RAISSA RACHEL SALUSTRIANO DA SILVA-MATOS
LÍDIA FERREIRA MORAES
FABÍOLA LUZIA DE SOUSA SILVA
(ORGANIZADORAS)

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APRESENTAÇÃO

O agronegócio brasileiro vem se expandindo cada vez mais, isso se deve ao constante crescimento populacional, com isso tem-se ume demanda maior por alimentos e insumos necessários para os processos produtivos, as importações e exportações também tem a sua influência para tal acontecimento, já que o Brasil se destaca entre os países que mais produzem.

Entretanto, mesmo com toda informação já existente ainda se faz necessário o desenvolvimento de novos estudos, a fim de capacitar e minimizar alguns entraves existentes no sistema de produção, considerando o cenário atual a demanda por informações de boa qualidade é indispensável.

Com isso, o uso de tecnologias, técnicas e pesquisas necessitam estar atreladas na produção agrícola para desde modo obter sucesso e alta produtividade. Com base nisso a obra “Desenvolvimento da pesquisa científica, tecnologia e inovação na agronomia 2” vem com o intuito de trazer aos seus leitores informações essenciais para o sistema agrícola.

Apresentando trabalhos desenvolvidos e resultados concretos, com o objetivo de informatização e capacitação acerca deste setor, oferecendo a possibilidade do leitor de agregar conhecimentos sobre pesquisas desenvolvidas para a agricultura. Pesquisas que buscam contribuir para o aprimoramento dos pequenos, médios e grandes produtores. Desejamos a todos, uma excelente leitura!

Raissa Rachel Salustriano da Silva-Matos
Lídia Ferreira Moraes
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SUMÁRIO

CAPÍTULO 3

CHANGES IN THE CHEMICAL QUALITY OF PINK PEPPER FRUITS DURING STORAGE

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ABSTRACT: In this work, the effect of three storage conditions of pink pepper fruits, during six months: was evaluated at room temperature in shade, fridge and freezer. Each month, samples were taken of the pink pepper fruits in each condition and destined for extraction/analysis of fixed oils and essential oils. Among the results obtained, we verified that the storage condition did not negatively affect the content of fixed oil and fatty acid analyzed in pink pepper fruits. On the other hand, we found a change in the chemical profile from the third month of storage with an increase in oxygenated terpenes (elmol and α-eudesmol), decrease in monoterpene hydrocarbons (α-pinene and β-pinene, sylvestrene and α-phellandrene) and decrease in essential oil content in the sixth month of storage. The results obtained contribute to the perception of the need to carry out the storage of pink pepper fruits for an adequate time.

KEYWORDS: Schinus terebinthifolius, essential oil, lipids, storage.

ALTERAÇÕES NA QUALIDADE QUÍMICA DOS FRUTOS DE PIMENTA ROSA DURANTE O ARMAZENAMENTO

RESUMO: Neste trabalho, avaliou-se o efeito de três condições de armazenamento de frutos de pimenta rosa, durante seis meses: à temperatura ambiente à sombra, geladeira e freezer. A cada mês, foram colhidas amostras dos frutos da pimenta rosa em cada condição e destinadas à extração/análise de óleos fixos e óleos essenciais. Dentre os resultados obtidos, verificamos que a condição de armazenamento não afetou negativamente o teor de óleo fixo

e ácidos graxos analisados em frutos de pimenta rosa. Por outro lado, encontramos uma mudança no perfil químico a partir do terceiro mês de armazenamento com aumento dos terpenos oxigenados (elmol e α-eudesmol), diminuição dos hidrocarbonetos monoterpenos (α-pineno e β-pineno, silvestreno e α- felandreno) e diminuição do teor de óleo essencial no sexto mês de armazenamento. Os resultados obtidos contribuem para a percepção da necessidade de realizar o armazenamento dos frutos de pimenta rosa por tempo adequado.
KEYWORDS: Schinus terebinthifolius, óleo essencial, lipídeos, armazenamento.

1 | INTRODUCTION

Pink pepper is an aromatic plant that occurs in the *Mata Atlântica* (Atlantic forest), has economic and social importance, mainly because the bioactives extracted from the fruits have medicinal, cosmetic and nutritional properties (Alves *et al.*, 2020; Feriani *et al.*, 2021; Giuffrida *et al.*, 2020; Tlili *et al.*, 2018). All pink pepper plant is useful: the bark and leaves have medicinal properties; seedlings are used in the recovery of degraded areas, in urban landscaping, and wood for making posts (Mendonça, Silva-Mann e Rabbani, 2014; Santos *et al.*, 2012).

However, the fruit is the part of the plant that presents the greatest biotechnological potential, among the reasons: because the pink pepper seed is used to the species propagation; due to the presence of essential oils and lipids that confer sensory and nutritional properties and, therefore, appreciated and used as a condiment (Tlili *et al.*, 2018), also by the fact that the essential oil to have bioactive properties and be used for the preparation of drugs and cosmetics (Mendonça, Silva-Mann e Rabbani, 2014).

Given the importance of pink pepper fruits for the aromas and fragrances sector, some studies have been developed to evaluate the effects of some post-harvest variables, such as drying (Fonseca *et al.*, 2021; Silva *et al.*, 2017), storage (Ribeiro *et al.*, 2018; Silveira *et al.*, 2021) and extractive methods (Andrade, Poncelet e Ferreira, 2017; Bittencourt Fagundes *et al.*, 2020) on the production and quality of bioactives obtained from the fruits. However, studies to evaluate the effects of temperature and storage time of pink pepper fruits have been based on the seeds germination capacity (Ribeiro *et al.*, 2018; Silveira *et al.*, 2021), despite the chemical profile being an important parameter with implications for the properties of fruits and their products.

Regardless of the extraction methods involved, the lipids and essential oils levels obtained from pink pepper fruits range from 8–14% and 0.5–5%, respectively (Fonseca *et al.*, 2021; Oliveira *et al.*, 2014; Silva *et al.*, 2017; Tlili *et al.*, 2018). It is known that the compounds that make up these two chemical classes (lipids and terpenes) are very susceptible to oxidation in the presence of moisture, light, heat and O_2 (Turek e Stintzing, 2013) and the exposure time to any of these conditions can amplify the formation of oxygenated artifacts.

