

VALORIZATION OF AUTOCHTHONOUS HALOPHYTE PLANTS THROUGH HYDROPHILIC INTERACTION CHROMATOGRAPHY COUPLED TO Q EXACTIVE MASS SPECTROMETER



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INTRODUCTION

Plant lipidomics is a newly emerging omics platform aiming to cover the most diverse topics related to lipid including metabolism adaptation to external conditions. Halophytes are salt tolerant plants rich in lipids, namely polar lipids stored in their cell membranes and in their seeds. These plants have great potential for different biotechnological applications: phytoremediation of organic rich effluents, biodiesel and food for human consumption. However, the lack of information on the lipidome of halophytes is currently a gap in knowledge that impairs researchers to unravel the true biotechnological potential of these remarkable plants.

To overcome this drawback we used a lipidomic approach based on hydrophilic interaction chromatography coupled to Q-Exactive[®] high resolution mass spectrometer (Thermo Scientific) to characterize the lipidome of sea purslane (*Halimione portulacoides*), an autochthonous halophyte present in Portuguese saltmarshes.

RESULTS

HILIC-LC-Q-EXACTIVE-MS

HILIC coupled to a Thermo Scientific Q Exactive[®] HRMS, was used to identify the lipidome signature of *Halimione portulacoides*.

Analysis was performed by LC-MS and MS/MS in simultaneous positive and negative modes with high resolution mass spectrometry information from the Q-Exactive[®] mass spectrometer with Orbitrap[®] technology.

