Chemical composition of the essential oil from leaves of *Chenopodium ambrosioides* L. grown in Recife-PE, Brazil

Composição química do óleo essencial de folhas de *Chenopodium ambrosioides* L. cultivadas em Recife-PE, Brasil

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RESUMO

Chenopodium ambrosioides L. é uma erva aromática amplamente distribuída ao longo da América, África e também em países da Europa. O óleo essencial das folhas de *C. ambrosioides* é o componente mais usado na medicina popular. Com o objetivo de conhecer a sua composição química, o óleo essencial das folhas da espécie, cultivada em Recife-PE, Brasil, foi extraído por hidrodestilação e analisado por cromatografia gasosa acoplada à espectrometria de massas. O rendimento médio do óleo foi de 0,7% ± 0,1%. Os componentes majoritários encontrados foram α-terpineno (42,14%), α-terpinenil-acetato (31,57%) e timol (7,90%).

Palavras-chave: α-terpineno, *Chenopodium ambrosioides* L., Mastruz

ABSTRACT

Chenopodium ambrosioides L. is an aromatic herb widely distributed throughout the Americas, Africa and also in European countries. The essential oil from the leaves of *C. ambrosioides* is the main component used in folk medicine as an anthelmintic and for the treatment of influenza. In order to know the chemical composition, the essential oil from the leaves of the species cultivated in Recife-PE, Brazil, was extracted by hydrodistillation and analyzed by gas chromatography-mass spectrometry. The average yield of oil was 0.7% ± 0.1. The major components were α-terpinene (42.14%), α-terpinenyl-acetate (31.57%) and thymol (7.90%).

Keywords: α-terpinene, *Chenopodium ambrosioides* L., wormseed

INTRODUCTION

The genus *Chenopodium*, of the family Chenopodiaceae, includes several species native to Asia, Europe and America, used as food, condiment and/or medicinal plants (Kokanova-Nedialkova, Nedialkov & Nikolov, 2010, p. 280; Blanckaert *et al*., 2012, p. 559). *Chenopodium ambrosioides* L. is an herb popularly known as epazote, Mexican tea, American wormseed, paico, mastruz and erva-de-Santa-Maria (Kliks, 1985, p. 879; Albuquerque *et al*., 2009, p. 136). The species is native to Central and South America, it originated, probably, from Mexico. It has spontaneous growth in tropical, subtropical and temperate areas and is found on all continents (Kismann, 1991, 608p.). In Brazil, its distribution is extensive, occurring in almost all the territory (Sousa *et al*., 2004, 448p.; Lima *et al*., 2006, 82p.). Senna (2012) states that *C. ambrosioides* is native to Brazil, but not endemic. It has been used by local people as anthelmintic and in the treatments of influenza (Morais *et al*., 2005, p. 171; Lima *et al*., 2006, 82p.).

The essential oil (EO) of *C. ambrosioides* is also used as anthelmintic, especially against ascariasis (Guimaraes, Llanos & Acevedo, 2001, p. 212). Additionally, the EO presents several properties, such as antioxidant, antifungal, insecticidal and allelopathic (Hegazy & Farrag, 2007, p. 01; Kumar *et al*., 2007, p. 159; Jaramillo, Duarte & Delgado, 2012, p. 54).

In many Chenopodiaceae species, the secondary metabolites that compose the EO are synthesized in specialized secretory structures such as glandular trichomes (Metcalfe & Chalk, 1950, p. 1074). The biosynthesis of these metabolites can be influenced by external factors like environmental conditions, season and soil composition (Martins *et al*., 2006, p. 1203; Demuner *et al*., 2011, p. 1182). In many cases genetic variability can also result in plant of different chemotypes (Castro *et al*., 2004, p. 55; Silva *et al*., 2007, p. 474).

Previous studies have reported the composition of the EO of the plant in other countries, like Colombia (Jaramillo, Duarte & Delgado, 2012, p. 54) and Nigeria (Gbolade, Tira-Picos & Noguerria, 2010, p. 654). However, there are few studies on the species cultivated in Brazil.

The plant is one of the 71 plant species that arouse the interest of the government for the production of phytotherapics, and is present in the list of National Medicinal Plants of Interest to the Unified Health System (Brasil, 2009).

