



## TABLE OF CONTENTS

	Introduction	1
<b>1</b>	<b><u>GENERAL CONCEPTS</u></b>	<b>3</b>
<b>1.1</b>	<b>Baby Banana (<i>Musa acuminata</i> AA Simmonds cv. Bocadillo)</b>	<b>4</b>
1.1.1	Taxonomic Classification of Bananas ( <i>Musa acuminata</i> )	4
1.1.2	Plant morphology	5
1.1.3	Pre-harvest	8
1.1.4	Post-harvest	10
1.1.4.1	<i>Hyperpigmentation phenomenon</i>	12
<b>1.2</b>	<b>Chlorophyll degradation</b>	<b>14</b>
<b>1.3</b>	<b>High-Speed Countercurrent Chromatography</b>	<b>24</b>
<b>1.4</b>	<b>Spiral-Coil Low-Speed Rotary Countercurrent Chromatography (Spiral-Coil LSRCCC)</b>	<b>27</b>
<b>2</b>	<b><u>RESULTS AND DISCUSSION</u></b>	<b>29</b>
<b>2.1</b>	<b>Phytochemical profile of Baby Banana control peels</b>	<b>30</b>
2.1.1	Phosphatidylcholine (Lecithin)	31
2.1.2	Glycosyldiacylglycerolipids	38
2.1.2.1	<i>O-<math>\alpha</math>-D-Galp (1''<math>\rightarrow</math> 6')- O-<math>\beta</math>-D-Glup (1'<math>\rightarrow</math> 3)- 2, 1-diacyl-L-glycerol (GGDG)</i>	39
2.1.2.2	<i>O-<math>\beta</math>-D-Galp(1'''-3)-2, 1 diacyl-L-glycerol (MGDG)</i>	55
2.1.2.3	<i>Compositional analysis of the glycolipid GGDG and MGDG</i>	70
2.1.2.4	<i>Summary</i>	75
2.1.3	Molecular species of glucocerebroside	79
2.1.4	Sulphoquinovosyl-diacylglycerol (SQDG)	88



2.1.5	Phosphatidylethanolamine (PE)	90
2.1.6	Ceramide aminoethylphosphonate (CAEP)	92
2.1.6.1	<i>Summary</i>	93
2.1.7	Linoleic acid	94
2.1.7.1	<i>Summary</i>	103
2.1.8	Lutein	104
2.1.9	Violaxanthin-di-laurate and $\beta$ -Carotene isomers	110
2.1.9.1	<i>Summary</i>	123
2.1.10	4-Epi-cycloeucalenone and 4-Epi-cyclomusalenone	127
2.1.11	$\beta$ -Sitosterol	134
2.1.11.1	<i>Summary</i>	138
2.1.12	Pheophytin-a	141
2.1.13	Summary	152
<b>2.2</b>	<b>Optimization of the solvent system for the separation of chlorophylls and derivatives by means of High-Speed Countercurrent Chromatography in Baby Banana peels, spinach and grass</b>	154
2.2.1	Experimental Design	154
2.2.2	Isolation of chlorophylls and derivatives from spinach by HSCCC	167
2.2.3	Role of dichloromethane and chloroform as stationary phase in the solvent system for the isolating of chlorophylls and xanthophylls from plant extracts by HSCCC	178
2.2.4	Chlorophylls <i>a/b</i> ratio and derivatives in Baby Banana peels and other plants by HSCCC	184
<b>2.3</b>	<b>Preparative Spiral-Coil Low Speed Rotary Countercurrent chromatography of Baby Banana peels (<i>Musa acuminata</i>) with hyperpigmentation.</b>	190
2.3.1	APCI-HPLC-MS-MS analysis of Spiral-Coil-LSRCCC fractions from Baby Banana peels with hyperpigmentation	194
2.3.2	Identification of polar compounds in Spiral-Coil-LSRCCC	204