For this reason, this study aimed to evaluate the effect of storage on the chemical

quality of pink pepper fruits. In this context, we paid attention to the chemical composition of fatty acids and essential oils extracted from pink pepper fruits, under the effect of three storage conditions (room temperature, cold-room and freezing) and storage time (six months).

2 | MATERIALS AND METHODS

General

Pink pepper (*Schinus terebinthifolius* Raddi) fruits were collected in 2018 from plants with 2.5 years old of the active germplasm collection of Federal Rural University of Rio de Janeiro (UFRRJ), maintained by the department of plant science on BR 465, Km 7, CEP 23897-000, Seropédica, Rio de Janeiro, Brazil. Alkane and fatty acid methyl ester standards and analytical/chromatographic grade reagents were purchased from Sigma-Aldrich (São Paulo, Brazil). The lipids and essential oils extraction and other procedures related to the distillates were carried out at the Laboratory of Plantas Aromáticas e Medicinais (UFRRJ, Seropédica, Brazil). Chromatographic analysis, GC/FID (Hewlett-Packard 5890 II, Palo Alto, USA) and GC/MS (QP-2010 Plus, Shimadzu, Japan) were performed in the Central Analítica Multiusuário of the Pos-graduate Program in Chemistry (UFRRJ, Seropédica, Brazil).

Treatments

A composite sampling of pink pepper fruits collected from the UFRRJ germplasm collection was dried in a forced-air circulation oven (Eletrolab EL 402, São Paulo, Brazil) for 48 hours at 37°C until reaching constant weight (with about 13% humidity), then, fractionated into three sub-samples, and each one was stored at room temperature in shade (29 ±3 °C) or in the cold chamber or fridge (5 ±1 °C) or a freezer (-15±3 °C), protected from moisture and direct light. Pink pepper fruits sample from each treatment (storage condition) was removed per month, until the sixth month of storage, for the extraction of fixed oil and essential oil. The control sample consisted of dried pink pepper fruits not stored.

Essential oil extraction and analysis

Pink pepper fruits samples (30 g) were subjected to hydrodistillation by triplicate, using the Clevenger apparatus for two consecutive hours. The essential oil was separated by phase difference, the moisture removed with anhydrous sodium sulfate and then stored in amber glass bottles at -20 °C for chemical analysis.

The analysis of essential oils esters was performed through three injections of samples 1μL (1%, *v*/*v*). Gas chromatographic (GC) analysis was carried out with a Hewlett-Packard 5890 II apparatus equipped with a flame ionization detector (FID), and a split/splitless injector at a 1:20 split ratio was used to separate and detect volatile oil

constituents. Substances were separated into a VF-5ms fused silica capillary column (30 m × 0.25 mm i.d., film thickness 0.25 μm, Agilent J&W). The oven, injector and detector temperatures were programmed as reported by Adams (2007). The carrier gas used was He (1 mL/min). Injected volume was 1 μL at a 1:20 split ratio. The percentage of volatile oil compounds was calculated from the relative area of each peak analyzed by FID. Volatile oils were also analyzed with a Shimadzu GC/MS QP-2010 Plus instrument. Carrier gas flow, capillary column and temperature conditions for GC/MS analysis were the same as those described for GC/FID and reported by Adams (2007). Mass spectrometer operating conditions were ionization voltage of 70 eV, mass range of 40-400 m/z and 0.5 scan/s. The compounds' retention indexes were calculated based on co-injection of samples with a C8-C20 hydrocarbon mixture. Constituents were identified by comparison of their mass spectra with the National Institute of Standards and Technology library (NIST-2011, Gaithersburg, Maryland, USA) and with those reported by Adams (2007).

Fixed oil extraction, derivatization and analysis

Pink pepper fruits samples (5g) were subjected to distillation by triplicate by Soxhlet apparatus for 3h at 80 °C with petroleum ether, then stored in amber glass vials at -20 °C for the derivatization step. For the methylation process, 1.5 mg of fixed oil was used in duplicate. In test tubes containing the oil samples, 0.4 ml of a sodium hydroxide solution in anhydrous methanol (0.5 N) were added and incubated for 10 min in a water bath at 100 °C. After, 0.5 ml of Boron trifluoride-methanol (BF3, 10%) was added for a further 2 min. At room temperature, the solutions were partitioned with 2 mL of petroleum ether and anhydrous sodium sulfate, after then, the organic phase (containing fatty acid methyl ester, FAME) was collected and transferred to 1.8 mL amber glass vials, and stored at -20 °C for chemical analysis.

The analysis of FAME was performed through three injections of samples and standards 1μL. Standards were properly weighed and dissolved in dichloromethane (0,2 mg/mL). Gas chromatographic (GC/FID) analysis was carried out with a Hewlett-Packard 5890 II apparatus (Palo Alto, USA) equipped with a flame ionization detector, and a split/splitless injector at a 1:20 split ratio was used to separate and detect volatile oil constituents. The percentage of compounds was calculated from the relative area of each peak analyzed by FID. Compounds were separated into a VF-5ms fused silica capillary column (30 m × 0.25 mm i.d., film thickness 0.25 μm, Agilent J&W). The injector and detector temperatures were 240 and 280 °C, respectively. The oven temperature was 180 °C, for one minute, followed by heating at 5 °C/min to 240°C, 0.5 °C/min to 270 °C and 10 °C/min to 290°C, which remained for 10 min. The same samples were also analyzed with a GC/MS QP-2010 Plus instrument (Shimadzu, Japan). Carrier gas flow, capillary column and temperature conditions for GC/MS analysis were the same as those described for GC/FID. Mass spectrometer operating conditions were ionization voltage of 70 eV, mass range of 40-

400 m/z and 0.5 scan/s. The FAME characterization was performed by comparing their retention times with those obtained by injecting authentic FAME standards under the same chromatographic conditions.

