be a problem for controlling the content of these substances to ensure the safety and health of the consumer.

CONCLUSIONS

The storage of cachaça in PET packaging is an alternative to replacing glass packaging. However, 60% of the samples analyzed contained concentrations of contaminants outside the legal limits, and most samples had low concentrations of ethanol. All samples stored in polymeric packaging contained PAHs with concentrations higher than those found in the literature for cachaça stored in other types of packaging. This may be associated with the thermal processes involved in the production of these packages. Establishing limits for these compounds in legislation would be necessary to ensure product quality and consumer safety.

AUTHOR CONTRIBUTIONS

Conceptual idea: Cardoso, M. G.; Barbosa, R. B.; Methodology design: Cardoso, M. G.; Barbosa, R. B.; Santiago, W. D.; Data collection: Barbosa, R. B.; Alvarenga, G. F.; Ferreira, V. R. F.; Data analysis and interpretation: Barbosa, R. B.; Cardoso, M. G.; Writing and editing: Barbosa, R. B.; Alvarenga, G. F.; Nelson, D. L.; Cardoso, M. G.

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