

# Time-scale and intraspecific variability of *Lychnophora ericoides* Mart. leave's volatile constituents.

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# Background



## Biodiversity



## Bioprospection

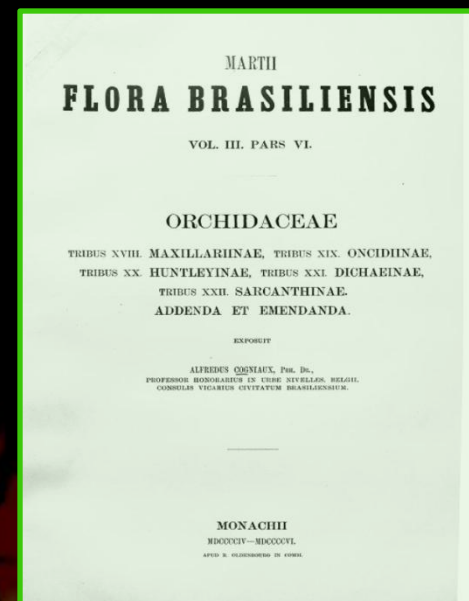


## Chemodiversity (1)

23.000 species: 1840 – 1906 <sup>(2)</sup>

➤ von Martius, Eichler, Urban.

JBRJ (2010) <sup>(3)</sup>: 40.982 entries for species.



2010 lista de espécies  
flora do brasil

1- GERSHENZON; DUDAREVA. Nature Chemical Biology. 2007.

2- <<http://florabrasiliensis.cria.org.br/opus/>>

3- <<http://floradobrasil.jbrj.gov.br/2010/>>.

# *L. ericoides*



Asteraceae

Vernonieae

Lychnophorinae

*Lychnophora ericoides*

10- SEMIR et al. 2011.

5- SEMIR. PhD dissertation. UNICAMP, 1991.

6- MANSANARES. PhD dissertation. UNICAMP, 2004.

# *L. ericoides*

Small trees/shrubs – candelabra pattern<sup>(5)</sup>

Rupestrian fields<sup>(5)</sup>

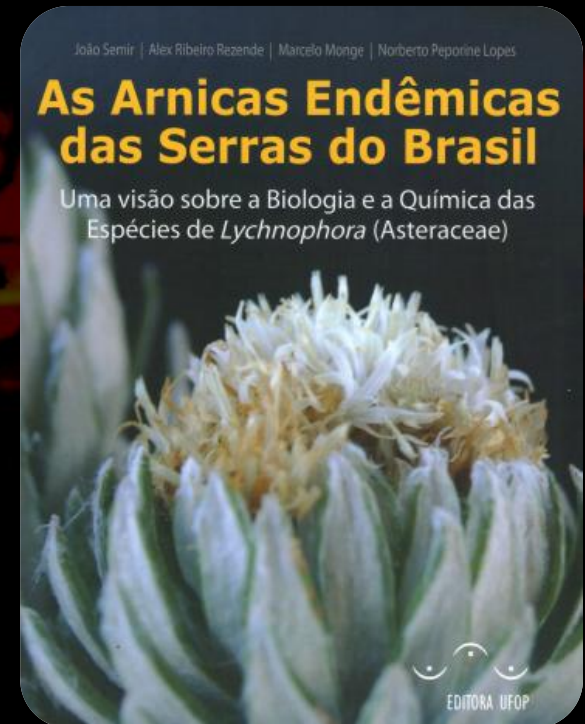
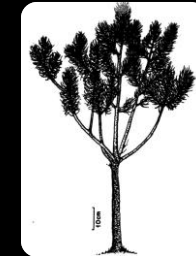
Essential oil: - Acaricidal<sup>(7)</sup>

- Antihypernociceptive<sup>(X)</sup>

Ethnopharmacology

↳ Seasonality on antiinflammatory<sup>(8)</sup>

Polymorfism<sup>(6)</sup>



10- SEMIR et al. 2011.

6- MANSANARES. PhD dissertation. UNICAMP, 2004.

7-BALDIN et al. Boletín de Sanidad Vegetal. Plagas. 2010.

8-LOPES. FAPESP – Pesquisa. 2000.

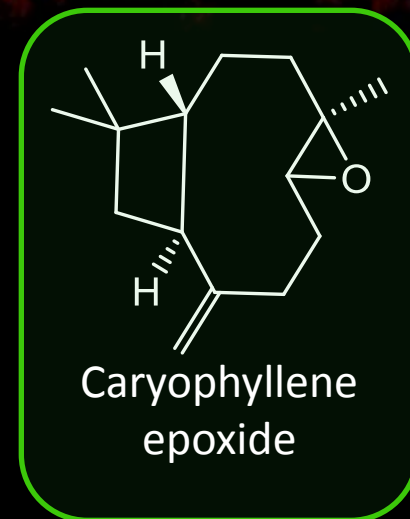
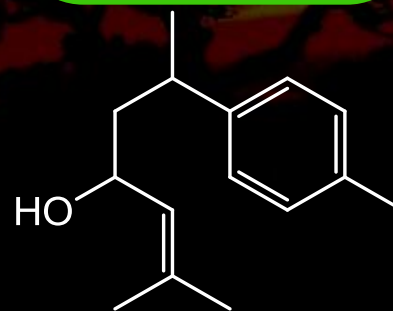
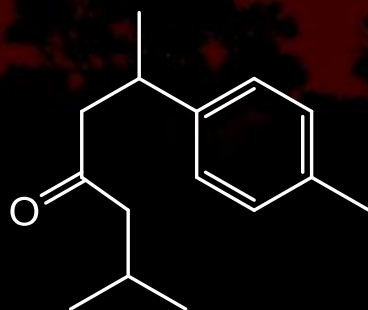
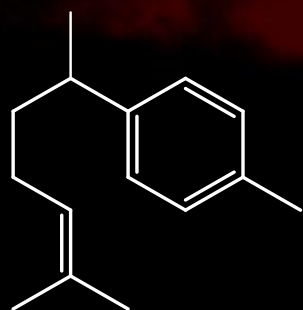
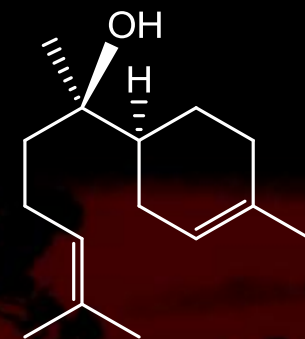
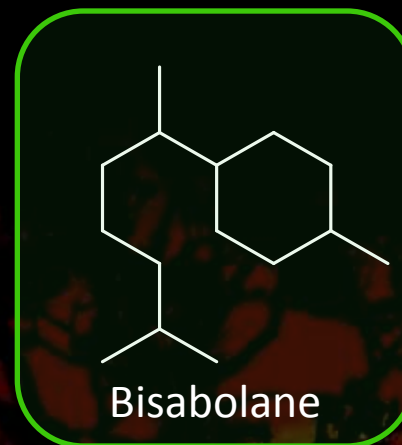
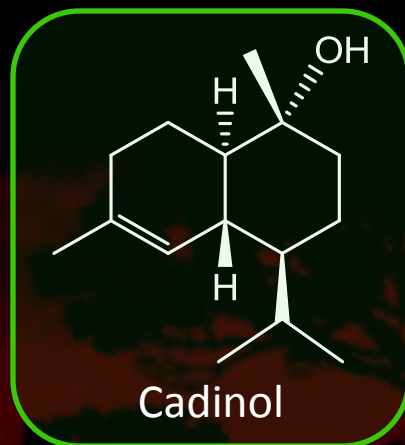
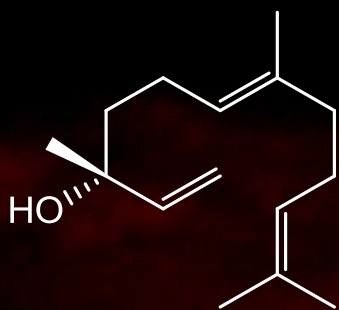
X- PAVARINI et al. UNPUBLISHED