Statistical Analysis

The fixed oil and essential oil content (%) from different treatments were obtained in triplicate (n=3). The chromatographic analysis of fixed oil and essential oil was performed in duplicate (n=2) and a composite sampling from three samples of essential oil (n=1), respectively. Data were expressed as arithmetic means ± standard deviation (SD) and submitted to analysis of variance (ANOVA), and differences between means were determined using the Tukey test at P=0.05. For experimental values were evaluated data normality based on D'Agostino-Pearson and Shapiro-Wilk tests and homoscedasticity test.

For the multivariate analysis, the independent variables consisted of 19 essential oil samples, one control and 18 treatments (three storage conditions x six months of storage) and the dependent variables consisted of 26 compounds present in the essential oil (the main substances were defined by those that appeared in at least one sample in a concentration greater than 1.9%), data standardization was not performed. Then, the data matrix was submitted to principal component analysis (PCA), to verify the accumulated variance in the first two or three principal components. Cluster analysis was performed using the hierarchical agglomerative method for the union between groups, the method of mean connection between the groups (UPGMA) was used and the cophenetic correlation between the phenetic and generic matrix was verified to verify its consistency.

3 | RESULTS AND DISCUSSION

Samples of pink pepper fruits under the effects of the treatments were subjected to 3 hours of hydrodistillation with petroleum ether and the content (%, m/m) of fixed oil obtained did not show significant differences as a function of the condition (ambient temperature, cold room and freezer) or storage time (up to six months). The fixed oil content obtained from pink pepper fruits exposed to the treatments also did not differ significantly from the control, which was 7.2% (Fig. 1). On the other hand, samples of pink pepper fruits submitted to 2 hours of hydrodistillation, to obtain the essential oil, showed lower contents (%, m/m) in the sixth month of storage. It was not possible to confirm a significant result between treatments under different storage conditions. (Fig. 1).

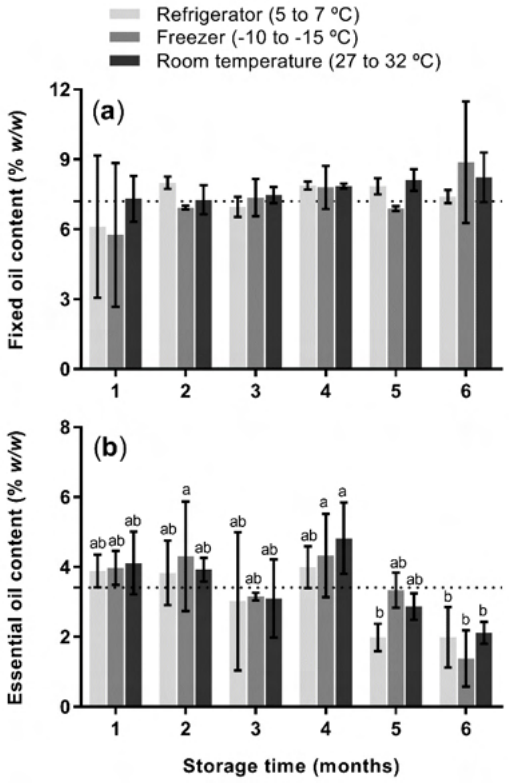

Figure 1 Content (%, w/w) of fixed oil (a) and essential oil (b) from dried pink pepper fruits stored under different conditions and time periods. The numbers one to six represent storage times by month. Columns and bars represent the sample mean ± standard deviation. Light-gray column – cold chamber or refrigerator (5 ±1 °). Gray column – freezer (-15±3 °C). Dark gray column – room temperature in shade (29 ±3 °C). Dotted line indicates means of control sample. Different letters indicate significant results based on the Tukey test ($P < 0.05$).

The oleic (29.2–51.9%), linoleic (19.4–29.9%), palmitic (13.1–24.3%) and arachidonic (4.1–9.2%) fatty acids were the most abundant and together represented on average 89.9% of compounds present in the fixed oil of pink pepper fruits (Fig. 2). It was not possible to observe a significant difference in the contents of linoleic and arachidonic acids as a function of the treatments. As for palmitic and oleic acids, specific significant differences were observed, but it was not possible to point out a trend as a function of treatment and control effects (Fig. 2).

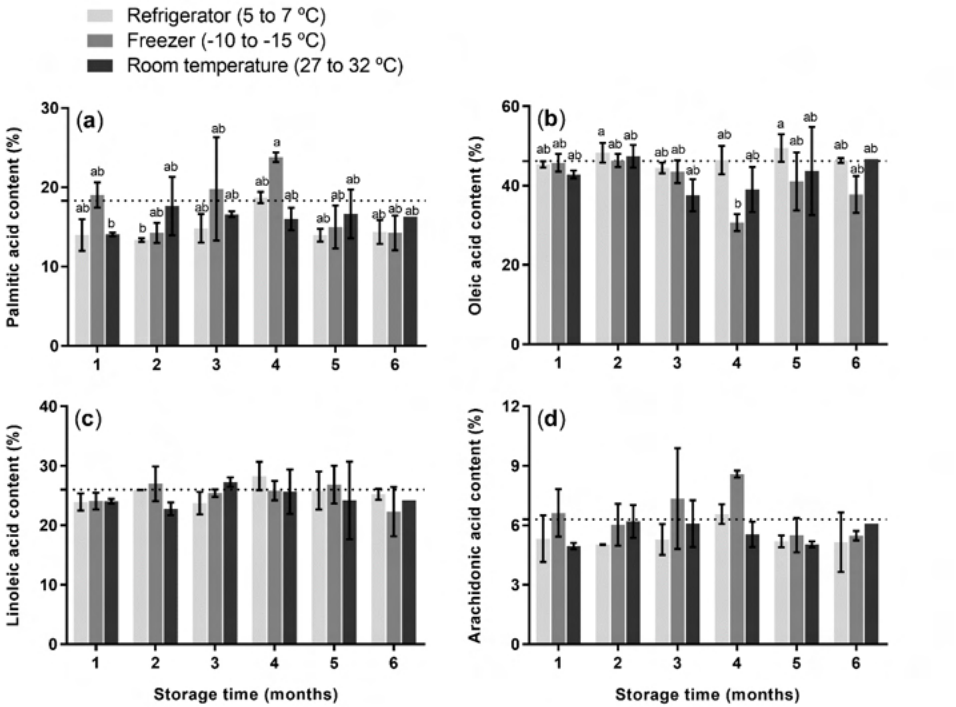

Figure 2 Concentration (%) of palmitic (a), oleic (b), linoleic (c) and arachidonic (d) acids in the fixed oil extracted from dried fruits stored under different conditions and times. The numbers one to six represent storage times by month. Columns and bars represent the sample mean ± standard deviation. Light-gray column – cold chamber or refrigerator (5 ±1 °). Gray column – freezer (-15±3 °C). Dark gray column – room temperature in shade (29 ±3 °C). Dotted line indicates means of control sample. Different letters indicate significant results based on the Tukey test ($P < 0.05$).

