

Compounds identified on hexane and dichloromethane extracts of *Salicornia ramosissima*

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Salicornia ramosissima J. Woods [4]

Introduction

Salicornia ramosissima J. Woods (common purple glasswort) is an annual halophyte, widely distributed in the salt marsh of Ria de Aveiro (Portugal), that belongs to the *Salicornia* L. genus (Chenopodiaceae) [1]. Although phytochemical studies on this genus report the presence of compounds which are well-recognized for their biological activities, such as flavonoids, chromones and alkaloids [2], too little is known about secondary metabolites on purple glasswort. In our previous work we identify ethyl *o*-hydroxycinnamate, (*E*)-fatty alcohol ferulic acid and scopoletin from the dichloromethane extract of *Salicornia ramosissima* aerial parts [3]. Here we report the most recent results obtained in further work to characterize the secondary metabolites of *Salicornia ramosissima*.

Aliphatic profile

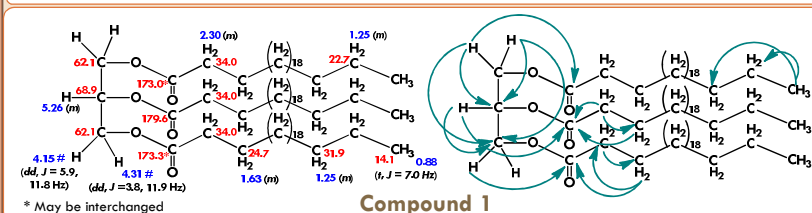
Analysis of hexane extracts by GC-MS revealed the presence of 27 fatty acids of which 6 are unsaturated, 15 alcohols, 7 sterols, 5 sugars and 3 cinnamic acids. The fatty acids, alcohols and sterols are the most abundant ones. Table 1 shows some of the identified compounds. Palmitic acid, tetracosan-1-ol and Docosan-1-ol represent the main components of the extracts.

Table 1 – Identified compounds by GC-MS

Identified Compounds		NE (%)	NE-H (%)	E (%)	E-H (%)
Fatty acids					
Palmitic acid	C16:0	2.54	5.32	3.32	6.55
Tetracosanoic acid	C24:0	0.63	1.02	0.56	1.17
Docosanoic acid	C22:0	0.56	0.95	0.65	1.39
Octadec-11-enoic acid	C18:1 (11E)	0.15	0.41	0.18	0.57
Tricosanoic acid	C23:0	-	0.31	-	0.40
2-Hydroxy-heptanoic acid	COOHCH(OH)(CH ₂) ₄ CH ₃	-	0.03	-	0.04
Fatty alcohols					
Tetracosan-1-ol	C24-OH	2.50	2.33	2.42	3.26
Dososan-1-ol	C22-OH	1.55	2.70	1.87	3.17
Hexacosan-1-ol	C26-OH	1.45	1.13	1.28	1.69
tridecan-2-ol	C13-OH	-	0.05	-	0.10
Pentacosan-1-ol	C25-OH	-	-	0.14	0.20
Sterols					
Stigmasterol		0.78	0.60	0.67	0.92
β-sitosterol		0.66	0.77	0.74	1.13
Sitostanol		0.46	0.64	0.60	0.87
Aromatic acid					
Isoferulic acid		-	0.51	-	0.23

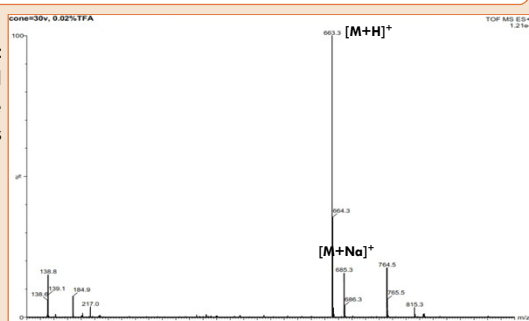
The lipid profile among *Salicornia* species is quite distinct however the fatty acid content in *Salicornia ramosissima* is similar to the *Salicornia fruticosa* [5], the saturated fatty acids are present in higher amount (87%) than unsaturated (13%) and palmitic acid (41,9%) represent the main component of the fatty acids fraction.

The analysis of the dichloromethane extract from *S. ramosissima* aerial parts allowed the isolation of the triacylglycerol (1) and the 2,2'-((oxybis(3-(tert-butyl)-6,1-phenylene))bis(propane-2,2-diyl))bis(4-(tert-butyl)phenol) (2). Here we present their unequivocal structure elucidation.



The spectroscopic characterization of compound 2 led to an unique structure with uncommon *t*-butyl groups. Is not the first time that a compound like this is discovered in nature [5]. The structure's was confirmed by mass spectroscopy (Fig 3).

Fig 3



Acknowledgments

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References

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Material and Methods

Plant collected in Ria de Aveiro (Portugal) from distinct growth conditions:
NE – growth with natural irrigation
E - growth with additional irrigation with canal water from 15 to 15 days

Plant air dried and aerial parts extract with dichloromethane or hexane at room temperature

Dichloromethane extract

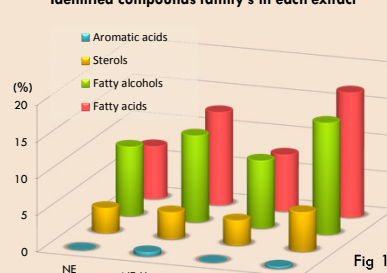
Silica gel column chromatography (gradient mixtures of hexane and ethyl acetate)

Silica gel TLC (different polarity eluent mixtures)

Compounds 1 and 2

Two hexane extracts: NE and E
 Alkaline hydrolysis
 Silylation
 GC-MS
 Compounds in table 1

Identified compounds family's in each extract

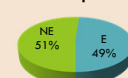


Prior to the hydrolysis of long chain alcohols are the most abundant family of compounds. This will change significantly after hydrolysis since fatty acids become majority (Fig 1).

The compounds tricosanoic, 2-hydroxy-heptanoic, isoferulic acids and the tridecan-2-ol exist exclusively in ester form, since they appears only after hydrolysis.

In the plant irrigated in excess (E), is observed an increase in the production of esterified compounds (Fig 2). The main alteration is the production of pentacosan-1-ol, nonexistent in NE.

Free compounds



Esterified compounds

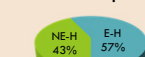


Fig 2

Spectroscopic characterization

In order to elucidate the fatty acid linked to the glycerol, the compound 1 was submitted to an alkaline hydrolysis and the result analyzed by GC-MS. The tetracosanoic acid was identified.