# *L. ericoides*

Lychnophorinae<sup>(9)</sup>

→ Terpenoids, STLs and flavonoids (polar fractions)



# Analytical Methodology

- **SPME: soft pre-concentration of volatiles**
- **GC-MS: chemicals separation and identification**
- ***prep*TLC: isolation of unknown/novel chemicals**
- **Clevenger: hydrodestilation → essential oil**
- **ESI-MS/MS: “*soft ionization*”**
- **NMR: further structural information**



# Aims



**1. To identify the major compounds in a softly concentrated volatile fraction of *Lychnophora ericoides* leaves.**



**2. To understand the dynamics behind its terpenes level along the time and within geographically distinct individuals.**



# Experimental

# Harvest

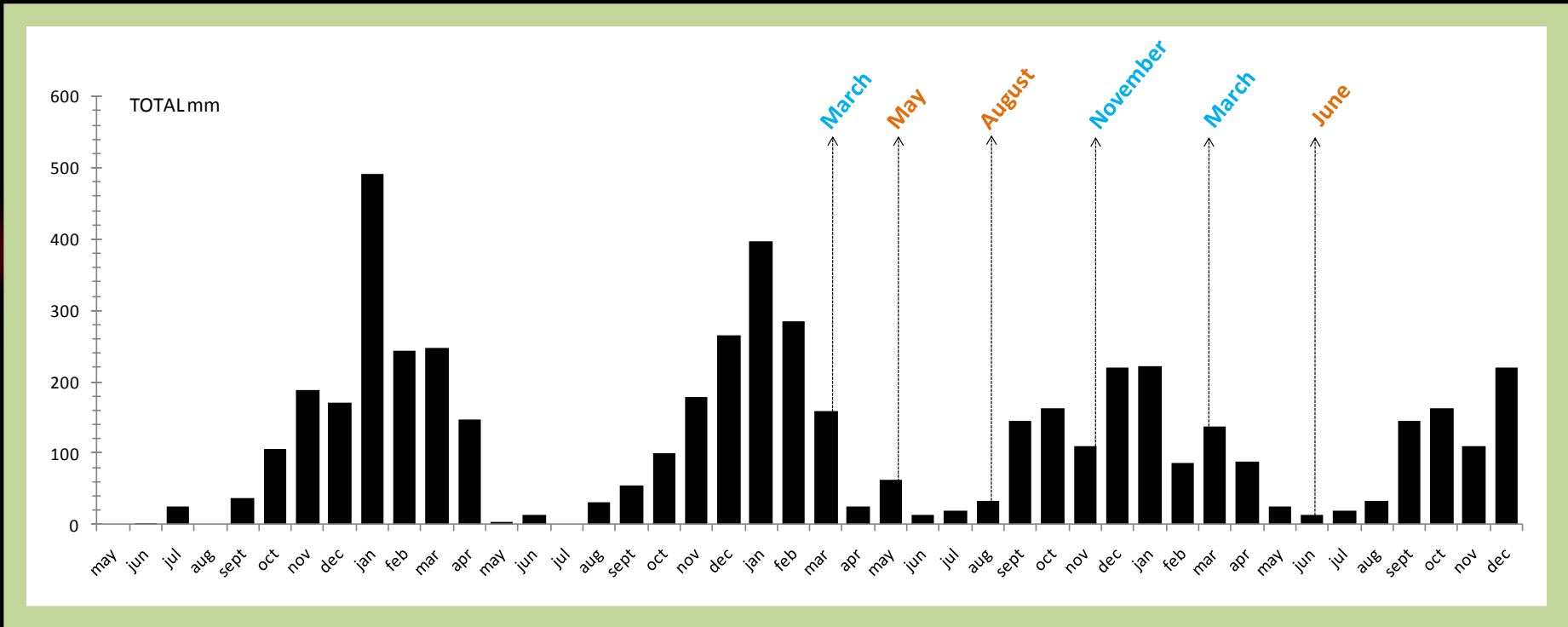
Allowance nº 010143/2011-4  
Allowance nº 010145/2011-7



S 20 38' 31.6", W 046 15' 31.8";  
1010 m high ("pedreira").

S 20 37' 54.0", W 046 19' 39.1";  
900 m high ("paraíso").