Considering the importance of the EO of *C. ambrosioides* for the development of new natural products and their wide popular and institutional interest, the aim of this study was to analyze the composition of the EO from leaves of *C. ambrosioides* by gas chromatography coupled to mass spectrometry, from plants grown in Recife-PE, Brazil.

MATERIALS AND METHODS

Plant Material

The plant was collected in the months of september, october and november of 2012, in the garden of the Laboratório de Fitoterapia, of the company Pernambuco Participações e Investimentos S/A (PERPART), located in the city of Recife, state of Pernambuco, Brazil. A voucher specimen was deposited in the herbarium UFP-Geraldo Mariz, of the Federal University of Pernambuco, Brazil, under number 69718.

Essential Oil Extraction

The EO was extracted from 30 g of fresh leaves by hydrodistillation for 2 h, in a Clevenger apparatus. The EO was collected, dried with anhydrous sodium sulfate, the volume determined and the yield calculated as the mean of three replications, on a fresh weight basis (Santos *et al*., 2004, p. 4). The EO was stored at 4°C until analyzed.

Gas Chromatography-Mass Spectrometry

Gas chromatography-mass spectrometry (GC-MS) analysis was performed using a GCMS-QP2010S (Shimadzu) instrument and a fused-silica capillary column RTX-5 (30 m x 0.25 mm i.d., film thickness, 1 μm; Restek). The operating conditions were: programmed column temperature 50° to 280°C at 4°C/min; injector port temperature, 250°C; carrier gas, 2.52 mL/min He; injection volume, 1 μL, splitless; interface temperature 260°C; electron impact ionization at 70 eV. Constituents were identified by matching their mass spectra with those recorded in the Wiley 229 library.

RESULTS AND DISCUSSION

A yellow-colored EO was obtained by hydrodistillation procedure yielding 0.7%. Ten compounds were identified (Table 1), most of them being monoterpenes and oxygenated monoterpenes. The major components found in the EO were α-terpinene (42.14%) and α-terpinenyl-acetate (31.57%), followed by thymol (7.90%).

Table 1. Chemical composition of the essential oil from leaves of *Chenopodium ambrosioides* L.

Compound	Retention time (min)	Area %
α-terpinene	18.704	42.14
ρ-cymene	19.138	7.29
γ-terpinene	21.220	0.75
α-terpinenyl-acetate	32.543	31.57
Ascaridole	33.490	0.87
Thymol	35.262	7.90
Carvacrol	35.851	4.27
Isoascaridole	36.396	2.61
Limonene oxide	49.494	0.44
Phytol	55.281	2.16

The composition of the EO of *C. ambrosioides* has been the subject of several studies and data from the literature shows that it has no constancy neither with respect to the compounds or their percentage. The factors that determine the composition and yield of essential oils are numerous, and in some cases, it is difficult to isolate these factors from each other. These variables may include seasonality and plant maturity, geographical origin, genetic variation, the stages of growth, the plant part used and the drying and post-harvest storage (Anwar *et al*, 2009, p. 188).

In the case of *C. ambrosioides*, realizes that the geographical origin of the plant has the greatest influence. The α-terpinene is the major component in the EO of plant collected in Nigeria, (Onocha *et al*., 1999, p. 220; Kasali *et al*., 2006, p. 13; Gbolade, Tira-Picos & Nogueria, 2010, p. 654), Rwanda (Muhayimana, Chalchat & Garry,1998, p. 690), Colombia (Jaramillo, Duarte & Delgado, 2012, p. 54), Cameroon (Chekem *et al*., 2010, 2900) and India (Gupta *et al*., 2002, p. 93; Singh *et al*., 2008, p. 378). In contrast, in India was also found *m*-cymene as major portion, corresponding to 43.9% of the oil (Prasad *et al*., 2009, p. 125). In Cameroon, Tapondjou *et al*. (2002, p. 398) found ρ-cymene corresponding to 50% the chemical constitution of EO.