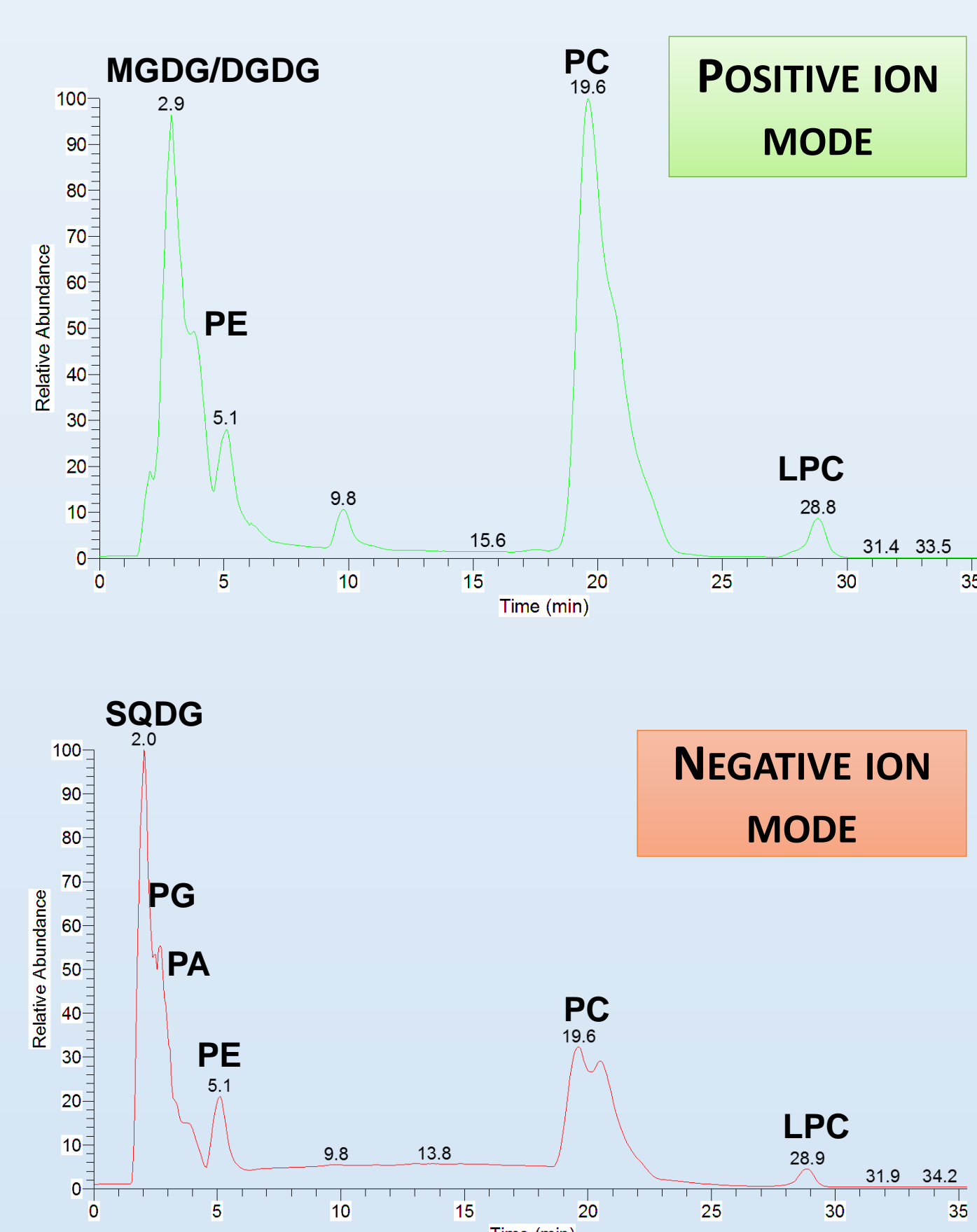


Figure 1: Total ion chromatograms obtained in positive and negative ion modes that allowed the separation of each lipid class from total lipid extract of *Halimione portulacoides*.

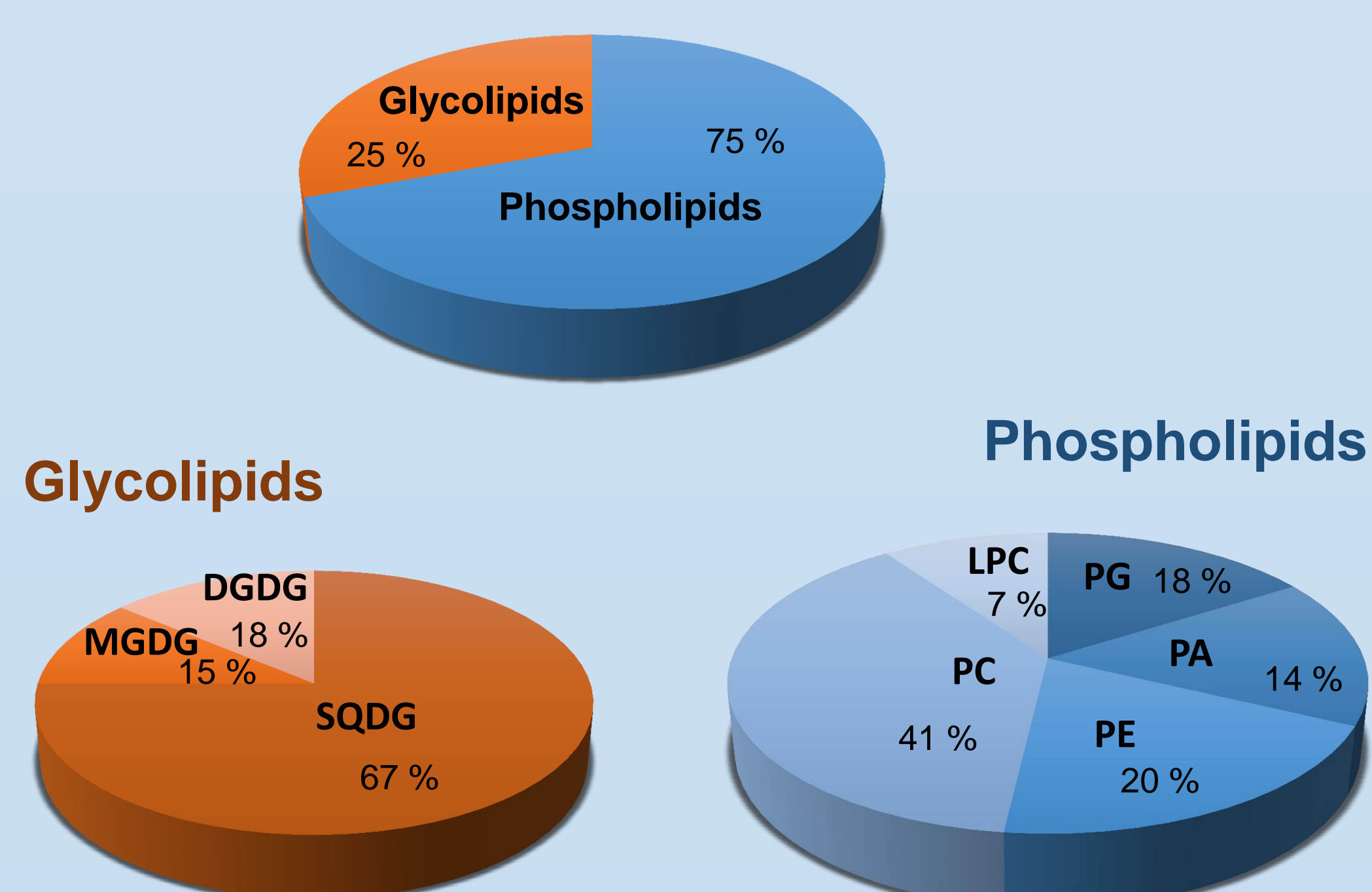
Table 1- Representative structure of the phospholipids and glycolipids identified in leaves of *Halimione portulacoides*