The chromatographic analysis (GC-EM and GC-DIC) of the essential oils revealed a rich composition of monoterpene hydrocarbons, except in the sixth month of storage, which showed a higher concentration of oxygenated sesquiterpenes. Overall, the concentration of oxygenated terpenes increased, while that of monoterpene hydrocarbons decreased (Fig. 1). The substances found in the highest concentration in essential oils were β-pinene (2.8–16%), α-pinene (5.6–14.9%), α-phellandrene (1.7–14.6%), elemol (3.7–14.4%), germacrene D (6.5–12.8%), sylvestrene (2.8-11.6%), myrcene (0.3-11.5%) and α-eudesmol (2.7-10.2%), with recurrence in all samples (Table 1).

The data from the chromatographic analysis of the essential oil samples (substances and concentration) were subjected to principal component analysis (PCA) and the two main components (PC1 and PC2) represented 73.1% of all variance (Fig. 3). It was possible to observe the dispersion of the samples, mainly due to the first main component and the factors that contributed the most were: negatively the oxygenated sesquiterpenes elmol and α-eudesmol and positively the hydrocarbons of monoterpenes β-pinene, α-pinene,

sylvestrene and α-phellandrene. The samples were dispersed as a function of storage time, with months 1, 2, 3 and the control (CRT) positioned to the right of PC1, months 4 and 5 to the left of the origin and the sixth month further away and to left of PC1 (Fig. 3).

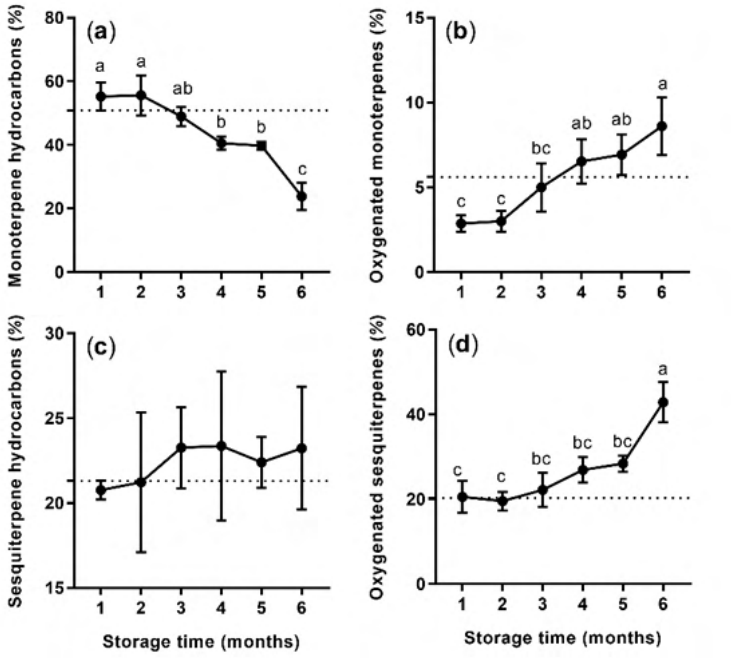

Figure 3 Concentration (%) of monoterpene hydrocarbons (a), oxygenated monoterpenes (b), sesquiterpene hydrocarbons (c) and oxygenated sesquiterpenes (d) in the essential oil extracted from dried fruits stored for different times. The numbers one to six represent storage times by month. Black symbols and bars represent the sample mean ± standard deviation. Dotted line indicates means of control sample. Different letters indicate significant results based on the Tukey test ($P < 0.05$).

The same essential oil data were subjected to analysis of hierarchical clusters, a dendrogram was made by the method of mean link between group (UPGMA) using dissimilarity measures (Euclidean distance) and the results pointed to the formation of six groups (G1 to G6). Group G1 consisted mainly of samples submitted to the first three months of storage and control (CTR), group G2 by samples from the fourth and fifth month, groups G4 and G5 by samples from the sixth month and, finally, group G3 and G6 by samples 2FG and 3FG respectively (Fig. 4). In general, it was possible to see the grouping of samples based on storage months, similar to PCA (Fig. 3).

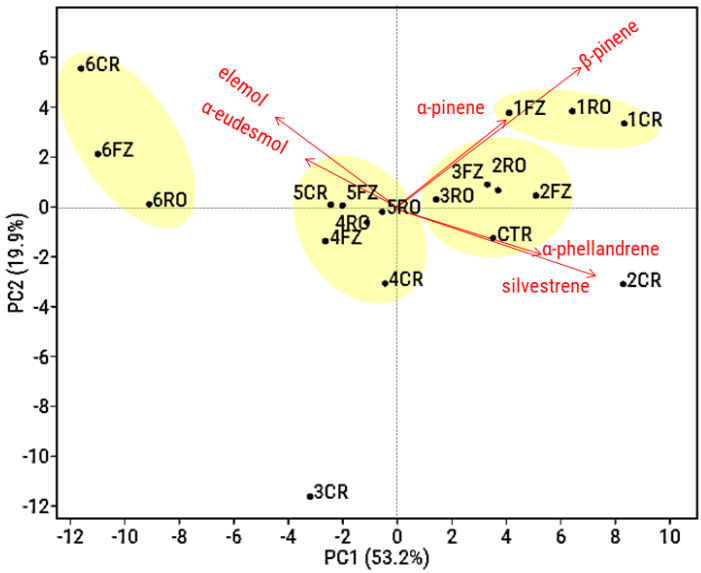

Figure 4 Bi-plot from principal component analysis, based on 26 compounds (dependent variables) in the 19 essential oil samples (independent variables). Red arrows represent the main factors that contribute to variability of samples. Standardization of data was not performed. The numbers one to six represent storage times by month. CTR – control samples. CR – cold chamber or refrigerator (5 ±1 °C). FZ – freezer (-15±3 °C). RO – room temperature in shade (29 ±3 °C).