# Harvest

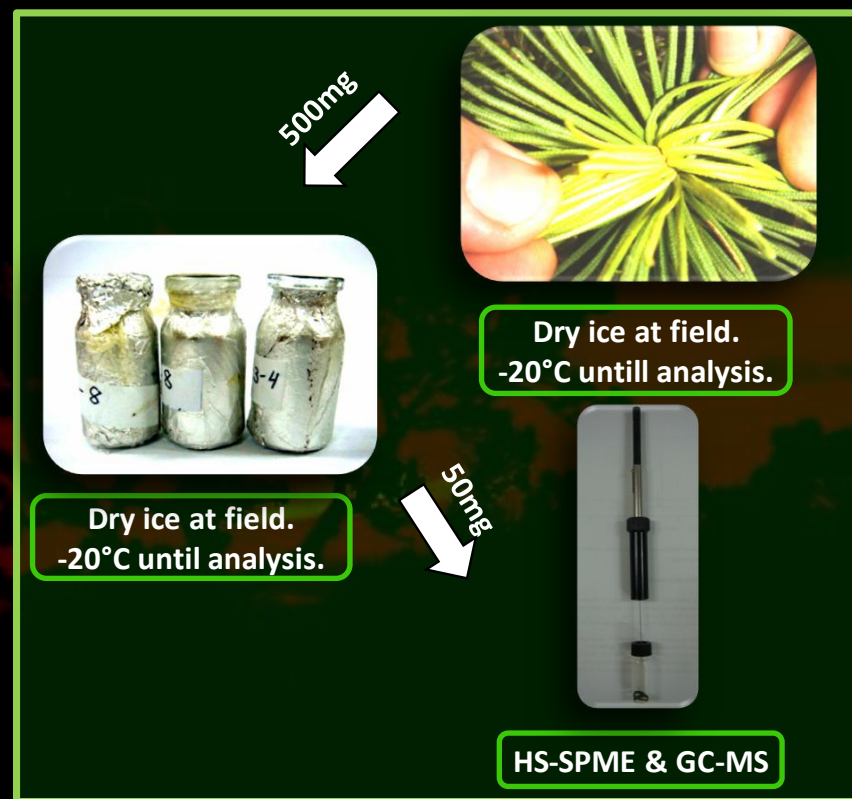


Total precipitation (mm /m<sup>2</sup>) per month

# Intraplant investigation



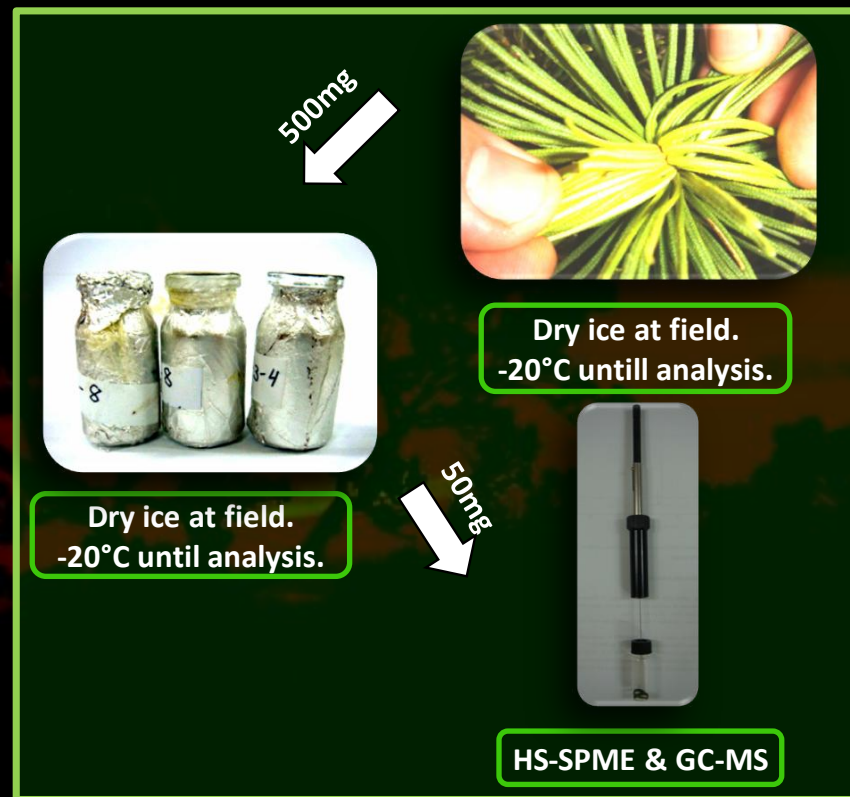
11h30min





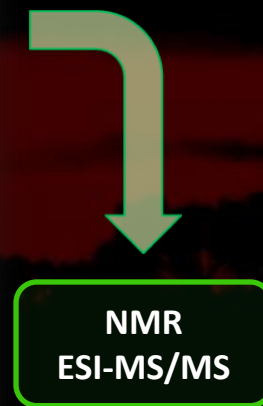
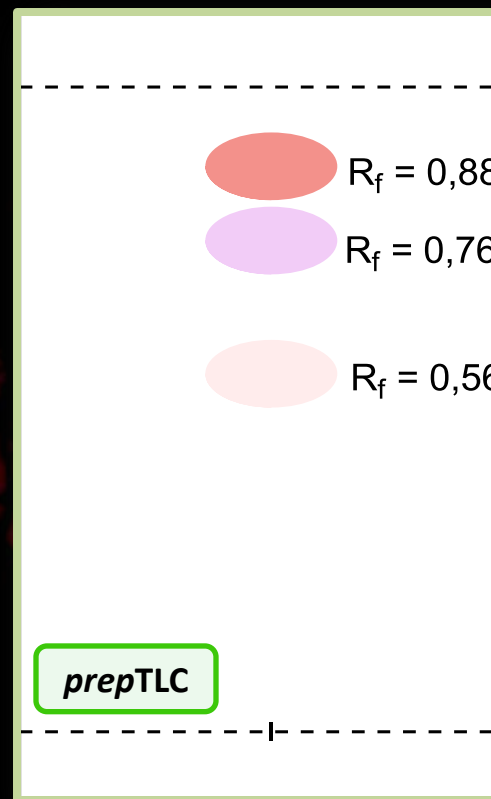
# Circadian investigation

Harvest	Day time
1	15h30min
2	19h30min
3	23h30min
4	03h30min
5	07h30min
6	11h30min





# Chemical profiling



GC-MS (Column DB5-MS)

Mass spectra libraries (>95%) (a)

Relative Retention indexes (>99%) (b)

a. Willey 7, NIST 62 e FFNSC Ver.1.3.

