SHORT COMMUNICATION

Chemical composition of the essential oil from the leaves of Anaxagorea brevipes (Annonaceae) and evaluation of its bioactivity

Danielle Cardoso de Alencara, Maria Lúcia Belém Pinheiro, José Lamak da Silva Pereira, João Ernesto de Carvalho, Francinete Ramos Campos, Alessandra Freitas Serain, Ricardo Brunelli Tirico, Alvaro José Hernández-Tasco, Emmanoel Vilaça Costa and Marcos José Salvador

ABSTRACT

The essential oil obtained by hydrodistillation from leaves of Anaxagorea brevipes was analysed by gas chromatography fitted with a flame ionisation detector (GC–FID) and coupled to mass spectrometry (GC–MS). Thirty one components were identified, representing around 75.7% of total oil. The major components were β-eudesmol (13.16%), α-eudesmol (13.05%), γ-eudesmol (7.54%), guaiol (5.12%), caryophyllene oxide (4.18%) and β-bisabolene (4.10%). The essential oil showed antimicrobial activity against Gram-positive bacteria and yeast with the MIC values between 25.0 and 100 μg/mL. The highest antiproliferative activity was observed for the oil against MCF-7 (breast, TGI = 12.8 μg/mL), NCI-H460 (lung, TGI = 13.0 μg/mL) and PC-3 (prostate, TGI = 9.6 μg/mL) cell lines, while against no cancer cell line HaCat (keratinocyte) the TGI was 38.8 μg/mL. The oil exhibited a small antioxidant activity assessed through ORAC-FL assay (517 μmol TE/g). This is the first report regarding the chemical composition and bioactivity of A. brevipes essential oil.

ARTICLE HISTORY

Received 25 February 2015
Accepted 27 August 2015

KEYWORDS

Anaxagorea brevipes; Annonaceae; essential oil; bioactivity

CONTACT
Marcos José Salvador marcosjs@unicamp.br, mjsalvador@bol.com.br

© 2015 Taylor & Francis
1. Introduction

Annonaceae family is widely distributed around the world and is the largest family in Magnoliales with approximately 130 genera and 2500 species. Represented by trees, shrubs, and woody climbers, they are valuable for their large pulpy fruits and others are used in traditional medicine (Richardson et al. 2004). Essential oil of several Annonaceae plants has been demonstrated promissory biological effects such as: insecticidal (Babarinde et al. 2015); spasmolytic (Correia et al. 2015); antitumour and trypanocidal activities (Costa et al. 2013). The genus Anaxagorea A. St. Hil includes 26 species with pantropical distribution across the USA and Asia and in Brazil with occurrence in Amazon forest (Lobão et al. 2005). Of the total species of the Anaxagorea, only five were phytochemically investigated, from which lignoids (Puentes De Díaz 1997), flavonoids (Gonda et al. 2002), polyprenoides (Sasak & Chojnacki 1973), xanthisones (Gonda et al. 2002), steroids (Puentes De Díaz 1997) and alkaloids were isolated (Hocquemiller et al. 1981). The essential oil of Anaxagorea dolicchocarpa showed terpenoid β-elemene in the leaves and the δ-cadinol, caryophyllene oxide, δ-cadineno, α-copaene and the γ-muurolene in the fruits as the main components (Fournier et al. 1994). Investigation of fragrances in the flowers of Anaxagorea brevipes and A. dolicchocarpa showed esters of aliphatic acids, including ethyl 2-methylbutanoate in A. brevipes and ethyl 3-methylbutanoate in A. dolicchocarpa as the major components (Jürgens et al. 2000).

The A. brevipes is commonly known as the ‘envira preta’ in Colombia, Venezuela, Suriname and in much of the Amazon. To our knowledge, this is the first report on the analysis of the chemical composition and evaluation of antimicrobial, antiproliferative and antioxidant activities of their essential oil and for the bioactivity of this plant.

2. Results and discussion

2.1. Essential oils analysis

The hydrodistillation of A. brevipes leaves produced a yellow crude essential oil, with a yield of 0.52%, in relation to the dry weight of the plant material. In total, 31 compounds were identified in the essential oil. The sesquiterpenes were the most abundant constituents with 72.34% of the crude essential oil (Table S1). The major compounds identified in the essential oil of A. brevipes were β-eudesmol (13.16%), α-eudesmol (13.05%), γ-eudesmol (7.54%), guaiol (5.12%), caryophyllene oxide (4.18%) and β-bisabolene (4.10%), and have been described for the first time in this plant. However, some compounds identified in the essential oil of A. brevipes have been described in other species of Anaxagorea (Fournier et al. 1994; Jürgens et al. 2000), indicating that this species is a typical member of the Anaxagorea genus. In addition, some of these compounds are also reported as constituents in the essential oil of other genera of Annonaceae family (Costa et al. 2008; Britto et al. 2012; Thang et al. 2013, 2014).