Although the essential oil and fixed oil compounds are made up of carbon, hydrogen and oxygen, they have different molecular structures, physicochemical characteristics, biosynthesis/degradation routes and biological functions. Terpenes are the main compounds present in the pink pepper fruit essential oils (Santos Cavalcanti, dos *et al.*, 2015), and triacylglycerols in fixed oils (Ennigrou *et al.*, 2017; Tlili *et al.*, 2018). The first is made up of isoprene units that have unsaturations and may or not be oxygenated, while the second is formed by fatty acids, esterified to glycerol and that may also be saturated or unsaturated.

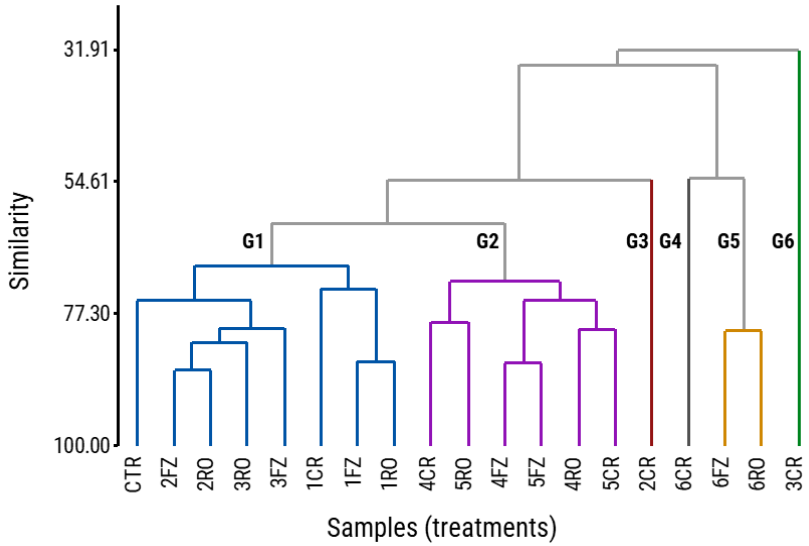

Figure 5 Cluster analysis by Unweighted Pair-Group Method using Arithmetic Avarages (UPGMA) based on the similarity index (Euclidian distance) considering the 26 compounds (dependent variables) in the 19 essential oil samples (independent variables). Cophenetic correlation between the phenetic and generic matrix was calculated to verify its consistency. The G1 to G6 clusters were formed with at least 65% similarity. Cophenetic correlation = 0,8477. The numbers one to six represent storage times by month. CTR – control samples. CR – cold chamber or refrigerator (5 ±1 °C). FZ – freezer (-15±3 °C). RO – room temperature in shade (29 ±3 °C).

In this work, there was no variation in the fatty acid content and composition under the effect of the treatments, only punctual variations in the palmitic and oleic acid contents, nothing that would allow any conclusion (Fig. 1 and 2). However, previous work has shown that fruit maturation (Ennigrou *et al.*, 2017), the type of tissue analyzed (Tlili *et al.*, 2018) promote changes in fatty acid composition.

The sum of oleic C18:1(9)$_{\omega 9}$, linoleic C18:2(9,12)$_{\omega 6}$ and arachidonic C20:4(5,8,11,14)$_{\omega 6}$ unsaturated fatty acids were more abundant than palmitic C18:0 saturated fatty acid, which confers nutritional quality to the pink pepper oil (Fig. 2). In terms of the concentration range of major fatty acids in pink pepper oil, the results presented in this work are similar to showed by other authors (Ennigrou *et al.*, 2017; Oliveira, de, Augusta, *et al.*, 2020). Some works have demonstrated the pink pepper fruits and fixed oil potential as a natural antioxidant in foods (Bittencourt Fagundes *et al.*, 2020; Oliveira, de, Cháves, *et al.*, 2020).

While fixed oils mostly contain compounds with low vapor pressure, essential oils are usually made up of compounds with greater volatility. For this reason, it is to be expected that storage time causes the loss of more volatile compounds, which are abundant in essential oils. However, there was no significant decrease in the essential oil extracted from fruits stored for up to five months, only in the sixth month there was a decrease (Fig. 1). The structures involved with the secretion of volatile compounds are secretory cavities in the

pink pepper fruit pericarp, therefore, protected internally by the tissue (Carmello-Guerreiro e Paoli, 2002; Machado e Carmello-Guerreiro, 2001).

Likely, the decrease in essential oil content only in the sixth month of storage is due to internal processes involved in the transformation of more volatile terpenes into less volatile ones, such as oxygenated terpenes (Turek e Stintzing, 2013). In the distillation process itself, oxygenated sesquiterpenes have a lower extraction rate, as observed in the literature (Oliveira *et al.*, 2020). Terpenes can also be transformed and degraded while still in the tissue (Pott, Vallarino e Osorio, 2020; Whitaker, 2008). These facts added together may be contributing to the lowest amount of essential oil observed in the sixth month of storage.

It was possible to prove an increase in the oxygenated monoterpenes and sesquiterpenes concentration and a decrease in monoterpene hydrocarbons from the first to the sixth month of storage (Fig. 3). This finding has important implications from a sensory and biological point of view, considering fluctuations in the concentration of certain compounds.

The hierarchical cluster analysis showed the samples similarity from the first months and a relative distance from those from the sixth month (Fig. 5), similar to the analysis of principal components (Fig. 4), the latter even allowed to prove that the compounds (factors) that most contributed to the differentiation of the essential oil samples were α-pinene, β-pinene, sylvestrene and α-phellandrene (monoterpene hydrocarbons), which decreased from the first to the sixth month of storage, and on the contrary, elmol and α-eudesmol (oxygenated sesquiterpenes) that increased from the first to the sixth month of storage (Fig. 4).