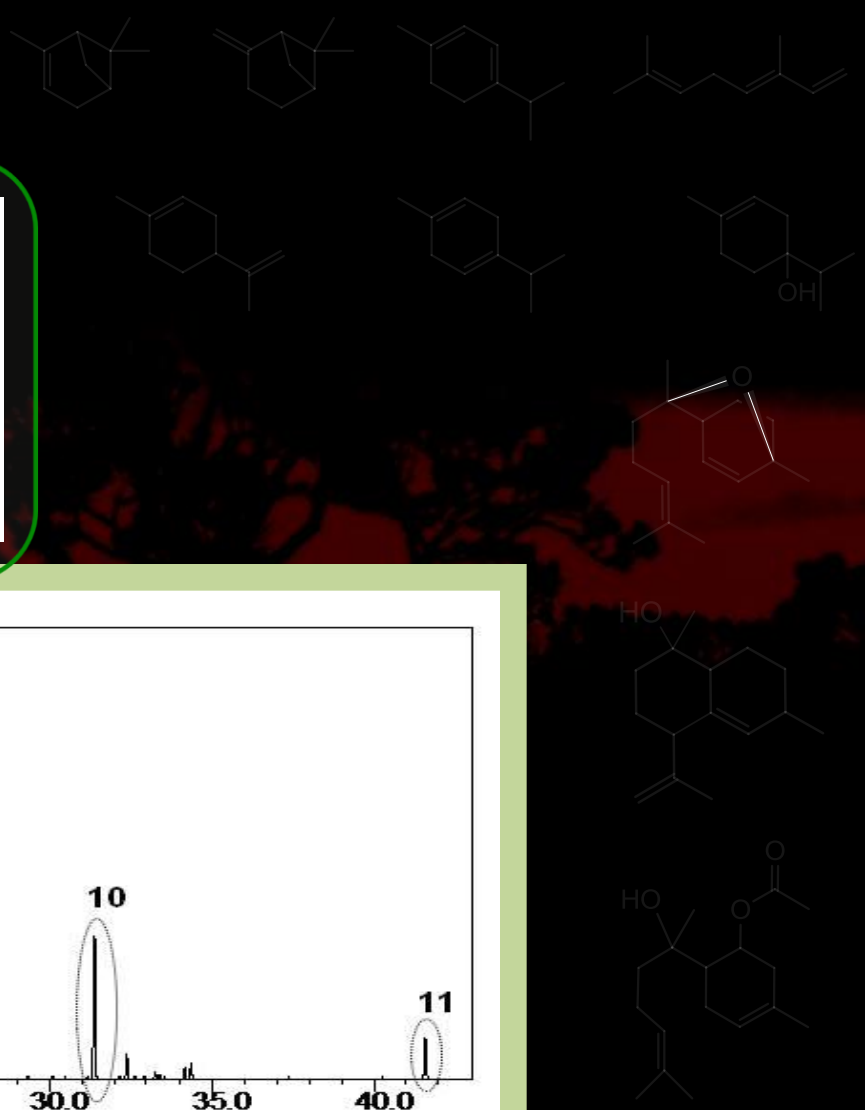
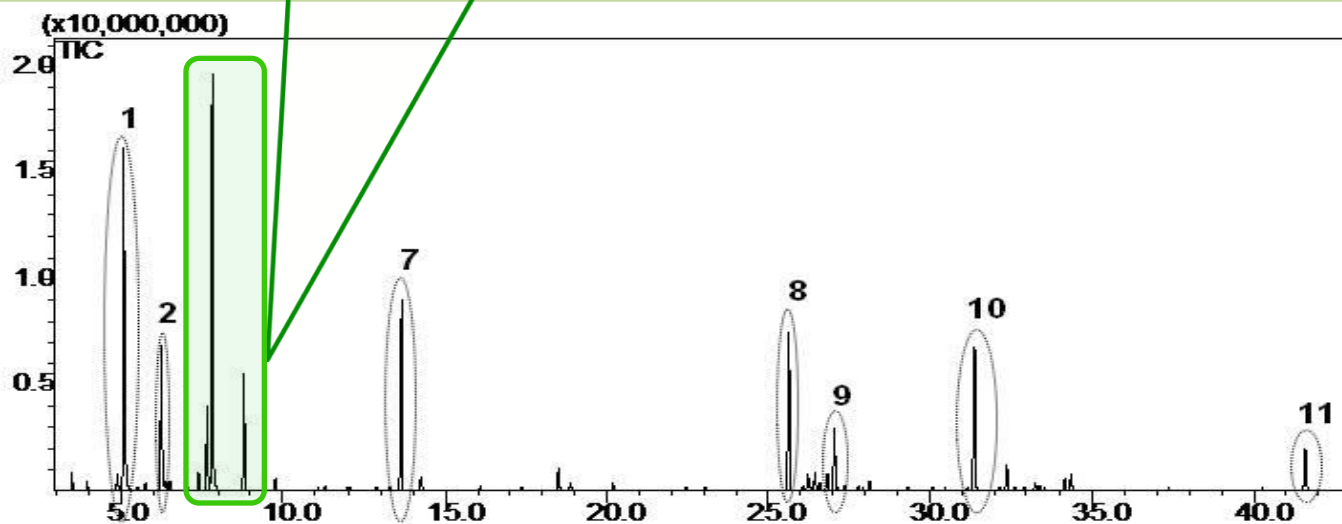
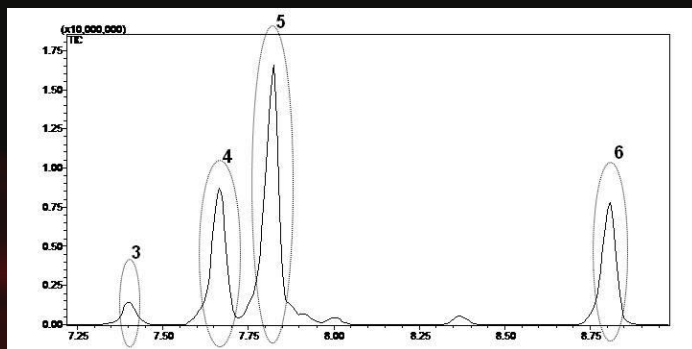
b. ADAMS, R. P. Identification Oil Components by Gas Chromatography/Mass Spectroscopy,1995.



