Journal of Applied Research on Medicinal and Aromatic Plants xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Journal of Applied Research on Medicinal and Aromatic Plants

journal homepage: www.elsevier.com/locate/jarmap

Short communication

Antioxidant capacity, proximate composition, and lipid constituents of *Aloe vera* flowers

Jaime López-Cervantes[a], Dalia I. Sánchez-Machado[a,*], Paola Cruz-Flores[a], María F. Mariscal-Domínguez[a], Gabriela Servín de la Mora-López[b], Olga N. Campas-Baypoli[a]

[a] Departamento de Biotecnología y Ciencias Alimentarias, Instituto Tecnológico de Sonora, 5 de febrero 818 Sur, 85000 Ciudad Obregón, Sonora, México
[b] Facultad de Ciencias Químico-Biológicas, Universidad Autónoma de Sinaloa, Ciudad Universitaria, AP 354, 80000 Culiacán, Sinaloa, México

ARTICLE INFO

Keyword:
Flowers
Aloe vera
α-Tocopherol
Lipid
HPLC

ABSTRACT

This study presents the chemical characteristics of flowers of *Aloe vera* as ingredient in food. The α-tocopherol, β-carotene, and β-sitosterol content as well as the oleoresin extraction for *Aloe vera* (L.) Burm f. flowers were evaluated using two agitation methods and four solvents. Then tocopherol, β-carotene, and β-sitosterol were quantified using an HPLC method. The statistical analysis revealed that the optimal extraction method for α-tocopherol was obtained by vortex agitation and acetone or ethyl acetate extraction, while for β-carotene, it was with acetone alone with no significant difference between agitation methods For β-sitosterol, the optimal method of extraction was acetone and vortex agitation. A proximate analysis of the dried Aloe flowers showed the average moisture, protein, lipid, fibre, and ash content to be 8.45%, 11.75%, 2.30%, 12.65%, and 8.07%, respectively. The best oleoresin extraction yield was by sonication using hexane as a solvent. Furthermore, Aloe flowers present an average of 30.71% inhibition for radical scavenging activity. Myristoleic acid was the most predominant fatty acid in the flowers, whereas arachidonic acid was the least abundant. Consequently, *Aloe vera* flowers were characterized to find their nutritional value and possible application in the food and pharmaceutical industry.

1. Introduction

Aloes are xerophytes belonging to the Liliaceae family, and consisting of more than 360 species, which exist in a variety of different habitats due to their adaptive capacity for survival (Choi and Chung, 2003). The majority of Aloe species is originally from the tropical and subtropical latitudes of Africa, but can be found in abundance on the Arabian Peninsula and in Madagascar. Many countries have endemic Aloe species with Madagascar being the number one country with the highest rate of endemism overall. However, the widely propagated *Aloe vera* plant has become established in many tropical and sub-tropical countries, including the Americas (Newton, 2004). Currently, México holds the number one spot for the cultivation of Aloe species with around 14 000 ha and about 36% of worldwide production. In particular, the northeastern region of México has the greatest cultivated surface area at 89.9% of the total nation (Moreno et al., 2012). The widespread nature and multiple beneficial properties of Aloe have led to its popular usage as a medicinal plant and for centuries has been used as a medicinal herb for healing people and animals (Sedaghathoor et al., 2017). Thus, Aloe species have become of great interest to researchers who have tried to identify the compounds responsible for these beneficial effects.

Diverse bioactive compounds have been identified in different parts of the Aloe plant including glycoproteins, polysaccharides, amino acids, anthraquinones, antioxidant compounds, and vitamins, such as A, E, and B_{12}. Most of the previous research has been conducted with the Aloe gel and latex (Kanama et al., 2015; Wyk et al., 1995). The Aloe gel contains carbohydrates linked to therapeutic characteristics, such as wound healing and anti-inflammatory activity, while the Aloe latex is comprised of the majority of the anthraquinones present in the whole leaf with therapeutic claims for anti-inflammatory activity, purgative action, and as an antioxidant (Choi and Chung, 2003).