Volatile compounds are susceptible to chemical transformations during and after the extraction process, as during storage under the temperature effects, humidity, light and the presence of oxygen (Turek e Stintzing, 2013). Nothing prevents the storage condition of pink pepper fruits also creating favorable conditions for chemical transformations inside the fruit, by enzymatic catalysis or not (Pott, Vallarino e Osorio, 2020; Turek e Stintzing, 2013; Whitaker, 2008).

For example, Zhao et al. (2019) observed a decrease in the monoterpene hydrocarbons α-pinene and limonene and an increase in oxygenated sesquiterpenes caryophyllene oxides, alloaromadendrene, isolongifolene-9,10-dehydro and isoaromadendrene epoxide, in short storage time at low temperature. Some studies also pointed to changes in the chemical quality of aromatic species as a function of drying conditions (Fonseca *et al.*, 2021; Silva *et al.*, 2017)and maturation time (Ennigrou *et al.*, 2017; Schimitberger *et al.*, 2018).

The change in the essential oil chemical composition as a function of storage time may be following a natural flow. However, it should be noted that due to possible biotechnological applications, it is important to predict these changes, otherwise, the material will lose the biological function that matters to the product of interest.

			% (relative area)																	
Nome	LRI_C	LRI_L	CTR	1CR	1FZ	1RO	2CR	2FZ	2RO	3CR	3FZ	3RO	4CR	4FZ	4RO	5CR	5FZ	5RO	6CR	6FZ	6RO
β-pineno	979	974	10.7	14.8	14.6	16.0	10.0	10.8	11.1	2.8	10.7	9.7	6.5	7.9	7.4	8.5	9.1	9.3	4.7	5.0	5.2
α-Pineno	935	932	8.3	14.9	11.2	12.0	11.6	11.1	10.0	5.6	11.4	9.7	7.1	7.5	8.3	6.9	8.6	7.5	7.7	6.8	7.8
α-felandreno	1008	1002	10.5	10.8	8.3	9.4	14.6	11.7	10.0	7.8	8.6	8.6	9.3	5.1	7.0	6.0	4.0	7.8	3.1	1.7	2.9
silvestreno	1030	1025	8.8	8.2	7.8	8.0	11.6	9.3	8.2	6.0	7.8	7.3	8.2	6.9	6.3	6.3	6.6	7.6	3.0	2.8	3.9
Mirceno	992	988	2.5	2.1	1.7	1.8	2.9	2.1	2.1	11.5	2.0	1.9	2.1	1.7	1.8	1.6	1.7	1.7	0.3	0.4	2.2
sabineno	974	969	2.0	3.2	2.4	1.3	2.0	1.9	2.3	7.0	4.5	2.3	1.5	3.1	3.6	3.2	2.9	1.1	1.1	1.3	2.8
o-cymeno	1025	1022	4.3	2.3	2.1	2.2	6.3	3.1	2.9	3.8	2.6	2.8	5.3	4.4	2.5	2.7	4.1	3.5	1.4	2.3	3.3
terpinoleno	1088	1086	1.3	0.9	0.8	0.9	1.6	1.2	1.1	1.4	1.0	1.0	1.4	0.7	1.1	1.0	0.6	0.9	0.2	0.1	0.2
gamma-terpinene	1057	1054	0.4	0.5	0.5	0.4	0.2	0.3	0.5	1.3	0.6	0.5	0.2	0.2	0.5	0.8	0.2	0.3	0.0	0.0	0.0
Canphene	947	946	1.0	1.2	0.9	1.1	0.9	0.9	1.0	0.4	1.1	0.9	0.6	0.7	0.9	0.7	0.9	0.7	0.2	0.4	0.3
alpha-Thujene	926	924	0.7	0.7	0.6	0.6	0.8	0.6	0.7	1.0	0.7	0.6	0.5	0.4	0.5	0.5	0.5	0.5	0.1	0.2	0.2
alpha-terpinene	1017	1014	0.3	0.4	0.4	0.3	0.2	0.3	0.4	0.9	0.4	0.3	0.2	0.1	0.3	0.6	0.1	0.2	0.0	0.0	0.0
Tricyclene	922	921	0.2	0.3	0.2	0.2	0.2	0.2	0.2	0.0	0.3	0.2	0.1	0.1	0.0	0.1	0.2	0.1	0.0	0.0	0.0
2-(1-Metil-2-oxopropil) ciclohexanone	1252	-	0.6	0.0	0.3	0.3	0.7	0.4	0.4	0.4	0.6	0.7	1.1	3.3	1.2	0.4	2.9	2.1	0.6	3.3	4.1
4-terpineol	1177	1174	0.9	0.8	1.0	0.6	0.2	0.5	1.0	2.8	1.4	0.9	0.5	1.3	1.3	1.9	1.3	0.7	1.2	1.0	3.2
Bornyl acetate	1285	1287	1.0	1.0	1.1	1.2	0.7	1.0	1.1	0.4	1.2	1.1	0.8	1.2	1.5	1.4	1.5	1.0	1.1	1.7	0.9
Citronellyl acetate	1354	1350	0.9	0.3	0.4	0.4	0.8	0.6	0.5	1.2	0.5	0.6	1.1	0.7	0.6	0.5	0.5	0.7	0.6	0.5	0.9
neoiso-3-thujanol	1148	1147	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.5	0.0	0.0	1.2	0.6	0.0	0.1	0.1
carvacrol	1305	1298	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.9	0.2	0.0	0.4	0.0	0.2	0.0	0.0	0.0
alpha-terpineol	1191	1186	0.8	0.3	0.4	0.6	0.0	0.3	0.4	0.3	0.5	0.4	0.4	0.6	0.5	0.7	0.7	0.7	0.0	0.6	0.5
