

Identification of compounds present in two different Caatinga medicinal plants obtained in free markets in Senhor do Bonfim – BA

Análise dos compostos de duas plantas medicinais da Caatinga obtidas na feira livre de Senhor do Bonfim – BA

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ABSTRACT

Medicinal plants of Brazilian native flora are consumed with little or no proof of its pharmacological properties, based only on users' or traders' reports. Although they are natural and, therefore, popularly considered free of side effects, medicinal plants' toxicity is a serious public health concern, either because of the plant's own compounds, either by adulterations or interaction with other medicines. Since these medicinal plants are usually sold in markets and free fairs by people without formal education, there may be labeling or manipulation problems, resulting in the absence of the desired effects or presence of adverse effects. In this work, compounds present in different medicinal plants samples found in Senhor do Bonfim free market (BA) were identified: six samples of Sene (*Senna occidentalis*; Fabaceae) and six samples of Umburana (*Amburana cearensis*; Fabaceae). For the compounds extraction of *S. occidentalis*, hydrodestillation was used, and for *A. cearensis* the compounds were extracted using maceration with hexane. All extracts were analyzed by GC-MS. Compounds in both plants had a high variation among samples, which can mean either that some of the medicinal plants sold in free markets belong to a species different from the announced one or, at least, that compounds found in the sample had degraded between harvest and sale.

Keywords: medicinal plants, essential oil, umburana, sene, Caatinga.

RESUMO

As plantas medicinais da flora nativa brasileira são consumidas com pouca ou nenhuma comprovação de suas propriedades farmacológicas, baseadas apenas em relatos de usuários ou comerciantes. Apesar de serem naturais e, portanto, popularmente consideradas livres de efeitos colaterais, a toxicidade das plantas medicinais é um sério problema de saúde pública, seja pelos compostos da própria planta, seja por adulterações ou interação com outros medicamentos. Como essas plantas medicinais costumam ser vendidas em mercados e feiras livres por pessoas sem educação formal, pode haver problemas de rotulagem ou manipulação, resultando na ausência dos efeitos desejados ou na presença de efeitos adversos. Neste trabalho, compostos presentes em diferentes amostras de plantas medicinais encontradas na feira livre de Senhor do Bonfim (BA) foram identificados: seis amostras de Sene (*Senna occidentalis*; Fabaceae) e seis amostras de Umburana (*Amburana cearensis*; Fabaceae). Para a extração dos compostos em *S. occidentalis*, foi utilizada a hidrodestilação, e para *A. cearensis* os compostos foram extraídos por maceração em hexano. Todos os extratos foram analisados por GC-MS. Os compostos em ambas as plantas tiveram uma alta variação entre as amostras, o que pode

significar ou que algumas das plantas medicinais vendidas nas feiras livres pertencem a uma espécie diferente da anunciada ou, pelo menos, que os compostos encontrados na amostra haviam degradado entre a coleta e a venda.

Palavras-chave: plantas medicinais, óleo essencial, umburana, sene, Caatinga.

1 INTRODUCTION

Since ancient times, humans have used plants for medicinal purposes, for the treatment, cure, and prevention of diseases. In the early 1990s, the World Health Organization reported that 65-80% of the population in developing countries used medicinal plants as the only way to treat diseases (Akerele, 1993). These plants are marketed supported by advertisements that promise safe benefits, since it is a natural source. However, the alleged pharmacological properties were often not verified in clinical or pre-clinical tests. This appeal for the consumption of natural products continues to increase, in the belief that these plants, used for centuries or even millennia, are safe for the population (Veiga Junior, Pinto e Maciel, 2005).

A survey carried out in the USA in 1997 showed that 42% of respondents had used medicinal plants at least once in the previous year, in alternative treatments. This percentage is about 33.8% higher than that obtained in the previous survey, from 1990 (Eisenberg *et al.*, 1998). In Germany, medicinal plants are used by the population to treat various diseases (Calixto, 2000), and self-medication with preparations based on medicinal plants is very common. In addition to self-medication, 70% of German general practitioners prescribe hundreds of herbs licensed in that country (Blumenthal, Busse e Bundesinstitut für Arzneimittel und Medizinprodukte (Germany), 1998).