All Aloe plants have the additional attraction of mass flower production throughout the year. However, of all the Aloe parts, the flower has seldom been studied and the biological compounds remain mostly unknown. Most Aloe species produce diurnal and tubular brightly colored flowers, usually red or yellow (Newton, 2004). The use of edible flowers has gained popularity, as seen in European and Asian cuisine, containing a diverse range of natural antioxidants.

A diet rich in antioxidants can reduce the risk of cardiovascular and

* Corresponding author.
E-mail address: dalia.sanchez@itson.edu.mx (D.I. Sánchez-Machado).

https://doi.org/10.1016/j.jarmap.2018.02.004
Received 26 October 2017; Received in revised form 9 January 2018; Accepted 12 February 2018

chronic diseases, as well as cancer (Loizzo et al., 2015). There is sufficient evidence for the protective role of vitamin E against cardiovascular diseases, DNA damage by free radicals and oxidation, and skin disorders. However, tocopherols, which can accumulate in the flowers, are part of the vitamin E group and can only be synthesized by a photosynthetic organism (Arrom and Munné-Bosch, 2010). Several studies of edible flowers have reported the carotenoid and α-tocopherol composition in *Capparis spinosa* flowers (Tlili et al., 2009). Studies related to *Aloe vera* flowers include the distribution of lectins in different parts of the Aloe flower (Vakhania et al., 2014), as well as the amino acid profile and antinutritional factor (Sotelo et al., 2007). To our knowledge, there has not been any previous research of Aloe flowers on the characterization and quantification of active compounds with antioxidant activity or their fatty acid profile. Fatty acids have been shown to exhibit biological functions in the skin such as prevention of trans epidermal water loss, maintenance of the stratum corneum epidermal barrier, and disruption of melanogenesis in epidermal melanocytes (Dorni et al., 2017).

The quantification of these compounds will generate knowledge about possible uses for the abundant Aloe flowers. Therefore, in this study we generated a complete proximal analysis of the flowers. Then we identified and quantified α-tocopherol, β-carotene, and β-sitosterol by HPLC in the samples as well as the oleoresin of the flowers. A fatty acid profile was also generated by gas chromatography.

2. Materials and methods

2.1. Chemicals and standards

HPLC-grade acetonitrile and methanol were purchased from J.T. Baker (Center Valley, PA, USA). Analytical-grade pyrocatechol, methanol, toluene, potassium carbonate, Trizma base, 2,2-diphenyl-1-pikryl-hydrazyl (DPPH), MES hydrate, and 6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox) were obtained from Sigma (St. Louis, MO, USA). The standards, α-tocopherol, β-carotene, and β-sitosterol, and sodium sulfate were supplied by Fluka (Milwaukee, WI, USA). Potassium hydroxide and boric acid were purchased from Fermont (Monterrey, Nuevo León, México). Hexane (HEX), acetone (AC), ethyl acetate (EAC), petroleum ether (PE) and chloroform were purchased from Fagalab (Guamúchil, Sinaloa, México). Sulfuric acid and sodium hydroxide were obtained from Productos Monterrey (Monterrey, Nuevo León, México). Selenium reagent was purchased from Merck (Kenilworth, NJ, USA). Fatty acid standards, FAME Mix C8-C22 and PUFA No.3 were obtained from Supelco (Bellefont, PA, USA). All reagents were analytical grade, unless otherwise noted. All aqueous solutions were prepared with ultrapure water purified with a Nanopure Diamond UV system (Barnstead International, Dubuque, Iowa, USA).

Alpha-tocopherol (0.376 mg/ml), β-carotene (0.014 mg/ml), and β-sitosterol (0.2 mg/ml) standard solutions were prepared with HPLC grade methanol and stored at 4 °C, away from light. Working solutions were prepared from these standard solutions and diluted with methanol. For quantification, the peak heights were correlated with the concentration according to the calibration curve. All samples were analyzed in duplicate. Alpha-tocopherol, β-carotene, and β-sitosterol content are presented as means ± the standard deviation.