2.3.3	Identification of chlorophyll derivatives esterified with sterols in Spiral-Coil-LSRCCC	207
2.3.4	Spiral-Coil-LSRCCC of Baby Banana peels with hyperpigmentation: comparison with HSCCC of Baby Banana control	210
2.3.4.1	<i>Elucidation of tocopherols and tocotrienols in Fraction 14 of Spiral-Coil-LSRCCC separation from Baby Banana peels with hyperpigmentation extract</i>	217
2.3.4.2	<i>Elucidation of Triterpene Alcohol Ferulates in Fraction 14 of Spiral-Coil-LSRCCC separation from Baby Banana peels with hyperpigmentation extract</i>	229
2.3.5	Summary	233
<b>3</b>	<b><u>SUMMARY AND OUTLOOK</u></b>	<b>235</b>
<b>4</b>	<b><u>MATERIAL AND METHODS</u></b>	<b>239</b>
<b>4.1</b>	<b>Plant Material and Chemicals</b>	<b>240</b>
4.1.1	Plant Material	240
4.1.2	Chemicals and Solvents	240
<b>4.2</b>	<b>Equipments and Parameter</b>	<b>242</b>
4.2.1	High-Performance Liquid Chromatography (HPLC)	242
4.2.1.1	<i>Jasco-System (HPLC-DAD)</i>	242
4.2.1.2	<i>Knauer-System (preparative) – Equipment I</i>	242
4.2.1.3	<i>Knauer-System (preparative) – Equipment II</i>	242
4.2.1.4	<i>Analytical Columns</i>	243
4.2.1.5	<i>Preparative Columns</i>	243
4.2.1.6	<i>Solvent System and Gradients of HPLC – DAD</i>	243
4.2.1.7	<i>Solvent System and Gradients of Preparative HPLC</i>	244
4.2.2	Mass spectrometry	245
4.2.2.1	<i>APCI-HPLC-MS-MS</i>	245
4.2.2.2	<i>Column APCI-HPLC-MS-MS</i>	245



4.2.2.3	<i>Parameter Mode APCI-HPLC-MS-MS</i>	245
4.2.2.4	<i>Solvent system and gradient of APCI-HPLC-MS</i>	245
4.2.3	High-Speed Countercurrent Chromatography (HSCCC)	246
4.2.3.1	<i>Solvent System of High-Speed Countercurrent Chromatography (HSCCC)</i>	246
4.2.4	Gas Chromatography	247
4.2.4.1	<i>GC-MS I</i>	247
4.2.4.2	<i>GC-MS II</i>	247
4.2.4.3	<i>GC-MS III</i>	247
4.2.5	Spiral-Coil Low-Speed Rotary Countercurrent Chromatography (Spiral-Coil LSRCCC)	248
4.2.5.1	<i>Solvent system for Spiral-Coil LSRCCC</i>	248
4.2.6	Nuclear Magnetic Resonance Spectroscopy	248
<b>4.3</b>	<b>Methods</b>	249
4.3.1	Extraction Methods	249
4.3.1.1	<i>First Extraction Method</i>	249
4.3.1.2	<i>Second Extraction Method</i>	249
4.3.2	Thin Layer Chromatography (TLC)	250
4.3.2.1	<i>System 1</i>	250
4.3.2.2	<i>System 2</i>	250
4.3.3	Compositional analysis of glycolipids	250
4.3.3.1	<i>Analysis of fatty acid components by Gas chromatography–mass spectrometry (GC-MS)</i>	250
4.3.3.2	<i>Sugar composition of glycolipids (SCG)</i>	251
4.3.4	Isolation of pheophytin from methanol phase of Baby Banana peels by means of Normal Phase Chromatography	252
4.3.5	Isolation of carotenoids from acetone extract of Baby Banana peels with hyperpigmentation by means of Normal Phase Chromatography	252



4.3.6	Partial isolation of chlorophyllase and proteins from flavedo of Baby Banana peels	253
4.3.6.1	<i>SDS/PAGE condition</i>	254
5	<b><u>BIBLIOGRAPHY</u></b>	255