In Brazil, medicinal plants from native flora are consumed with little or no evidence of their pharmacological properties, based only on reports from users or traders. Often, they are even used to treat diseases different from those for which the foresters used them. Despite being natural and, in popular opinion, free from side effects, the toxicity of medicinal plants is a serious public health problem, either because of the plant compounds themselves, by adulterations of the samples or by interaction with other drugs (Veiga Junior, Pinto e Maciel, 2005). Data from the National System of Toxic-Pharmacological Information-SINITOX record the occurrence of 8,501 cases of intoxication by plants in Brazil from 2004 to 2008. Of these intoxications, 12.4% were related to intentional circumstances in which the victim sought pharmacological

properties of the plant (SINITOX, 2010). Research carried out in Brazil to assess the safe use of medicinal plants is still incipient, as is the commercialization control by official bodies (Veiga Junior, Pinto e Maciel, 2005).

Since these medicinal plants are usually sold in markets and open fairs, by people without adequate formal education, there may be a problem of labeling or manipulation, resulting in the absence of the desired effects or even the presence of adverse effects (Amaral, do *et al.*, [s.d.]; Boer, de, Ichim e Newmaster, 2015; Veiga Junior, Pinto e Maciel, 2005). In addition, the so-called “common names” of plants can be attributed to more than one species, consequently grouping different organisms with different metabolites into the same category of “medicinal plant”, resulting in different or unexpected bioactivity (Andrade, Silva e Trigueiros, 2017).

Another factor that must be considered is the poor quality of some commercialized medicinal plants. These plants should have a more rigorous control regarding the knowledge of the plants’ origin, time and form of collection, storage, drying, conditioning, and contamination (Ustulin *et al.*, 2009). It is common to find epiphytic organisms on the surface of leaves and trunks, in addition to symbiotic organisms on roots, such as fungi (Amaral, do *et al.*, [s.d.]; Rocha, Soares e Corrêa, 2004). If there is no correct handling, these organisms can be ingested by the consumer, altering the therapeutic effect of the medicinal plant (Andrade, Silva e Trigueiros, 2017; Veiga Junior, Pinto e Maciel, 2005). Even contamination by heavy metals has been reported in the literature (Veiga Junior, Pinto e Maciel, 2005).

Since 1995, many efforts have been made in Brazil to encourage studies with medicinal plants and promote growth in the pharmaceutical sector. Unfortunately, despite the vast biodiversity of Brazilian flora, most of the medicinal plants studied and used in the preparation of medicines are not native from Brazil, due to the lack of scientific studies that prove their effects (Brandão, 2009).

Several studies have analyzed the usage of medicinal plants in different regions of Brazil, such as surveys performed at Estação Ecológica do Jataí, in the municipality of Santo Antônio-SP (Castellucci *et al.*, 2000), Cidade de Goiás and Pirenópolis-GO (Rizzo, 1999), Tanquinho -BA (Costa-Neto e Oliveira, 2000), Mossâmedes-GO (Vila Verde, Paula e Caneiro, 2003), Santa Tereza-ES (Medeiros, Silva e Senna-Valle, 2004), Campo Grande-MS (Nunes *et al.*, 2003), Goiânia, Anápolis, Trindade and Aparecida de Goiânia-GO (Tresvenzol *et al.*, 2006), Bezerros-PE (Lira, Sousa e Lins, 2020), São João do

Paraíso-MA (Parente *et al.*, 2022), Coelho Neto-MA (Mesquita *et al.*, 2022) and Pesqueira-PE (Cavalcanti, Andrade e Lima, 2020).