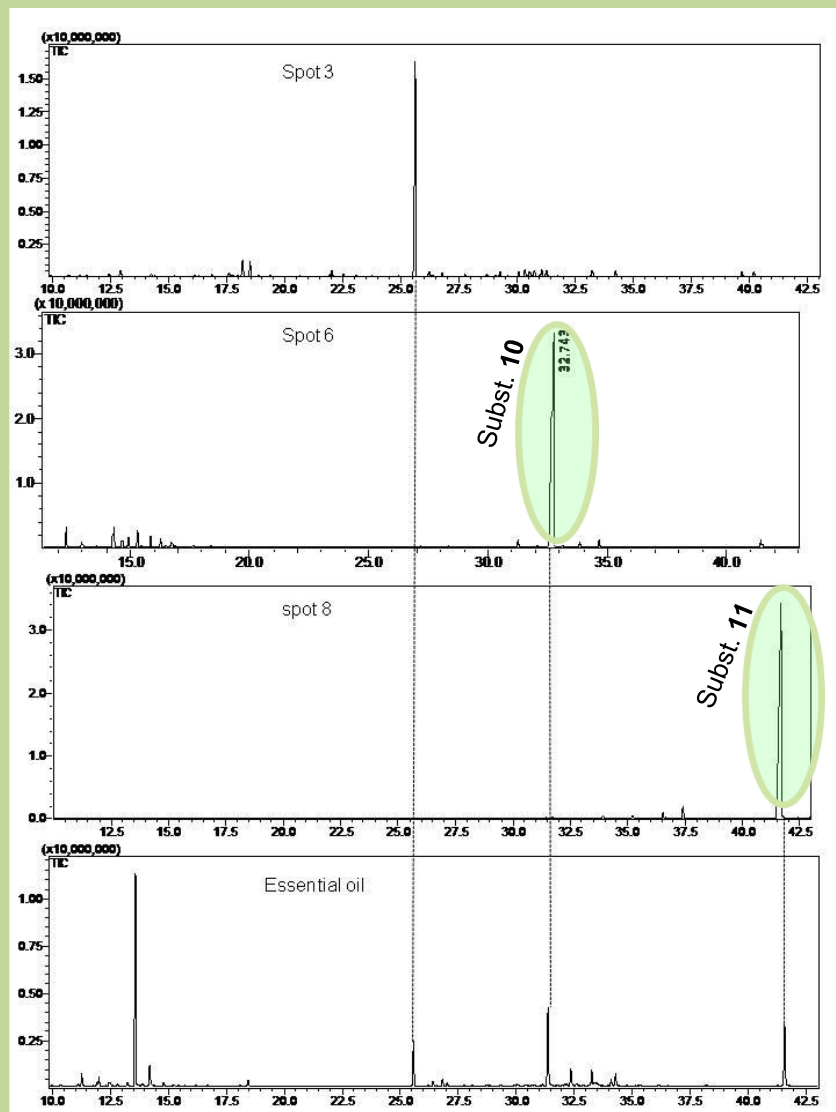
# Results and Discussion

# Chemical profile

Result/discus.



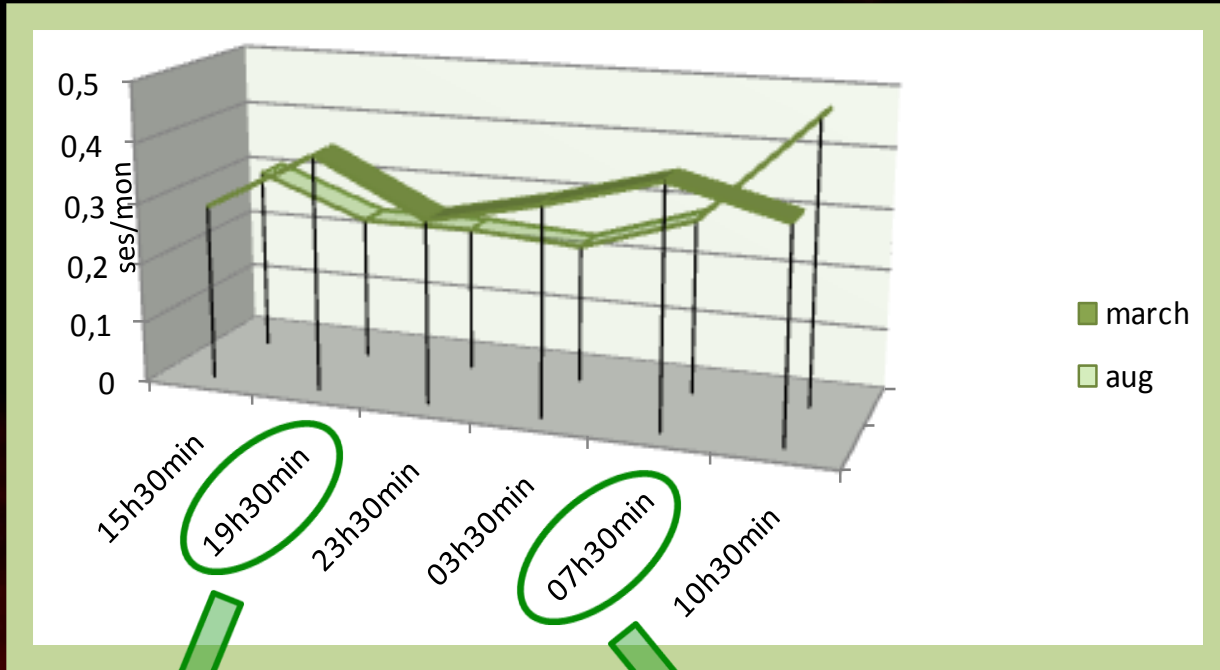
# Sesquiterpenes isolation



Comparing chromatograms of isolated compounds.