Climatic conditions and water available in the soil can change the vegetal secondary metabolism and, consequently, alter the composition of essential oils, throughout the seasons of the year (Silva *et al*., 2012, p. 195). The results of this study were obtained with plant material collected during the spring in Pernambuco. Torres *et al*. (2003, p. 31) analyzed the EO composition of *C. ambrosioides* collected in Argentina in three seasons: summer, autumn and spring. In summer, the major component was α-phellandrene (40.0%), which was found in traces in autumn. In autumn were found α-pinene (32.7%) and limonene (32.5%) in higher concentrations. In the spring, these proportions fell to 17.4% for α-pinene and to 27.7% for limonene. The content of ascaridole showed little variation during the three seasons, being found in summer 8.6%, 9.2% in autumn and 9.5% in the spring.

The content of ascaridole, component for which are attributed some biological activities of the EO (Jardim *et al*., 2008, p. 1213; Chu, Hu & Liu, 2011, p. 714) is also affected by the place of origin of the plant. In Brazil, Jardim *et al*. (2008, p. 1213) reported a content of 80% of ascaridole in EO of plant collected in the Southeast region of the country. Ascaridole was also found as the main component of the EO of *C. ambrosioides* from Madagascar (Cavalli *et al*., 2004, p. 275), Togo (Koba *et al*., 2009, p. 435), the Eastern Mediterranean (Dembitsky, Shkrob & Hanus, p. 209) and China (Chu, Hu & Liu, 2011, p. 714). It was reported, however, accounting for only a small percentage of 0.1% in the EO of plant in Nigeria (Onocha *et al*., 1999, p. 220).

In the present study, with the cultivated species in the Northeast region of Brazil, only 0.87% of ascaridole was found. The chemical composition of the EO was quite different from that reported by Jardim *et al*. (2008, p. 1213). However, it is known that ascaridole is a heat-sensitive compound which rearranges on treatment at 150°C to form isoascaridole. Moreover, it has been noted that this isomerisation can give rise to inaccurate quantification of ascaridole (Cavalli *et al*., 2004, p. 275).

Futhermore, Johnson & Croteau (1984, p. 254) showed that the biosynthesis of ascaridole from α-terpinene was catalyzed by a soluble iodide peroxidase, which was isolated from homogenates of fruit and leaves of *C. ambrosioides*. The enzymatic synthesis of ascaridole was confirmed by GC-MS of the product. According Gobbo-Netto & Lopes (2007, p. 374) secondary metabolites represent a chemical interface between plants and the environment. Thus, stimulus arising from the environment in which the plant is located may redirect the metabolic route, leading to the biosynthesis of different compounds, which may also explain the variations in the chemical composition of the EO of *C. ambrosioides*.

Because it is a plant of great medicinal interest, and since the biological activities are related to the presence of secondary metabolites, further studies are needed about the factors that affect the composition of the EO of *C. ambrosioides*. The results are important for obtaining plants with greater accumulation of the compounds of interest and also for the optimization of harvesting techniques, post-harvest and extraction, visualizing potential commercial application of the plant (Silva *et al*., 2012, 195).

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REFERENCES

Albuquerque UP, Araújo TAS, Ramos MA, Nascimento VT, Lucena RFP, Monteiro JM, Alencar NL, Araújo EE. How ethnobotany can aid biodiversity conservation: reflections on investigations in the semi-arid region of NE Brazil. *Biodivers. Conser.* 18(1): 127-150, 2009.

Anwar F, Hussain AI, Sherazi TH, Bhanger MI. Changes in composition, antioxidant and antimicrobial activities of essential oil of fennel (*Foeniculum vulgare* Mill.) fruits at different stages of maturity. *J. Herbs Spices Med. Plants.* 15(2): 187-202, 2009.

Blanckaert I, Paredes-Flores M, Espinosa-García FJ, Piñero D, Lira R. Ethnobotanical, morphological, phytochemical and molecular evidence for the incipient domestication of Epazote (*Chenopodium ambrosioides* L.: Chenopodiaceae) in a semi-arid region of Mexico. *Genet. Resouc. Crop. Evol.* 59(4): 557-573, 2012.

Brasil. Plantas de interesse ao SUS. Disponível em: <http://portal.saude.gov.br/portal/saude/profissional/visualizar_texto.cfm?idtxt=30277&janela=1>. Acesso em maio de 2012.

Castro HG, Oliveira LO, Barbosa LCA, Ferreira FA, Silva DJH, Mosquim PR, Nascimento EA. Teor e composição do óleo essencial de cinco acessos de mentrasto. *Quim. Nova.* 27(1): 55-57, 2004.