Among the Brazilian native species used by the population in the treatment of diseases, there is *Amburana cearensis*, which is a species native to the Caatinga biome at Northeast Brazil, resistant to heat stresses and presenting good adaptation to the climate and belongs to the Fabaceae (Leguminosae) family and Faboideae subfamily (Pereira, 2010). It is a tree whose erect stem can reach 10 meters in height in the Caatinga and about 20 meters in the Zona da Mata (Cunha e Ferreira, 2003; Guedes *et al.*, 2010). Its roots, bark and seeds are used to fight and prevent diseases, mainly through the elaboration of home remedies made for the relief of respiratory system problems and anti-inflammatory activity (Silva *et al.*, 2020).

Senna occidentalis (L.) Link, also used as a medicinal plant, is an herbaceous species native to the Americas belonging to the Fabaceae (Leguminosae) family and Caesalpinioideae subfamily and has been used medicinally by Native Americans and African tribes as tonic, antipyretic, stomachic and laxative (Jain *et al.*, 1998). Furthermore, according to Tona and coworkers (2004), it is used in the Amazon and Africa for its antimalarial effects.

2 EXPERIMENTAL

2.1 FIELD OF STUDY

The study area comprised the open market in the municipality of Senhor do Bonfim, which is geographically located under the coordinates 10° 28' 00" south latitude and 40° 11' 00" west longitude of the Greenwich meridian (Ramalho, 2008). The climate recorded in the region is tropical semi-arid and the average annual temperatures oscillate between 24° and 33°C degrees. The market in the municipality of Senhor do Bonfim is the second largest in the northeast Brazil, with 1.2 km in length, attracting thousands of consumers every Saturday (Silva, 2021). Free markets have always represented a significant part of local commerce and have great cultural and economic importance for the region.

2.2 SAMPLE COLLECTION

First a survey was performed in the market in order to compile informations regarding the common names of the plants sold at eight different points in the city fair, considering their nature, their use, and if they were native to the caatinga biome. Two

species of interest were selected: umburana (*A. cearensis*) which is indicated, according to the sellers, to treat illnesses such as stomachache, headache, fight and prevent fevers, regulate menstrual periods, and act as an anti-inflammatory, in addition to many other benefits; and sena (*S. occidentalis*), which, according to information provided by the sellers, is recommended for constipation and headache. The selected plants were purchased and taken to the laboratory for the preparation of extracts.

2.3 COMPOUNDS EXTRACTION

To extract *A. cearensis* compounds, the seeds samples were weighed, and the biomass was then macerated using a mortar and pestle. After the maceration, 50 mL of hexane solvent were added, and this mixture was left to rest at room temperature for 30 minutes. The mixture was then filtered through filter paper into an Erlenmeyer. The process was repeated three times with 20 mL of the solvent and the extracts were combined. NaSO₄ was added to the Erlenmeyer to remove the water present in the extract, then the extract was transferred to another container and the solvent was evaporated. Subsequently, a fraction of 10 μ L of the concentrated extract was diluted again in 1mL of hexane and stored in freezer until analysis by gas chromatography coupled with mass spectrometry.

For the extraction of *S. occidentalis* essential oils, the leaves samples were weighed, transferred to a round-bottomed flask with approximately 300mL of distilled water, and attached to the Clevenger apparatus. A heating mantle was used to heat each sample at a temperature of around 100°C for 8 h over a period of two days. At the end of each day, the extracted compounds were collected in a vial. After extraction, a fraction of about 10 μ L was dissolved in hexane and stored in the freezer for GC-MS analysis.

Analysis of the extracts

The GC-MS analyzes were performed in splitless mode on an Agilent® GCMS 7890 equipment equipped with a DB-5 capillary column. The temperature for all analyzes ranged from 50°C to 300°C with an increment of 3°C per minute.