PHOSPHOLIPIDS	GLYCOLIPIDS
<chem>CCCCCCCCCCCCCCCC(=O)OCCOP(=O)(OCCO)OCCO</chem> Phosphatidylglycerol (PG)	<chem>OC[C@H]1O[C@@H](OC(=O)R2)[C@H](O)[C@@H](O)[C@H]1O</chem> Monogalactosyldiacylglycerol (MGDG)
<chem>CCCCCCCCCCCCCCCC(=O)OCCOP(=O)(O)O</chem> Phosphatidic acid (PA)	<chem>OC[C@H]1O[C@@H](OC(=O)R2)[C@H](O)[C@@H](O)[C@H]1O</chem> Digalactosyldiacylglycerol (DGDG)
<chem>CCCCCCCCCCCCCCCC(=O)OCCOP(=O)(OCCN)O</chem> Phosphatidylethanolamine (PE)	<chem>OC[C@H]1O[C@@H](OC(=O)R2)[C@H](O)[C@@H](O)[C@H]1O</chem> Sulfoquinovosyldiacylglycerol (SQDG)
<chem>CCCCCCCCCCCCCCCC(=O)OCCOP(=O)(OCCN(C)C)O</chem> Phosphatidylcholine (PC)	
<chem>CCCCCCCCCCCCCCCC(=O)OCCOP(=O)(OCCN(C)C)O</chem> Lysophosphatidylcholine (LPC)	

DISTRIBUTION OF LIPID SPECIES IDENTIFIED IN EACH LIPID CLASS

➤ Results gathered in the present work allowed to identify the **lipidome signature** of *Halimione portulacoides*

➤ More than 100 species different lipid species from phospholipids (5 classes) and glycolipids (3 classes) were assigned to the total polar lipid composition .



➤ The PC was the class with the greatest diversity of molecular species. All species are represented in Table 2.

➤ In both phospholipids and glycolipids the most abundant molecular species contain FA18:3 n-3. The α-Linolenic acid (18:3(n-3)) was the predominant fatty acid in all lipid classes.

Polar lipids have great potential as bioactive compounds, particularly in what concerns their putative use as anti-inflammatory, anti-proliferative, anti-microbial and anti-oxidative properties.