Cavalli J, Tomi F, Bernardini A, Casanova J. Combined analysis of the essential oil of *Chenopodium ambrosioides* by GC, GC-MS and ^{13}C-NMR spectroscopy: quantitative determination of ascaridole, a heat-sensitive compound. *Phytochem. Anal.* 15(5): 275-279, 2004.

Chekem MSG, Lunga PK, Tamokou JDD, Kuiate JR, Tane P, Vilarem G, Cerny M. Antifungal properties of *Chenopodium ambrosioides* essential oil against *Candida* Species. *Pharmaceuticals*. 3(9): 2900-2909, 2010.

Chu SS, Hu JF, Liu ZL. Composition of essential oil of chinese *Chenopodium ambrosioides* and insecticidal activity against maize weevil, *Sitophilus zeamais*. *Pest. Manag. Sci.* 67(6): 714-718, 2011.

Dembitsky V, Shkrob I, Hanus LO. Ascaridole and related peroxides from the genus *Chenopodium*. *Biomed. Pap. Med. Fac. Univ. Palacky Olomouc Czech Repub.* 152(2): 209-215, 2008.

Demuner AJ, Barbosa LCA, Magalhaes CG, Silva CJ, Maltha CRA, Pinheiro AL. Seasonal variation in the chemical composition and antimicrobial activity of volatile oils of three species of *Leptospermum* (Myrtaceae) grown in Brazil. *Molecules.* 16(2): 1181-1191, 2011.

Gbolade AA, Tira-Picos V, Nogueria JMF. Chemical constituents of *Chenopodium ambrosioides* L. var *anthelminticum* herb essential oil from Nigeria. *Chem. Nat. Comp.* 46(4): 654-655, 2010.

Gobbo-Neto L & Lopes NP. Plantas medicinais: fatores de influência no conteúdo de metabólitos secundários. *Quím. Nova*. 30(2): 374-381, 2007.

Guimaraes DL, Llanos RSN, Acevedo JHR. Ascaridiasis: comparison of the therapeutic efficacy between paico and albendazole in children from Huaraz. *Rev. Gastroenterol. Perú.* 21(3): 212-219, 2001.

Gupta D, Charles R, Mehta VK, Garg SN, Kumar S. Chemical composition of the essential oil of *Chenopodium ambrosioides* L. from the southern hills of India. *J. Essent. Oil Res.* 14(2): 93-94, 2002.

Hegazy AK & Farrag HK. Allelopathic potential of *Chenopodium ambrosioides* on germination and seedling growth of some cultivated and weed plants. *Global J. Biotech. Biochem.* 2(1): 01-09, 2007.

Jaramillo BE, Duarte E, Delgado W. Bioactividad del aceite esencial de *Chenopodium ambrosioides* colombiano. *Rev. Cubana Plant. Med*. 17(1): 54-64, 2012.

Jardim CM, Jham GN, Dhingra OD, Freire MM. Composition and antifungal activity of the essential oil of the brazilian *Chenopodium ambrosioides* L. *J. Chem. Ecol.* 34(9): 1213-1218, 2008.

Johnson MA & Croteau R. Biosynthesis of ascaridole: iodide peroxidase-catalyzed synthesis of a monoterpene endoperoxide in soluble extracts of *Chenopodium ambrosioides* fruit. *Arch. Biochem. Biophys.* 235(1): 254-266, 1984.

Kasali AA, Ekundayo O, Paul C, König WA, Eshilokun AO, Ige B. 1,2:3,4-diepoxy-p-menthane and 1,4-epoxy-p-menth-2-ene: rare monoterpenoids from the essential oil of *Chenopodium ambrosioides* L. var *ambrosioides* leaves. *J. Essent. Oil Res.* 18(1): 13-15, 2006.

Kismann KG. Plantas infestantes e nocivas. São Paulo: BASF Brasileira, 1991.

Kliks MM. Studies on the traditional herbal anthelmintic *Chenopodium ambrosioides* L.: ethnopharmacological evaluation and clinical field trials. *Soc. Sci. Med.* 21(8): 879-886, 1985.