3 RESULTS AND DISCUSSION

3.1 AMBURANA CEARENSIS

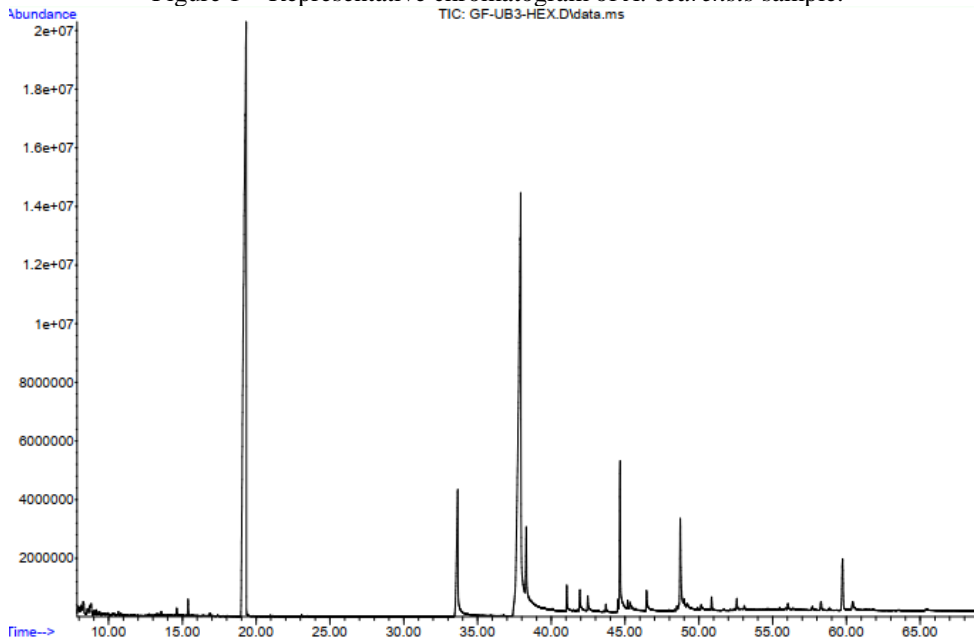
The samples were numbered from AC-1 to AC-6 according to the tent from which they were obtained. The yields calculated after the extraction are shown in table 1.

Table 1 – Extract yields obtained from samples of *A. cearensis*.

Sample	Sample mass (g)	Extract obtained (g)	Yield (%)
AC-1	8.58	1.82	21,2
AC-2	11.17	1.87	16,7
AC-3	5.02	0.89	17
AC-4	10.97	2.98	27
AC-5	9.77	2.19	22,45
AC-6	9.56	2.26	23,7

The identification of *A. cearensis* extracts was carried out using GC-MS analysis, and a representative chromatogram of one sample is shown in Figure 1.

Figure 1 – Representative chromatogram of *A. cearensis* sample.



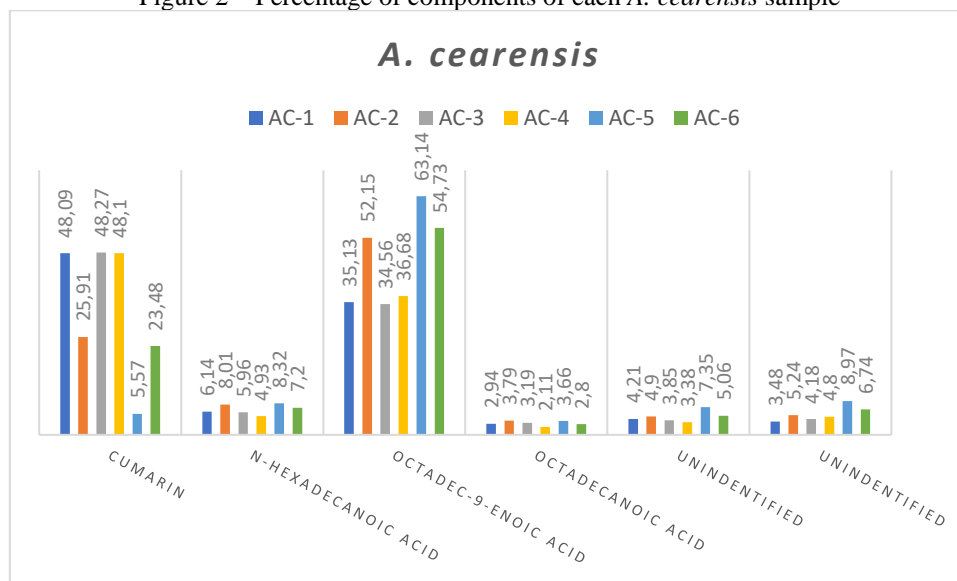
The mass spectra of the major compounds of the extracts were compared with the literature (Adams, 2007). From these data, the possible constituents of the samples were identified. Table 2 presents the major components and their percentage in each of *A. cearensis* samples.