METHODOLOGY

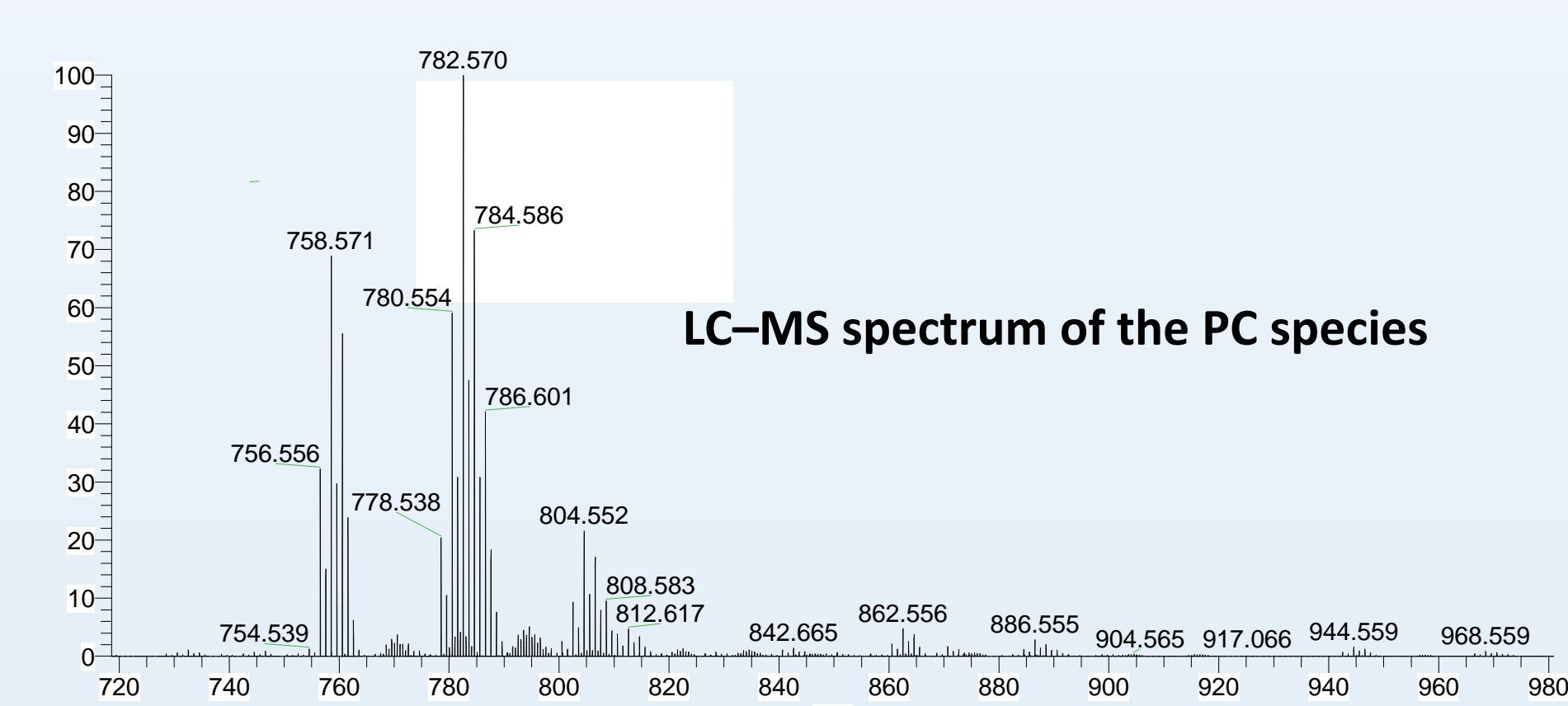
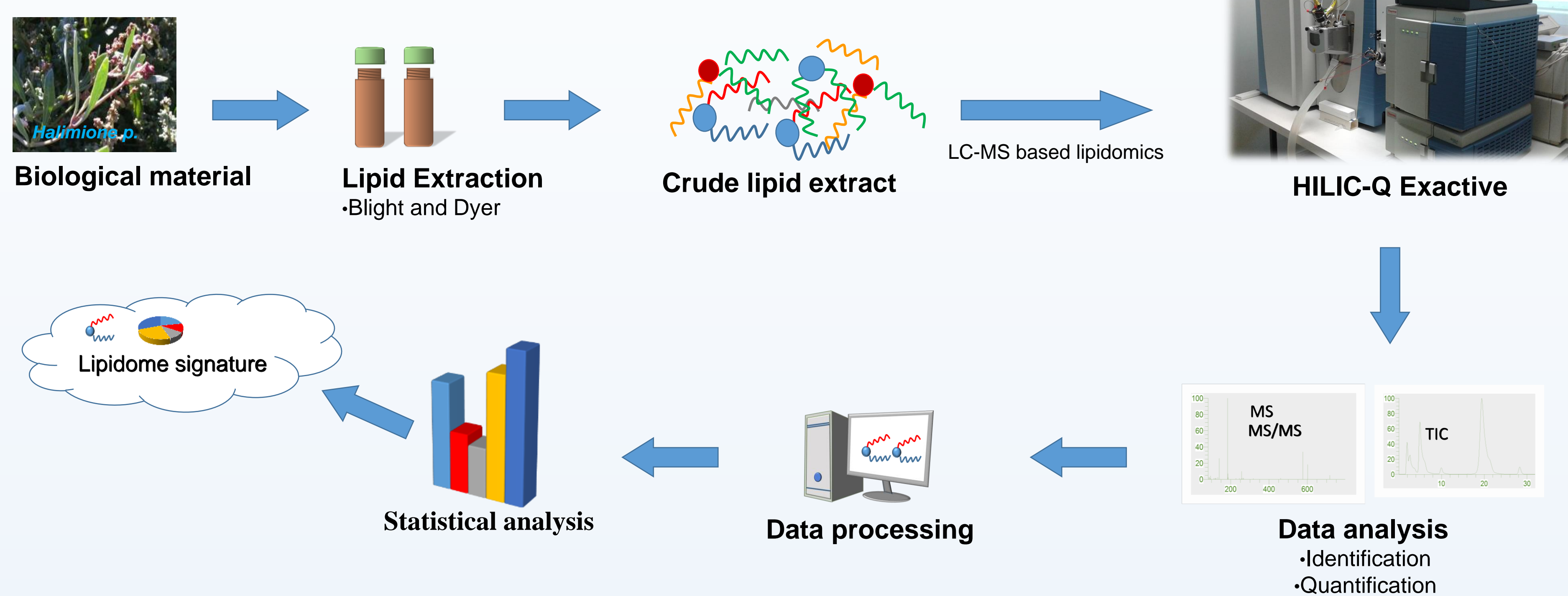