Koba K, Catherine G, Raynaud C, Chaumont J, Sanda K, Laurence N. Chemical composition and cytotoxic activity of *Chenopodium ambrosioides* L. essential oil from Togo. *Bangladesh J. Sci. Ind. Res.* 44(4): 435-440, 2009.

Kokanova-Nedialkova K, Nedialkov PT, Nikolov SD. The genus *Chenopodium*: phytochemistry, ethnopharmacology and pharmacology. *Phcog. Rev*. 3(6): 280-306, 2010.

Kumar R, Mishra AK, Dubey NK, Tripathi YB. Evaluation of *Chenopodium ambrosioides* oil as a potential source of antifungal, antiaflatoxigenic and antioxidant activity. *Int. J. Food Microbiol.* 115(2): 159-164, 2007.

Lima JLS et al. Plantas medicinais de uso comum no Nordeste do Brasil. Campina Grande: CEDAC, 2006.

Martins FT, Santos MH, Polo M, Barbosa LCA. Variação química do óleo essencial de *Hyptis suaveolens*(L.) Poit. sob condições de cultivo. *Quim. Nova.* 29(6): 1203-1209, 2006.

Metcalfe CR & Chalk L. Anatomy of the dicotyledons. Oxford: Clarendon Press, 1950.

Morais SM, Dantas JDP, Silva ARA, Magalhães EF. Plantas medicinais usadas pelos índios Tapebas do Ceará. *Rev. Bras. Farmacogn.* 15(2): 169-177, 2005.

Muhayimana A, Chalchat JC, Garry RP. Chemical composition of essential oils of *Chenopodium ambrosioides* L. from Rwanda. *J. Essent. Oil Res.* 10(6): 690-692, 1998.

Onocha PA, Ekundayo O, Eramo T, Laakso I. Essential oil constituents of *Chenopodium ambrosioides* L. leaves from Nigeria. *J. Essent. Oil Res.* 11(2): 220-222, 1999.

Prasad CS, Shukla R, Kumar A, Dubey NK. In vitro and in vivo antifungal activity of essential oils of *Cymbopogon martini* and *Chenopodium ambrosioides* and their synergism against dermatophytes. *Mycoses.* 53(2): 123-129, 2009.

Santos AS, Alves SM, Figueiredo FJC, Rocha Neto OG. Descrição de Sistema e de Métodos de Extração de Óleos Essenciais e Determinação de Umidade de Biomassa em Laboratório. *Comunicado Técnico-Embrapa*. 99: 1-6, 2004.

Senna L. Chenopodium in Lista de Espécies da Flora do Brasil. Jardim Botânico do Rio de Janeiro. Disponível em: <http://floradobrasil.jbrj.gov.br/jabot/floradobrasil/FB4313>. Acesso em abril de 2014.

Silva CJ, Barbosa LCA, Maltha CRA, Pinheiro AL, Ismail FMD. Comparative study of the essential oils of seven *Melaleuca* (Myrtaceae) species grown in Brazil. *Flavour Fragr. J.* 22(6): 474-478, 2007.

Silva CJ, Barbosa LCA, Demuner AJ, Montanari RM, Francino D, Meira RMSA, Souza AO. Chemical composition and histochemistry of *Sphagneticola trilobata* essential oil. *Rev. Bras. Farmacogn.* 22(3): 482-489, 2012.

Singh HP, Batish DR, Kohli RK, Mittal S, Yadav S. Chemical composition of essential oil from leaves of *Chenopodium ambrosioides* from Chandigarh, India. *Chem. Nat. Comp.* 44(3): 378-379, 2008.

Sousa MP et al. Constituintes químicos ativos e propriedades biológicas de plantas medicinais brasileiras. Fortaleza: Editora UFC, 2004.

Tapondjou LA, Adler C, Bouda H, Fontem DA. Efficacy of powder and essential oil from *Chenopodium ambrosioides* leaves as post-harvest grain protectants against six-stored product beetles. *J. Stored Prod. Res.* 38(4): 395-402, 2002.

Torres MC, Ricciardi GAL, Nassiff AEA, Ricciardi AIA, Bandoni AL. Larvicidal and nematicidal activities of the leaf essential oil of *Croton regelianus*. *Facena*. 5(12): 2724-2728, 2003.