cis-p-Menth-2-en-1-ol	1121	1118	0.4	0.0	0.0	0.0	0.0	0.0	0.2	0.4	0.2	0.2	0.3	0.2	0.2	0.3	0.2	0.2	0.0	0.0	0.0
beta-copaen-4-alpha-ol	1590	1590	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.4	0.0	0.1	0.1	0.0	0.1	0.2	0.1	0.2
borneol	1167	1165	0.2	0.0	0.0	0.0	0.1	0.0	0.0	0.2	0.0	0.0	0.3	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0
trans-p-Menth-2-en-1-ol	1139	1136	0.2	0.0	0.0	0.0	0.0	0.0	0.2	0.3	0.2	0.1	0.2	0.0	0.1	0.2	0.2	0.2	0.0	0.0	0.0
elemol	1554	1548	6.2	3.7	6.0	4.1	3.8	5.2	5.2	3.7	5.4	4.8	5.5	5.4	5.0	6.9	5.4	6.2	14.4	9.1	8.2
α-eudesmol	1657	1652	2.7	4.0	4.7	5.1	3.5	4.7	4.4	4.9	3.8	5.6	5.3	5.1	4.6	5.1	4.7	5.0	10.2	7.8	7.5
γ-eudesmol	1635	1630	3.1	2.8	4.4	3.9	2.7	3.3	3.4	3.7	2.4	3.7	3.5	4.0	3.4	3.8	3.4	4.3	6.7	6.4	6.3
β-eudesmol	1655	1649	3.9	2.3	4.1	3.3	2.2	3.2	2.8	3.5	2.5	3.7	3.7	4.0	2.9	4.4	3.3	5.3	6.0	5.4	5.1
criptomeridiol	1817	1813	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.8	4.9	4.3
hinesol	1640	1640	0.0	0.3	0.0	0.5	0.5	0.3	0.5	0.7	0.4	1.1	0.9	2.1	1.4	0.5	3.0	1.1	0.0	1.5	0.6
bulnesol	1673	1670	1.9	1.1	1.9	1.5	1.2	1.8	1.8	1.7	1.9	2.7	2.7	2.4	1.6	3.0	2.5	3.0	2.7	2.0	1.3
Spathulenol	1580	1577	0.7	0.6	0.4	0.5	1.1	0.6	0.5	0.7	0.6	0.7	2.2	1.5	1.0	1.0	1.2	1.0	2.4	2.4	1.9
agarospirol	1643	1646	0.0	1.2	1.3	1.5	0.7	0.0	1.3	1.6	0.0	2.0	1.8	1.7	1.5	2.0	1.0	1.8	0.7	1.8	0.4
Oxido de cariofileno	1582	1582	0.7	0.3	0.3	0.5	1.0	0.5	0.4	0.5	0.4	0.5	1.9	1.0	0.6	0.7	0.5	0.6	1.4	1.5	1.3
10-epi-gamma-eudesmol	1620	1622	0.5	0.3	0.5	0.5	0.4	0.6	0.6	0.6	0.4	0.7	0.8	0.7	0.6	0.6	0.6	0.8	0.0	0.7	0.6
salvial-4(14)-em-1-one	1595	1594	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.2	0.2	0.2	0.1	0.2	0.2	0.5	0.2	0.2
1-epi-cubenol	1628	1627	0.0	0.0	0.1	0.4	0.0	0.0	0.3	0.2	0.0	0.4	0.2	0.2	0.3	0.4	0.3	0.3	0.0	0.2	0.0
5-epi-7-epi-alpha-eudesmol	1608	1607	0.2	0.0	0.0	0.0	0.0	0.0	0.2	0.2	0.1	0.2	0.3	0.3	0.2	0.2	0.2	0.3	0.0	0.4	0.0
Germacrene D-4-ol	1577	1574	0.2	0.1	0.2	0.0	0.0	0.1	0.2	0.0	0.2	0.2	0.0	0.0	0.3	0.2	0.0	0.0	0.2	0.0	0.0
germacreno D	1484	1484	6.8	10.4	8.9	8.0	6.5	9.0	9.8	7.5	11.0	8.9	6.7	7.0	11.7	9.4	8.5	6.7	12.8	7.1	7.2
β-Cariofileno	1419	1417	4.1	2.2	2.6	2.6	3.7	3.1	2.9	2.5	2.6	2.9	4.1	3.1	2.7	2.7	2.3	3.2	3.8	2.9	3.1
germacreno B	1556	1559	1.5	1.6	2.0	2.0	2.4	2.7	2.0	1.7	2.4	2.1	2.4	2.2	2.2	1.7	1.8	1.9	3.2	1.9	1.5
Biciclogermacreno	1496	1500	1.9	0.0	2.0	2.0	1.4	2.1	2.3	1.8	2.4	2.2	1.6	1.6	2.7	2.2	1.6	1.8	2.1	1.0	0.8
γ-elemene	1337	1335	0.9	0.5	0.8	0.6	0.4	0.7	0.8	0.6	0.9	0.8	0.5	1.5	0.9	0.9	1.4	0.9	0.5	2.4	2.7
δ-cadinene	1524	1522	1.2	1.4	1.4	1.4	0.0	1.5	1.5	1.9	1.6	1.8	1.4	1.4	2.3	1.8	1.5	1.5	2.1	1.5	1.6
γ-amorphene	1492	1495	0.1	2.1	0.2	0.2	0.0	0.1	0.1	0.2	0.1	0.2	0.0	0.1	0.2	0.2	0.1	0.2	0.0	0.0	0.3
alpha-copaene	1375	1374	0.7	0.6	0.6	0.5	0.6	0.7	0.8	0.7	0.9	0.8	0.7	0.8	1.1	0.9	1.0	0.7	0.7	0.9	0.6
gamma-muurolene	1476	1478	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.7	0.9	1.0
alpha-humulene	1452	1452	0.9	0.4	0.5	0.5	0.6	0.7	0.7	0.7	0.7	0.6	0.8	0.7	0.7	0.7	0.6	0.7	0.5	0.6	0.5
alpha-muurolene	1500	1500	0.4	0.4	0.5	0.5	0.2	0.5	0.6	0.6	0.5	0.6	0.4	0.5	0.7	0.6	0.5	0.6	0.5	0.5	0.5
gamma-cadinene	1514	1513	0.4	0.5	0.5	0.5	0.2	0.5	0.5	0.7	0.5	0.6	0.4	0.5	0.7	0.6	0.5	0.5	0.4	0.6	0.6
germacrene A	1504	1508	0.5	0.1	0.3	0.2	0.2	0.3	0.3	0.2	0.3	0.3	0.2	0.3	0.3	0.3	0.3	0.4	0.0	0.4	0.6
beta-elemene	1391	1389	0.4	0.0	0.2	0.2	0.2	0.2	0.3	0.2	0.3	0.2	0.3	0.3	0.3	0.4	0.3	0.4	0.2	0.2	0.0
Alloaromadendrene	1459	1458	0.2	0.2	0.2	0.1	0.1	0.2	0.2	0.2	0.3	0.2	0.2	0.2	0.3	0.3	0.3	0.2	0.0	0.2	0.0
beta-cubebene	1389	1387	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.2	0.1	0.1	0.3	0.2	0.2	0.3	0.2	0.0	0.2	0.1