Figure 2: HILIC-Q Exactive-MS obtained in the Q Exactive Orbitrap between 19-21 min representative of the profile of phosphatidylcholine molecular species

Table 2: Molecular species observed by HILIC-Q Exactive-MS, as [M+H]⁺ ions; C means number of carbon atoms and N represents the number of double bonds in the fatty acyl side chains. m/z values in bold correspond to the most abundant species detected in the LC-MS spectrum.

[M+H] ⁺	C:N	PC	[M+H] ⁺	C:N	PC
732.554	32:1	16:0/16:1	800.522	38:9	18:4/20:5
734.569	32:0	16:0/16:0	802.538	38:8	18:3/20:5
756.556	34:3	16:0/18:3	804.552	38:7	18:3/20:4
758.571	34:2	16:0/18:2,16:1/18:1	806.569	38:6	18:2/20:4;18:1/20:5
760.585	34:1	16:0/16:1	808.583	38:5	18:3/20:1
768.554	35:4	17:1/18:3	810.600	38:4	18:0/20:4
770.569	35:3	17:0/18:3	812.616	38:3	18:3/20:0
772.585	35:2	17:0/18:2	814.632	38:2	18:1/20:1
774.601	35:1	17:0/18:1	826.538	40:10	20:5/20:5
778.538	36:6	18:3/18:3	828.553	40:9	20:4/20:5
780.554	36:5	18:2/18:3, 16:1/20:4	836.616	40:5	18:3/22:2
782.570	36:4	18:1/18:3; 18:2/18:2	838.632	40:4	18:3/22:1;18:2/22:2
784.586	36:3	18:1/18:2; 18:0/18:3	840.647	40:3	18:3/22:0;18:2/22:1
786.601	36:2	18:1/18:1, 18:0/18:2, 16:2/20:0	842.665	40:2	18:2/22:0;18:1/22:1
788.616	36:1	18:0/18:1, 16:1/20:0	844.679	40:1	18:1/22:0
792.554	37:6	17:1/20:5	868.679	42:3	18:2/24:1; 16:0/26:1
794.569	37:5	17:0/20:5	870.694	42:2	18:1/24:1;18:2/24:0
796.585	37:4	19:1/18:3	872.710	42:1	18:1/24:0
798.601	37:3	19:0/18:3	874.725	42:0	18:0/24:0

CONCLUDING REMARKS

The polar lipidome of halophytes is remarkably diverse and its screening represents a significant analytical challenge. Modern research platforms, particularly mass spectrometry based lipidomic approaches, have been recently used to address this challenge.

This presentation highlights the advantage of using a lipidomic platform to unravel potential bioactive phytochemicals. Moreover, this approach also promotes the added value of autochthonous halophyte crops that can foster new applications on the fields of healthy and functional food products, nutraceuticals and source of multiple bioactive compounds.

ACKNOWLEDGEMENTS

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