- Subst. 10 = 23,3mg
- Subst. 11 = 27,2mg

# Circadian shifts along the year



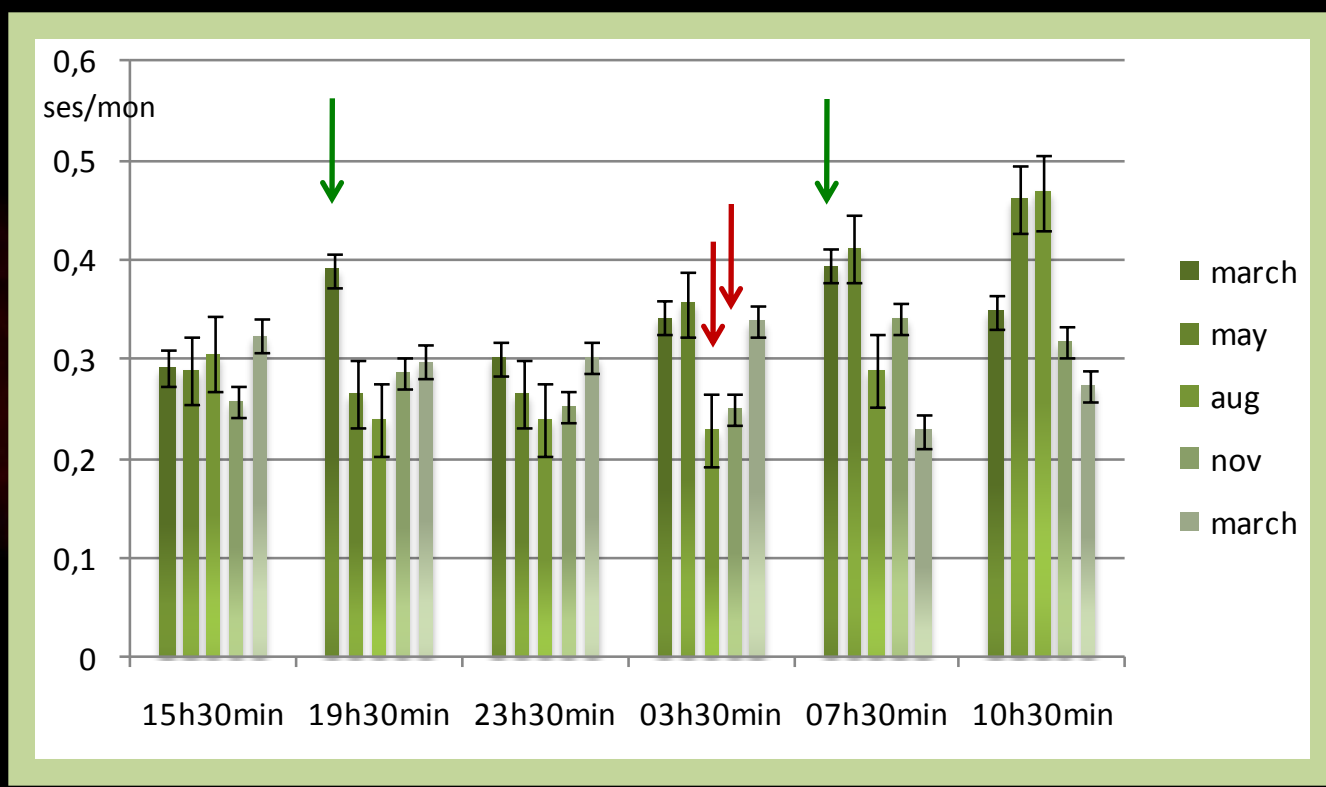
**Sunset**

**Sampled leaves during March**



**Sunrise**

# Circadian shifts along the year

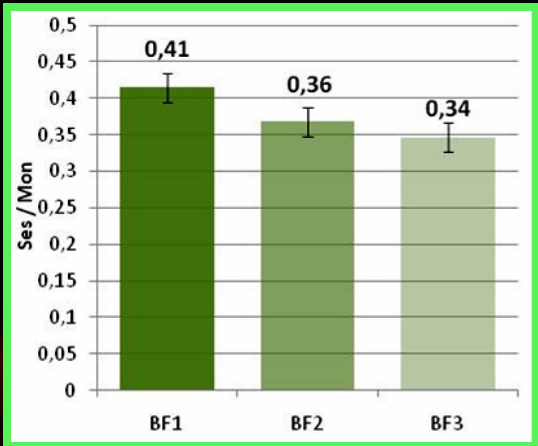


$$\frac{\sum \text{areas of 4 major sesquiterpenes}}{\sum \text{areas of 7 major monoterpenes}}$$

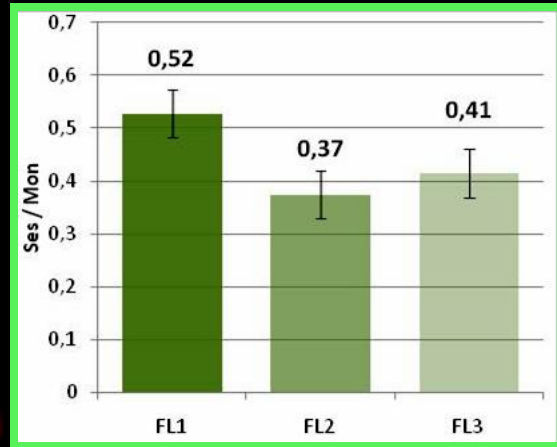


# Intraplant shifts

Result/discus.