The essential oil of A. dolicchocarpa showed as main component the terpenoid β-elemene in the leaves and the δ-cadinol, caryophyllene oxide, δ-cadineno, α-copaene and the γ-muurolene in the fruits (Fournier et al. 1994). The chemical presence of the eudesmols (α, β and γ) as constituents in the essential oil of A. brevipes is interesting from the point of view about the chemistry of the essential oil of Annonaceae species. These compounds α-, β- and γ-eudesmols together are the major compounds as observed only in Guatteria...
friesiana (W.A. Rodrigues) Erkens & Maas, indicating some chemotaxonomic relationship (Costa et al. 2008; Britto et al. 2012). Further investigations are necessary to confirm this hypothesis.

### 2.2. Biological activity

The essential oil of *A. brevipes* showed antibacterial and antifungal inhibitory effect, early against *Kocuria rhizophila* (ATCC 9341), *Staphylococcus aureus* penicillinase-negative (strain, 8-), *Candida albicans* (ATCC 10231) and *Candida parapsilosis* (ATCC 22019) with MIC values between 25.0 and 100 μg/mL (Table S2). This result is very similar to the recent studies performed with Annonaceae plants. These works have demonstrated that essential oils with high sesquiterpene contents, mainly germacrene D, caryophyllene and β-eudesmol, possess antimicrobial properties (Costa et al. 2008; Ho et al. 2010).

The antiproliferative activity of the essential oil from the leaves of *A. brevipes* was investigated on a series of cancer cell lines and highest activity was observed (Table S3). The bioactivity was particularly found against MCF-7 (breast, TGI (total growth inhibition) = 12.8 μg/mL), NCI-H460 (lung, TGI = 13.0 μg/mL) and PC-3 (prostate, TGI = 9.6 μg/mL) cell lines, while against no cancer cell line Hacat (keratinocyte, TGI = 38.8 μg/mL). Antitumor properties of the essential oil of Annonaceae species were described *in vitro* and *in vivo* and germacrene B, germacrene D and caryophyllene were found as the main compounds (Ferraz et al. 2013; Quintans et al. 2013). Also, *in vitro* and *in vivo* antitumour effects were documented for the essential oil from the leaves of *G. friesiana* and these effects could be assigned to its components α-, β- and γ-eudesmol. Therefore, the antiproliferative activity found here is also attributed to the sesquiterpenes constitution of the essential oil of *A. brevipes*, mainly the presence of α-, β- and γ-eudesmol (Britto et al. 2012). The same result was observed for the antimicrobial activity of the oil of *A. brevipes* (Costa et al. 2008).

Regarding antioxidant activity, the oil exhibited a small antioxidant activity assessed through TLC autographic assay for DPPH Radical-Scavenging (qualitative method) and by ORAC<sub>FL</sub> assay (quantitative method). In ORAC<sub>FL</sub> assay, the antioxidant capacity of the essential oil was 517.04 (15.20) μmol TE/g and in this assay is considered to have good antioxidant capacity with values >800 μmol TE/g (Costa et al. 2011). Similar results were verified for essential oil of other Annonaceae species (Costa et al. 2011; Siqueira et al. 2015).

### 3. Conclusion

In this study the chemical composition of the essential oil from the leaves of *A. brevipes*, and 31 components were identified, representing around 75.7% of total oil. The major components were β-bisabolene, caryophyllene oxide, guaiol and α-, β- and γ-eudesmol. The chemical composition of this oil is in agreement with the chemistry of the Annonaceae family. The essential oil showed antimicrobial and antiproliferative activity. This is the first report regarding the chemical composition and bioactivity of *A. brevipes* essential oil and the results confirm that the Annonaceae species are natural source of biologically active compounds. Further investigations (*in vitro* and *in vivo*) are necessary to confirm the potential of the *A. brevipes* essential oil as active and to study their toxicity and efficacy toward a clinical employment.
Supplementary material

Experimental details relating to this article are available online, alongside with Tables S1-S3.

Acknowledgements

The authors are grateful to FAPEAM, FAPITEC/SE, FAPESB, FAPESP, CAPES and CNPq for financial support and Professor Dr Antonio Carlos Webber of the DB/UFAM for the botanical identification.

Disclosure statement

No potential conflict of interest was reported by the authors.

References

Babarinde SA, Pitan OOR, Olatunde GO, Ajala MO. 2015. First report of toxicity of Xylopia parviflora (A. Rich.) Benth (Annonaceae) root bark’s essential oil against cowpea seed bruchid, Callosobruchus maculatus Fabricius (Coleoptera: Chrysomelidae: Bruchinae). Nat Prod Res. 29:349–352. doi: 10.1080/14786419.2014.940943

Britto ACS, De Oliveira ACA, Henriquez RM, Cardoso GMB, Bomfim DS, Carvalho AA, Moraes M, Pessoa C, Pinheiro M, Costa E, et al. 2012. In vitro and in vivo antitumor effects of the essential oil from the leaves of Guatteria friesiana. Planta Med. 78:409–414. doi:10.1055/s-0031-1298173