delta-amorphene	1507	1511	0.2	0.1	0.2	0.2	0.0	0.2	0.2	0.3	0.2	0.2	0.0	0.0	0.2	0.2	0.0	0.1	0.0	0.0	0.0
trans-muurola-3,5-diene	1448	1451	0.2	0.0	0.1	0.1	0.0	0.1	0.1	0.2	0.1	0.2	0.0	0.1	0.2	0.2	0.2	0.2	0.0	0.0	0.0
alpha-cadinene	1537	1537	0.1	0.1	0.2	0.2	0.0	0.2	0.2	0.2	0.1	0.2	0.1	0.1	0.2	0.2	0.2	0.2	0.0	0.0	0.0
beta-copaeno	1428	1430	0.1	0.0	0.1	0.1	0.0	0.1	0.2	0.2	0.2	0.2	0.1	0.2	0.2	0.2	0.2	0.2	0.0	0.2	0.0
beta-selinene	1487	1489	0.2	0.1	0.1	0.1	0.0	0.0	0.0	0.2	0.0	0.0	0.1	0.1	0.0	0.0	0.1	0.2	0.0	0.0	0.0
trans-cadina-1,4-diene	1532	1533	0.1	0.0	0.0	0.0	0.0	0.0	0.1	0.2	0.1	0.2	0.0	0.0	0.2	0.2	0.0	0.0	0.0	0.0	0.0
alpha.-Cubebene	1349	1345	0.2	0.0	0.1	0.1	0.0	0.1	0.2	0.2	0.2	0.2	0.1	0.1	0.2	0.2	0.2	0.2	0.0	0.0	0.0
gamma-elemene	1433	1434	0.2	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.1	0.1	0.2	0.1	0.1	0.1	0.1	0.2	0.0	0.0	0.0
Aromandendrene	1437	1439	0.2	0.0	0.0	0.1	0.0	0.1	0.1	0.0	0.0	0.2	0.0	0.1	0.2	0.1	0.1	0.2	0.0	0.0	0.0
ni	1835		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	1.8	0.9
eugenol	1357	1356	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	1.1	0.3	0.0	0.0	0.0	0.0	0.0
6-methyl-alpha-ionone	1522	1520	0.3	0.2	0.3	0.2	0.9	0.3	0.3	0.0	0.2	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0
cryptone	1185	1183	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.1	0.6	0.5	0.2	0.3	0.6	0.3	0.1	0.8	0.7
ni	1222	1195	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.2	0.5	0.1	0.0	0.5	0.2	0.0	0.6	0.7
ni	1295		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.2	0.1	0.0	0.4	0.3
ni	1170		0.2	0.0	0.0	0.0	0.1	0.0	0.0	0.2	0.0	0.0	0.3	0.1	0.1	0.2	0.1	0.1	0.0	0.0	0.0
Monoterpene hydrocarbons		MH	50.9	60.1	51.4	54.2	62.7	53.5	50.6	49.5	51.6	45.7	42.8	38.9	39.9	38.9	39.3	41.1	21.8	20.9	28.8
Oxygenated monoterpenes		OM	5.6	2.3	3.2	3.1	2.5	2.8	3.7	6.6	4.5	3.9	6.1	8.0	5.5	6.2	8.3	6.3	3.7	7.4	9.8
Sesquiterpene hydrocarbons		SH	21.3	20.8	21.3	20.2	16.5	23.2	24.0	20.7	25.4	23.7	20.4	21.3	28.4	24.0	22.2	21.0	27.4	21.4	20.9
Oxygenated sesquiterpenes		OS	20.3	16.5	23.7	21.6	17.1	20.2	21.4	22.2	18.2	26.3	28.9	28.4	23.5	29.0	26.3	29.9	46.9	44.2	37.6
Total identified			98.7	99.9	100	99.3	99.7	99.9	99.9	99.3	100	99.7	98.9	97.1	98.5	98.9	96.6	98.7	99.9	94.7	97.8

The chemical profile was analyzed by GC-MS and organized in the table in order of elution in the capillary chromatographic column (5% phenyl, 95% dimethylpolysiloxane). The concentration (%) was calculated based on the total peak area by GC-FID. LRI represents the literature and calculated (relative to n-alkanes C8-C20) linear retention indices. Ni – not identified. The numbers one to six represent stored times by month. CTR – control; CR – cold chamber or refrigerator (5 ±1 º), FZ – freezer (-15±3 ºC); RO – room temperature in shade (29 ±3 ºC).

Table S1. Compositions of essential oil from dried pink pepper fruits stored under different conditions (room temperature, cold chamber/refrigerator and freezer) and time periods (six months).

4 I CONCLUSION

Storage conditions did not provide significant changes in the chemical composition of fixed oils and essential oils. On the other hand, storage time caused changes in the essential oils chemical composition from the third month of storage, with an increase in oxygenated terpenes and a reduction in monoterpene hydrocarbons. There was also a reduction in the essential oil content in the sixth month of storage. Finally, we conclude that pink pepper fruits can be stored for up to 3 months at room temperature (29 ±3 °C), protected from humidity and direct light without significant change in the quality or quantity of fixed and essential oils.

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