Table 2 – Percentage of each compound found in *A. cearensis* extracts

Compound	AC-1	AC-2	AC-3	AC-4	AC-5	AC-6
Coumarin	48,09	25,91	48,27	48,1	5,57	23,48
n-hexadecanoic acid	6,14	8,01	5,96	4,93	8,32	7,2
octadec-9-enoic acid	35,13	52,15	34,56	36,68	63,14	54,73
octadecanoic acid	2,94	3,79	3,19	2,11	3,66	2,8
unidentified	4,21	4,9	3,85	3,38	7,35	5,06
unidentified	3,48	5,24	4,18	4,8	8,97	6,74

Coumarin was the major compound of samples AC-1, AC-3 and AC-4 (48,09 to 48,27%), followed by octadec-9-enoic acid (34,56 to 36,68%). In samples AC-2 and AC-6, octadec-9-enoic acid was the major compound (52,15 and 54,73%, respectively) and coumarin was the second major compound (25,91 and 23,48%, respectively). On the other hand, the sample AC-5 showed the most discrepant result of all samples, with octadec-9-enoic acid as the major component (63,14%) and with only 5,57% of coumarin. Figure 2 shows the percentage of compounds present in each sample.

Figure 2 – Percentage of components of each *A. cearensis* sample



Works such as the one by Almeida and coworkers (2011), which highlights the chemical and pharmacological composition of *A. cearensis*, shows that coumarin was the major compound in its ethanolic extracts and has hypotensive, antimicrobial, anti-inflammatory, antitumor, anti-malarial, leishmanicidal and anti-chagasic activities already attributed.

Therefore, samples with other compounds in quantities greater than coumarin or, as in the case of AC-5, in which coumarin appears in very low quantities, may

demonstrate that samples sold as Umburana may have been misidentified or, alternatively, they have already had their compounds degraded after inadequate storage for a long time.

3.2 SENNA OCCIDENTALIS

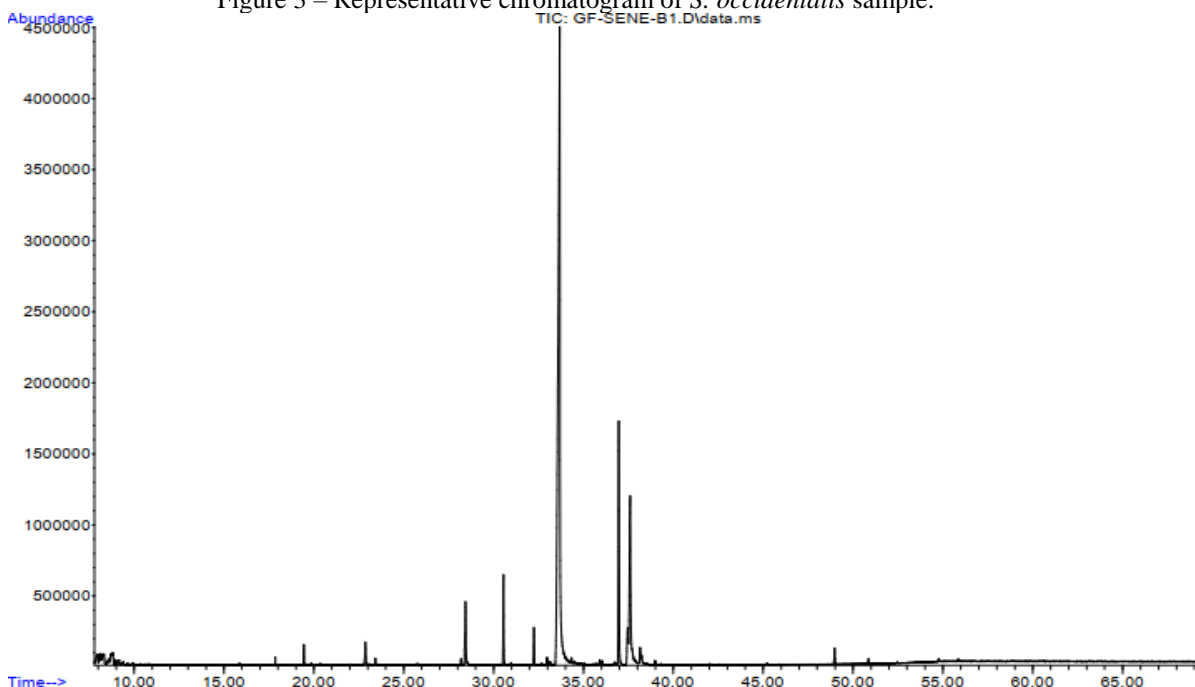
The samples of *S. occidentalis* were numbered from S1 to S6 according to the tent from which they were obtained. The yields calculated after the hydrodistillation extraction are shown in table 3.

