ABSTRACT

Name: AHMED MELEGI ABDEL-GHANY MELEGITitle of thesis : Plant extracts as effective compounds in insect controlDegree: Ph.D Thesis, Faculty of Science, Cairo university 2005/2006

The presence study is focusing on the chemical constituents of mango leaves that could implicate in the resistance of local Egyptian mango (*Mangifera indica*) cultivar called Alphonso to scale insect infestation. For comparison, the same chemical and biological investigation steps were also conducted on the highly susceptible mango cultivar (Sultani) in addition to three other susceptible mango cultivars (Zebda, Baladi and Hindi). The Acuminata scale insect, Kilifia acuminata (Signoret) nymphs were used as a bio-indicator. Leaf components were extracted with solvents with different polarities, and fractionated using several column chromatographies, preparative TLC and preparative HPLC. Finally, the isolated bioactive compounds were characterized and quantified with various spectroscopic methods including GC-MS, ¹H-NMR, ¹³C-NMR, *IR*, and *UV*. Six bioactive compounds were isolated, of which three were isolated from hexane extract of Alphonso leaves and were found to be moderately repellant to K. acuminata; two compounds were isolated from hexane extract of Sultani leaves and were found to be highly attractant to K. acuminata and one compound was isolated from methanol extract of Alphonso leaves and was found to be toxic against K. acuminata. The repellant compounds isolated from Alphonso leaves were identified as α -pinene, β -pinene and limonene. The attractant compounds extracted from Sultani leaves were identified as cis-ocimene and trans-ocimene while the toxic compounds isolated from Alphonso leaves was identified as pyrogallol. The later compound has not been previously reported as a constituent of *M. indica* species. The overall results of this study suggest that the combined action of three repellant secondary metabolites, namely: α -pinene, β -pinene and limonene and one toxic compound, namely pyrogallol play an important role in resistance of Alphonso mango cultivar against scale insect pests in general or at least against K. acuminata scale insect.

Key words: Plant extracts, Identification, Isolation, *kilifia acuminata*, Mango, Biological effect, Insect control.

Supervisors:

1- Prof. Dr. Fathy Abd El-gelil

2- Prof. Dr. Mohamed Farag El-Said

Prof. Dr. Refaat Hasan Helal

Chairman of Chemistry Department Faculty of Science – Cairo University

TABLE OF CONTENTS:

I -		INTRODUCTION	1
II-		REVIEW OF LITERATURE	5
	1.	1. Groups of plant secondary metabolites	6
	1.1	1.1. Terpenes (terpenoids)	6
	1.2	1.2. Phenolics	8
	1.3	1.3. Nitrogenous compounds	10
	2	2. Plant secondary metabolites as a resistance trait against herbivores	12
	3	3. Mango, <i>Mangifera indica</i> L	13
	3.1	3.1 Botanical Aspect of Mangifera indica L	13
	3.2	3.2. The Family Anacardiaceae	14
	3.3	3.3. Overview of the important secondary compounds within the Family Anacardiaceae	14
	3.4	3.4 Back ground on <i>Mangifera indica</i> cultivars	19
	4	4. Chemical Investigations on <i>Mangifera indica</i> cultivars	20
	5	5. Relation between M. indica cultivars and insect pests	33
	6	6. Structural skeletons of the major compounds constituents identified in <i>M. indica</i>	37
III-		MATERIALS AND METHODS	47
	1	1. Plant Material.	47
	2	2. Sample extraction	47
	3	3. Isolation of bioactive components	48
	3.1	Hexane extract	48
	3.1.1	Silica Gel Column Chromatography (Column 1)	48
	3.1.2	Silica Gel Column Chromatography (Column 2)	49
	3.1.3	Preparative TLC	49
	3.2	Methanol extract	50
	3.2.1	Sephadex Column Chromatography	50
	3.2.2	Analysis and Concentration of Fractions	50
	3.2.3	Assessment of the fractions	51
	3.2.4	Preparative high-performance liquid chromatography (HPLC)	51
	4	Identification of the bioactive compounds	51
	4.1	Gas Chromatographic-Mass Spectrometric (GC-MS) Analysis	52
	4.2	Nuclear Magnetic Resonance (NMR)	52
	4.2	Ultraviolet Spectroscopy	53
	4.4	FT-IR spectroscopy	53
	т.т 5	Quantitative assessment.	53
	6	Rapid screening of compounds presented in Methanol	53
	0	rupte servening of compounds presented in methanion	55

		fractions	
	7	Laboratory bioassays	54
	7.1	Experimental insect	54
	7.2	Insect collection	54
	7.3	Behavioral assays	54
	7.3.1	Multi Choice bioassay	54
	7.3.2	No-choice bioassay	56
	7.3.3	Toxicity bioassay	56
	8	Data analysis	57
IV-		RESULTS	58
	1.	Extraction	58
	2.	Biological effects of leaf extracts	60
	3.	Isolation, purification and characterization of bioactive	60
		compounds in hexane extracts of both Alphonso and Sultani	
		leaves	
	3.1	TLC	60
	3.2	Column chromatography	60
	3.3	Separation of H_{Alf2} and H_{Sul-2} fractions	65
	3.4	Preparative TLC	65
	3.5	Identification and quantification of the isolated bioactive	65
		components	
	3.6.1	Identification of compounds in the bioactive isolate number	69
		1	
	3.6.2	Identification of compounds in the bioactive isolate number	75
	262	2 Identification of compounds in the bioactive isolate number	01
	3.6.3	Identification of compounds in the bioactive isolate number	81
	3.6.4	5 Identification of compounds in the bioactive isolate number	81
	3.0.4	11	01
	3.7	Isolation, purification and characterization of bioactive	89
		compounds in methanol extract of Alphonso leaves	• •
	3.7.1	Sephadex LH-20 column chromatography	93
	3.7.2	Preparative HPLC	93
	3.7.3	Identification and quantification of the isolated bioactive	97
		compound M1	
	4	Combined effects of the isolated compounds from Alphonso	106
		leaves	
	5.	Screening of the identified bioactive compounds on other <i>K</i> .	109
		acuminata susceptible mango cultivars	
V -		DISCUSSIONS AND CONCLUSIONS	114
·	1.	Isolation and identification of the bioactive compounds	114
	2.	Conclusion	120
VI-	-•	SUMMARY	121
VII-		REFERENCES	122
VIII-		ARABIC SUMMARY	1
V 111-			

ii