2.2. Samples

Aloe vera (L.) Burm f. flowers were harvested from two regions in south Sonora in winter of 2015, México. Three samples obtained from intensively managed fields (latitude: N 27° 6′ 24.3′′, longitude: O 109° 28′ 52.7′′, altitude: 33 m), and five samples from wild plants (latitude: N 27° 37′ 51.3′′, longitude: O 109° 54′ 13.6′′, altitude: 41 m) in their natural habitat were analyzed. To obtain a random sample, the managed field was separated into five sections. A collection of all racemes from the inflorescence of 10 aloe plants was pooled to obtain managed field sample 1, 2, 3, 4 and 5. In the case of wild flowers, the same procedure was used to separate three sections and pool samples, wild grown 1, 2 and 3. Wild grown flowers (1,2,3,4,5) were collected in the same day, as well as managed field flowers (1,2,3). However, the managed field flowers collection was done one week after the wild grown. All were dried in the sunlight, conserved separately in airtight plastic bags covered with aluminum foil, and stored at room temperature (21 °C). Before the analysis, the flower was ground using a blender and sieved (0.0059 inches).

2.3. Proximate analysis of Aloe flower powders

The moisture content of the Aloe flower powder was determined by drying each sample to a constant weight at 60 °C (AOAC, 1995). Then, the nitrogen content was determined using the Rapid Kjeldahl method (Labconco, Model 652, Labconco Corp., Kansas City, MO, USA) and the protein content was calculated by multiplying the percentage of nitrogen by a factor of 6.25 (AOAC, 1995). After that, the ash content was gravimetrically estimated by incineration at 550 °C (AOAC, 1995). Then the lipids were extracted from samples with a 2:1 chloroform-methanol mixture, according to the method described by Sánchez-Machado et al. (2004), with some modifications. Specifically, 0.5 g of the dried sample was extracted with 3 ml of the solvent mixture, and after 2 min of vortex agitation, the content was filtered through Whatman paper No. 41. The process was repeated once more with the obtained residue. The two extracts were then combined and concentrated to dry in an oven at 60 °C. The weight of the resulting residue is reported as the total lipid content of the sample. The dietary fibre content was determined by an enzymatic-gravimetric method, using a MES-Tris buffer for total dietary fibre (Sullivan, 1993) with some modifications, specifically in using 0.5 g of the dried Aloe flowers, with residual protein and residual ash correction.

2.4. Sample preparation

Alpha-tocopherol, β-carotene, and β-sitosterol were extracted from dried Aloe flowers using the method of Sánchez-Machado et al. (2002), with some modifications. For saponification, a sample of 1.0 g was weighed out in a screw-top assay tube. Then, 200 µl of pyrocatechol, as an antioxidant, and 6 ml of KOH solution (0.5 M in methanol) were added, and immediately vortexed for 20 s. The tubes were placed in a water bath at 80 °C for 15 min and vortexed every 5 min for 15 s. After cooling in an ice bath, 1 ml of distilled water and 6 ml of hexane were added. The mixture was immediately vortexed for 1 min, then centrifuged for 2 min at 425 × *g*. Then 3 ml of the upper phase were transferred to a new test tube and dried under nitrogen. The residue was redissolved in 1 ml of the HPLC mobile phase (60:40 methanol:acetronitrile (v/v)) and filtered through a 0.45-µm pore size membrane filter before the analysis of α-tocopherol, β-carotene, and β-sitosterol by HPLC.

2.5. Extraction and quantification of the oleoresin yield

Extraction was performed using four solvents, acetone (AC), ethyl acetate (EAC), hexane (HEX), and petroleum ether (PE), and two methods of agitation, vortex (V) and sonication (S). The extraction procedure for oleoresins was followed in accordance to López-Cervantes et al. (2014) with some modifications. For each extraction, 1.0 g of sample was weighed into a tube, 10 ml of solvent was added, and the mixture was left to stand for 10 min. The tubes were then stirred for 5 min. After the first extraction, colored liquor was obtained and was filtered through Whatman paper No.41. The residue was treated a second time with 5 ml of solvent. To evaporate the solvent, the total extract was transferred to another test tube and dried under nitrogen. After the removal of the solvents, the extract obtained was referred to as oleoresin. To estimate the extraction efficiency, the