Table 3 – Extract yields obtained from samples of *S. occidentalis*.

Sample	Sample mass (g)	Essential oil obtained (g)	Yield (%)
SO-1	10,71 g	0,057 g	0,53%
SO-2	11,47 g	0,082 g	0,71%
SO-3	09,18 g	0,027 g	0,29%
SO-4	10,17 g	0,062 g	0,60%
SO-5	11,52 g	0,064 g	0,55%
SO-6	11,03 g	0,060 g	0,54%

The identification of *S. occidentalis* essential oil was carried out using GC-MS analysis, and a representative chromatogram of one sample is shown in Figure 3.

Figure 3 – Representative chromatogram of *S. occidentalis* sample.



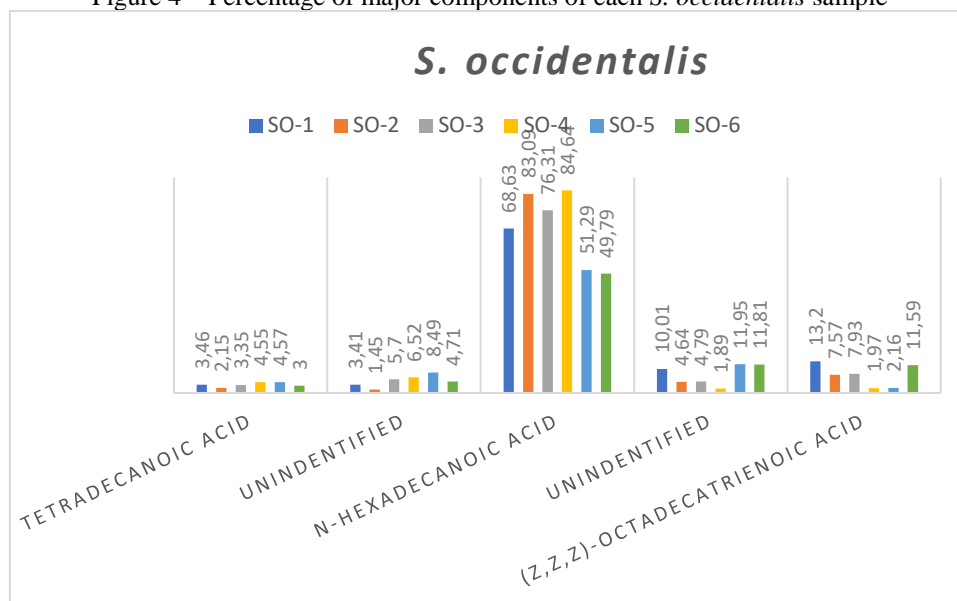
The mass spectra of the essential oil compounds were compared with the literature (Adams, 2007). From these data, the possible constituents of the samples were identified. Table 4 presents all components and their percentage in each of *S. occidentalis* samples.