Sampled leaves during March

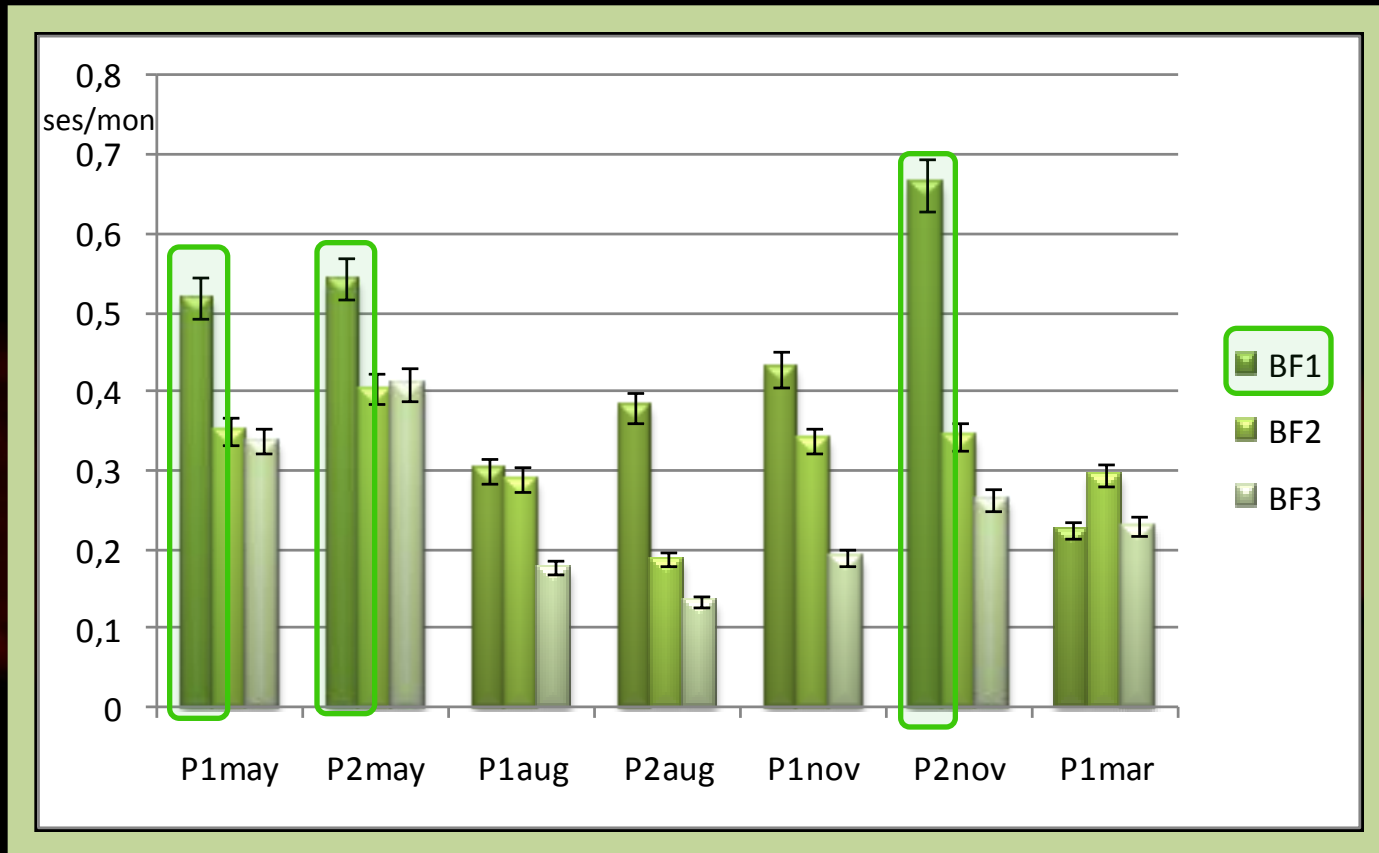




Flowerless branches



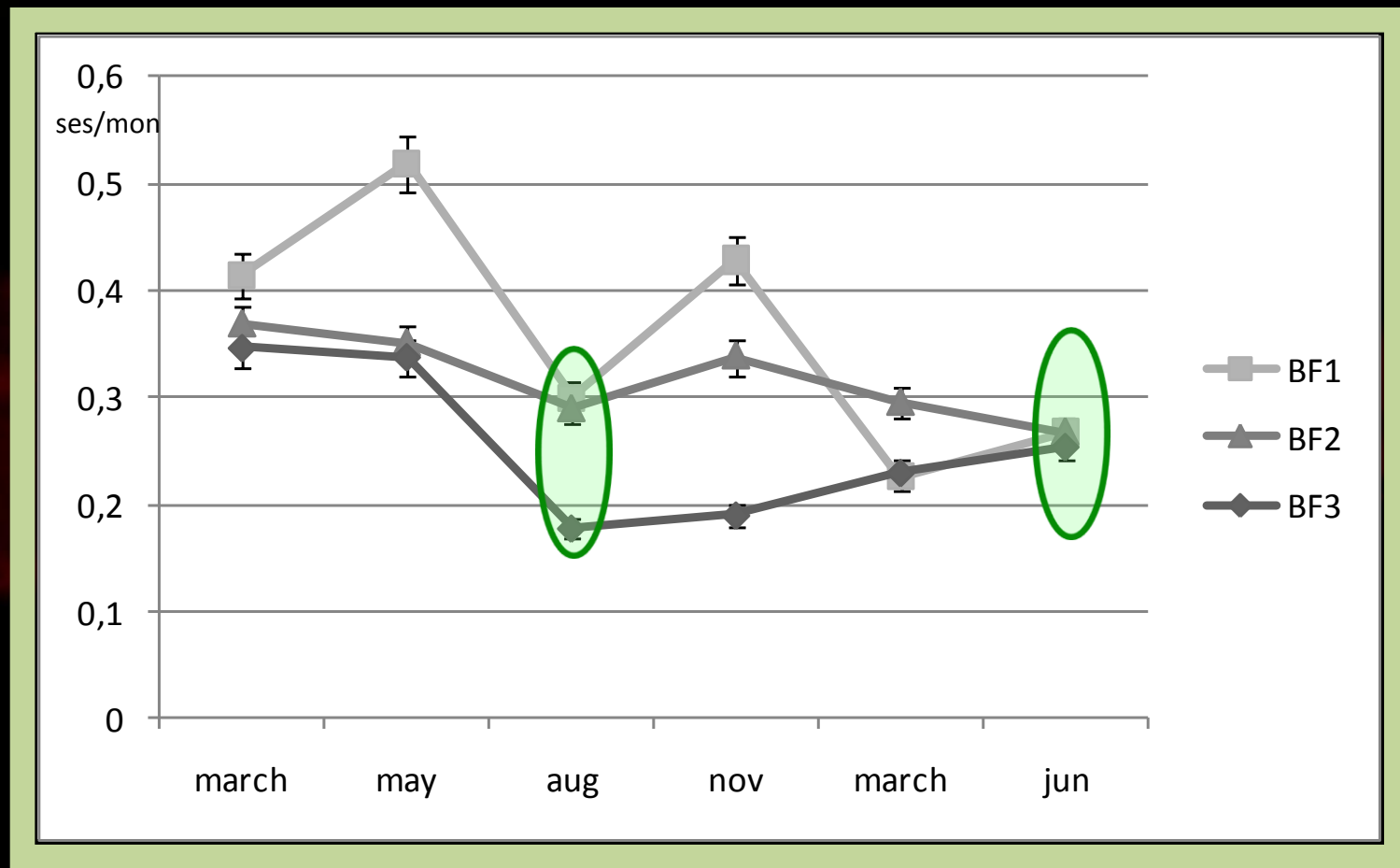
Capitulum branches

# Intraplant shifts



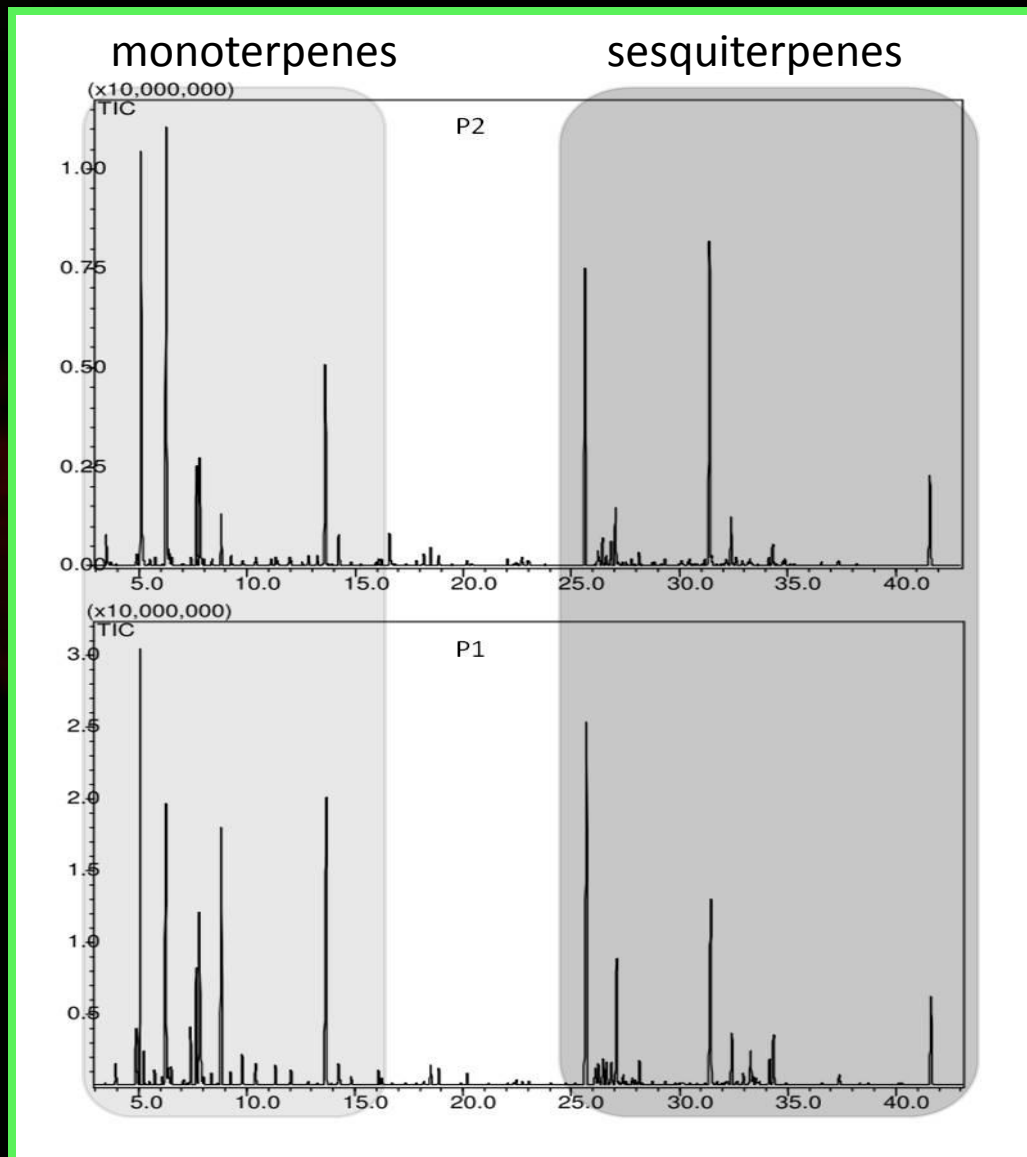
 P1= "paraíso"  
 P2= "pedreira"

# Intraplant shifts



 P1= "paraíso"

# Intraspecific uniformity



**Comparing chromatograms of BF1 leaves from sites pedreira (P2) and paraíso (P1). Period:May.**



# Main Remarks



**New sesquiterpenes are reported**



**A variety of bisabolane-like and one cadinane-like structure was observed in accordance with previous literature**



**Soft pre-concentration displayed a majority of monoterpenes and could also detect one major sesquiterpene, which was not present in essential oils**



**Data showed that previous investigations using essential oil were not in accordance with wild habitat behaviour**



**During the wet summer at the day time, in younger leaves, the individuals displayed higher relative levels of sesquiterpenes**



**Geographically different individuals are chemically quite similar**

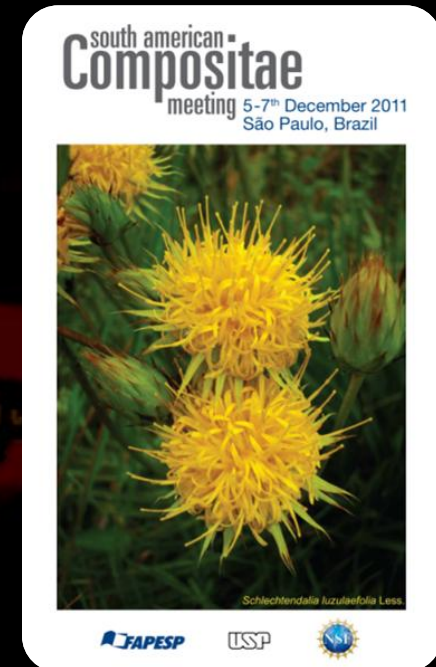


- 1-Gershenzon, J.; Dudareva, N. *Nature Chemical Biology*. V.3, P.408-414, 2007.
- 2- <http://florabrasiliensis.cria.org.br/opus>
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- 4- Stipanovic, R.D., Lopez, J.D., Jr., Dowd, M.K., Puckhaber, L.S. & Duke, S.E. *J. Chem. Ecol.* 32, 959–968 (2006).
- 5- Semir, J.. Tese De Doutorado, Instituto De Biologia, Unicamp, Campinas, P.515, 1991.
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- 8-Coile, N. C.; Jones, S. B.. *Brittonia*, V.33, N.4, P.528-542, 1981.
- 9-Curado, M.A.; Oliveira, C.B.A.; Jesus, J.B.; Santos, S.C.; Seraphin, J.C. Ferri, P.H. *Phytochemistry*. 67, P. 2363-2369, 2006.
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**FURNAS**

Sr. Luís Junio Leonel  
Mendes



# Time-scale and intraspecific variability of *Lychnophora ericoides* Mart. leave's volatile constituents.