Correia AC, Ferreira TF, Martins IRR, Macêdo CL, de S. Monteiro FS, Costa VCO, Tavares JF, Silva MS, Paredes-Gamero EJ, Buri MV, et al. 2015. Essential oil from the leaves of Xylopia langsdorffiana (Annonaceae) as a possible spasmolytic agent. Nat Prod Res. 29:980–984. doi:10.1080/14786419.2014.964706

Costa EC, Dutra LM, Salvador MJ, Ribeiro LHG, Gadelha FR, de Carvalho JE. 2013. Chemical composition of the essential oils of Annona pickelli and Annona salzmanni (Annonaceae), and their antitumour and trypanocidal activities. Nat Prod Res. 27:997–1001. doi:10.1080/14786419.2012.68691sd3

Costa EV, Dutra LM, Jesus HCR, Nogueira PCL, Moraes VRS, Salvador MJ, Cavalcanti SCH, Santos RLC, Prata, APN. 2011. Chemical composition and antioxidant, antimicrobial, and larvicidal activities of the essential oils of Annona salzmanni and A. pickelli (Annonaceae). Nat Prod Commun. 6:907–912.

Costa EV, Teixeira SD, Marques FA, Duarte MCT, Delarmelina C, Pinheiro MLB, Trigo JR, Sales Maia BHLNS. 2008. Chemical composition and antimicrobial activity of the essential oils of the Amazon Guatteria species. Phytochemistry 69:1895–1899. doi:10.1016/j.phytochem.2008.03.005

Ferraz RPC, Cardoso GMB, Da Silva TB, Fontes JEN, Prata APN, Carvalho AA, Moraes MO, Pessoa C, Costa EV, Bezerra DP. 2013. Antitumour properties of the leaf essential oil of Xylopia frutescens Aubl. (Annonaceae). Food Chem. 141:196–200. doi:10.1016/j.foodchem.2013.02.114

Fournier G, Hadjiakhoondi A, Charles B, Leboeuf M, Cavé A. 1994. Volatile components of Anaxagorea dolichocarpa fruit. Biochem Syst Ecol. 22:605–608. doi:10.1016/0305-1978(94)90073-6

Gonda R, Takeda T, Akiyama T. 2002. Studies on the constituents of Anaxagorea luzonensis A. Gray Nat Med. 56:10–12.

Ho C, Wang EI, Tseng Y, Liao P, Lin C, Chou J, Su Y. 2010. Composition and antimicrobial activity of the leaf and twig oils of Litseaum haenaeis and L. linii from Taiwan. Nat Prod Commun. 5:1823–1828.

Hocqueimiller R, Rasamizafy S, Moretti C, Jacquemin H, Cavé A. 1981. Anaxagoreine, a new aporphine alkaloid isolated from two species of the genus Anaxagorea. Planta Med. 41:48–50.

Jürgens A, Webber AC, Gottsberger G. 2000. Floral scent compounds of Amazonian Annonaceae species pollinated by small beetles and thrips. Phytochemistry 55:551–558. doi: 10.1016/S0031-9422(00)00241-7

Lobão A, Sue Dunn D, Kurtz B. 2005. Annonaceae das restings do estado do Rio de Janeiro, Brasil Rodriguesia 56:85–96.

Puentes De Díaz AM. 1997. Neolignans from Anaxagorea clavata. Phytochemistry 44:345–346.

Quintans JSS, Soares BM, Ferraz RPC, Oliveira ACA, Da Silva TB, Menezes LRA, Sampaio MFC, Prata APN,
Moraes MO, Pessoa C, et al. 2013. Chemical constituents and anticancer effects of the essential oil from leaves of *Xylopia laevigata*. Planta Med. 79:123–130. doi:10.1055/s-0032-1328091
Richardson JE, Chatrou LW, Mols JB, Erkens RH, Pirie MD. 2004. Historical biogeography of two cosmopolitan families of flowering plants: annonaceae and rhamnaceae. Philos Trans R Soc London, Ser B: Biol Sci. 359:1495–1508. doi:10.1098/rstb.2004.1537
Sasak W, Chojnacki L. 1973. Long chain polyprenols of tropical and subtropical plants. Acta Biochim Pol. 20:343–350.
Siqueira CAT, Serain AF, Pascoa ACRF, Andreazza NL, De Lourenço CC, Góis-Ruiz ALT, De Carvalho JE, De Souza ACO, Tonini JM, Tempone AG, Salvador MJ. 2015. Bioactivity and chemical composition of the essential oil from the leaves of *Guatteria australis* A.St.-Hil. Nat Prod Res. 29:1–4.
Thang TD, Dai DN, Hoi TM, Ogunwande IA. 2013. Study on the volatile oil contents of *Annona glabra* L., *Annona squamosa* L., *Annona muricata* L. and *Annona reticulata* L., from Vietnam. Nat Prod Res. 27:1232–1236. doi:10.1080/14786419.2012.724413
Thang TD, Luu HV, Dung VC, Tuan NN, Hung NH, Dai DN, Ogunwande IA. 2014. Chemical constituents of essential oils from the leaves and stem barks of four Vietnamese species of *Fissistigma* (Annonaceae). Nat Prod Res. 28:174–184. doi:10.1080/14786419.2013.863199