Table 4 – Percentage of each compound found in *S. occidentalis* essential oil.

Compound	SO-1	SO-2	SO-3	SO-4	SO-5	SO-6
Cariofilene	--	--	--	--	1,73	--
(E)-6,10-dimethyl-5,9-undecadien-2-one	--	--	--	--	1,91	2,82
dodecanoic acid	--	--	--	--	2,09	1,26
unidentified	--	--	--	--	2,28	--
ar-tumerone	--	0,7	--	--	2,03	--
tetradecanoic acid	3,46	2,15	3,35	4,55	4,57	3
unidentified	3,41	1,45	5,7	6,52	8,49	4,71
bis(2-methylpropyl) 1,2-Benzenedicarboxilic ester	--	--	--	--	0,61	--
pentadecanoic acid	--	--	--	--	0,33	--
unidentified	1,3	0,4	--	--	--	--
6,10,14-trimehyl-5,9,13-pentadecatrien-2-one	--	--	--	--	9,13	6,51
methyl hexadecanoate	--	--	--	--	0,43	--
unidentified	--	--	--	--	0,49	--
n-hexadecanoic acid	68,63	83,09	76,31	84,64	51,29	49,79
ethyl hexadecanoate	--	--	--	--	0,51	--
unidentified	--	--	--	0,44	--	--
unidentified	10,01	4,64	4,79	1,89	11,95	11,81
unidentified	--	--	1,92	--	--	--
(Z,Z)-9,12-octadecadienoic acid	--	--	--	--	--	3,52
(Z,Z,Z)-octadecatrienoic acid	13,2	7,57	7,93	1,97	2,16	11,59

N-hexanoic acid was the major compound of all samples, but there were great differences in the relative amount of this compound (49.79 to 84.64%). The second major compound in samples SO-1, SO-2 and SO-3 was (Z,Z,Z)-octadecanoic (13.2, 7.0 and 7.93%, respectively), although this compound was present in very low amounts in samples SO-4 (1.97%) and SO-5 (2.16%). In sample SO-6, (Z,Z,Z)-octadecanoic acid was the third major component, but in similar amount of SO-1 (11.59%). Some major components could not be identified. Figure 4 shows the percentage of major compounds present in each sample.

Figure 4 – Percentage of major components of each *S. occidentalis* sample



The results show that there is a significant variance in the amount of compounds extracted, considering that they are plants of the same species and from different tents. This distinction may be due to some factors. For instance, as pointed out by (Souza-Moreira, Salgado e Pietro, 2010), the chemical composition of essential oils is determined by genetic factors, but other factors can also cause significant changes in the production of secondary metabolites. The stimulus produced by the plant environment can alter the metabolic pathway, leading to the synthesis of different compounds. Among these factors, plant-microbe, plant-insect, and plant-plant interactions, in addition to age and developmental stage, luminosity, temperature, water, nutrition, and collection time.

Another factor that must be considered is the poor quality of some commercialized medicinal plants. They don't have a rigorous control regarding the knowledge of the origin of the plants, time and form of collection, storage, drying, conditioning, and contamination (Ustulin *et al.*, 2009). Some of the samples studied may be old and poorly stored, so the compounds have already started deteriorating. Despite the origin of the differences found, they reveal that the *S. occidentalis* plants sold in the tents of the Senhor do Bonfim fair have different compounds and, therefore, neither their efficacy nor their safety can be guaranteed.

4 CONCLUSIONS

The results show that some of the medicinal plants commercialized in the free markets may be of poor quality, old or misidentified. As samples AC-5, AC-2 and AC-6

of *A. cearensis* showed lower amounts of coumarin, it is expected that these plants don't show the expected bioactivity. Samples SO-5 and SO-6 of *S. occidentalis* may be old and poorly stored, because of the lower amount of n-hexadecanoic acid, that may have already started deteriorating.

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