

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قال :

وَهُوَ الَّذِي أَنْشَأَ السَّمَاوَاتِ وَالْأَرْضَ مِنْ هَذِهِ الْأَنْعَمِيَّةِ مِنْهُ مِنْهُ طَلْعَهَا قِنْوَانُ دَائِنَيَّةٍ وَجَنَابٌ مِنْ أَغْنَابِهِ وَالرَّزْمَانَ إِذَا أَتَمَرَ وَيَنْعِمُ إِنَّ فِي ذَلِكُمْ لَآيَاتٍ لِقَوْمٍ يُؤْمِنُونَ

صدق

سورة (99) : العنكبوت

Dedication

To ...

My mother and my father

My sisters

Acknowledgement

I wish I am able to give him the appreciation he deserves, he never stopped giving me effort and time, and he offered me advice and support and never stopped giving them to me: Prof. Moham- med Abd Elkareem.

I am indepted to those who supported me and guided me in my search for knowledge: Dr.Haidar Abd.Algadir Mohamed, Dr. Elk-heir Mugudam, Dr. Alshikh Abd Allah, Dr. Alawea abd Allah.

To all those who contributed to this research I am extremely grateful.

Abstract

Phytochemical screening of the alcoholic extract of the *Mitracarpus hirtus* indicated the presence of flavonoids, alkaloids and glycosides. Steroids were not detected. It was decided to investigate the flavonoids of this herb due to their medicinal value and relative abundance.

The crude product obtained from the alcoholic extract was re-extracted with hexane, chloroform and ethyl acetate. The ethyl acetate extractive gave positive test for flavonoid but the chloroform extractive did not. The ethyl acetate extractive was subjected to paper chromatography, which indicated the presence of one major flavonoid. This flavonoid was obtained in the pure state by preparative paper chromatography.

The spectral studies (UV, IR) of this flavonoid indicated that it is a 5, 7- dihydroxydihydroflavonol.

الخلاصة

اجريت اختبارات فيتوكيميائيه للمستخلص الكحولي لعشبه حنبوت ، حيث اتضح انه يحتوى على فلافونيدات و قلويات و جليكوسيدات و عدم احتوائه على استيرويدات.

بدأت دراسه تفصيلية للفلافونيدات التي يحتويها النبات لوفرتها بالنبات ولأهميةها الطبية، حيث تم استخلاص المستخلص الكحولي للنبات مرة اخرى باستخدام الهكسان والكلوروفورم و خلات الإيثيل. اختبار الفلافونيد اعطى نتیجه ايجابيه عند استخدام مستخلص خلات الايثيل بينما اعطى نتیجه سلبيه عند استخدام مستخلص الكلوروفورم . استخدمت كروماتografيا الورقه لفصل الفلافونويد الرئيسي في الهيئة النقية من مستخلص خلات الإيثيل.

اجريت دراسات طيفيه(UV, IR) لهذا المركب حيث اوضحت انه من المحتمل ان يكون عباره عن 7,5- ثنائى هيدروكسى ثنائى هيدروفلافونول.

List of contents

الأدبيه	i
Dedication	ii
Acknowledgment	iii
Abstract	iv
الخلاصة	v
List of contents	vi

Chapter one

1-Introduction	1
1.1- General approach	1
1.2- Nomenclature	1
1.2.1- Phenylbenzopyran (C ₆ -C ₃ -C ₆ Backbone)	3
1.2.2- Isoflavonoids	4
1.2.3- Neoflavonoids	7
1.2.4- Minor flavonoids	7
1.3- Biosynthesis of flavonoids	9
1.3.1- Biosynthesis of flavonoid precursors	9
1.3.2- Formation of chalcones	10
1.3.3- Formation of aurones	11
1.3.4- Formation of flavanones	14
1.3.5- Formation of isoflavone	14

1.3.6- Formation of flavones	16
1.3.7- Formation of flavonols	18
1.3.8- Glycosylation	19
1.3.9- Methylation	21
1.4- Separation and quantification of flavonoids	21
1.4.1- Extraction	21
1.4.2- Preparative Separation	24
1.4.2.1- Preliminary Purification	24
1.4.2.2- Preparative Methods	25
1.4.2.2.1- High-Performance Liquid Chromatography	27
1.4.2.2.2- Medium-Pressure Liquid Chromatography	28
1.4.2.2.3- Centrifugal Partition Chromatography	29
1.4.3- Analytical Methods	30
1.4.3.1-Thin-Layer Chromatography	30
1.4.3.2- High-Performance Liquid Chromatography	32
1.4.3.3- High-Performance Liquid Chromatography- Mass Spectrometry	33
1.4.3.4- High-Performance Liquid Chromatography- Nuclear Magnetic Resonance	35
1.4.3.5- Capillary Electrophoresis	36
1.4.3.6- The Ultraviolet–Visible Absorption Spectra of Flavonoids	37
1.4.3.6.1- The UV-VIS spectra of flavonoids in methanol	38
1.4.3.6.1.1- Flavones and Flavonols	38
1.4.3.6.1.2- Isoflavones, flavanones and dihydroflavonols	39
1.4.3.6.1.3- Chalcones and aurones	40
1.4.3.6.1.4- Anthocyanidins and anthocyanins	40
1.4.3.6.2-The structural significance of sodium methoxide induced Shift	41
1.4.3.6.2.1- Flavones and flavonols	41
1.4.3.6.2.2- Isoflavones, flavanones and dihydroflavonols	42
1.4.3.6.2.3- Chalcones and aurones	42
1.4.3.6.2.4- Anthocyanidins and anthocyanins	43
1.4.3.6.3- The structural significance of sodium acetate induced shifts	43
1.4.3.6.3.1- Flavones and flavonols	43
1.4.3.6.3.2- Isoflavones, flavanones and dihydroflavonols	44
1.4.3.6.3.3- Chalcones and aurones	44
1.4.3.6.4- The structural significance of borate induced	44

shifts	
1.4.3.6.5- The structural significance of aluminum chloride induced Shifts	45
1.4.3.6.5.1- Flavones and flavonols	45
1.4.3.6.5.2- Isoflavones, flavanones and dihydroflavonols	45
1.4.3.6.5.3- Chalcones and aurones	46
1.4.3.6.5.4- Anthocyanidins and anthocyanins	46
1.5- Flavonoids as nutraceuticals	47
1.5.1-Free Radicals	47
1.5.1.1- Reactive oxygen species	48
1.5.1.2- Reactive nitrogen species (RNS)	48
1.5.2- Cytoprotective effects of flavonoids	49
1.5.3- Protection against peroxide and other reactive oxygen species	50
1.5.4- Suppression of cancer growth	51
1.5.4.1- Antiproliferative effects	51
1.5.4.2- Inhibition of cell cycle progression	52
1.6- Flavonoid function in plants	53
1.6.1- Defense against pathogenic microbes	53
1.6.2- Protection from solar ultraviolet	54
1.6.3- Antioxidant activity	55
1.6.4- Heavy Metal Tolerance	55
1.7- <i>Mitracarpus hirtus</i>	56
1.8- Aim of this work	57

Chapter two

2- Materials and method	58
2.1- Materials	58
2.1.1- Collection of plant material	58
2.2- Methods	58
2.2.1- Preparation of test reagents for phytochemical screening	58
2.2.1.1- Flavonoid test reagents	58
2.2.1.2- Alkaloid test reagents	59
2.2.2- Preparation of plant extract	59
2.2.3- Phytochemical screening	59

2.2.3.1- Test for steroids	59
2.2.3.2- Test for alkaloids	60
2.2.3.3- Test for flavonoids	60
2.2.3.4 -Test for glycosides	61
2.2.4- Extraction of flavonoids from <i>Mitracarpus hirtus</i>	61
2.2.5- Paper chromatography of the ethyl acetate extractive	61
2.2.6- Preparative paper chromatography	62
2.2.7- UV shift reagents	62
2.2.8- The UV spectrum of FM in presence of UV shift reagents	63
2.2.8.1- The UV spectrum of FM in presence of sodium methoxide	63
2.2.8.2- The UV spectrum of FM in presence of AlCl ₃	63
2.2.8.3- The UV spectrum of FM in presence of AlCl ₃ /HCl	63
2.2.8.4-The UV spectrum of FM in presence of sodium acetate	64
2.2.8.5-The UV spectrum of FM in presence of boric acid/ sodium acetate	64

Chapter three

3-Results and discussion	65
3.1-Phytochemical screening	65
3.2-Identification of FM	65
Recommendations	72
References	

