# AIN SHAMS UNIVERSITY FACULTY OF SCIENCE

# A THESIS Entitled

### CHEMICAL STUDIES ON SOME EGYPTIAN DESERT PLANTS (ASPHODELUS SPECIES)

Submitted For the

Degree of Doctor of Philosophy

By

MEDHAT MAHROUS HASSAN SEIF EL-NASSR B.Sc., M.Se.



Supervised by

Prof. Dr. A A SAMMOUR
Dr. A M RIZK

Pharmaceutical Sciences Laboratory
National Research Centre
Cairo, U.A.R.



1973

## Chemical Studies on Some Egyptian Desert Plants (Asphodelus sjecies)

Thesis Advisor

Prof. Dr. A. Sammour

Prof. Dr. S. Tobia

Head of Chemistry Department



#### ACKHOWLEDGEMBHT

I wish to express my sincere thanks to Prof.

Dr. A. Samsour, Professor of Organic Chemistry, Chemistry

Department, Faculty of Science, Ain Shams University,

for soponsoring the work reviewing the text and for

his constructive criticism.

I am deeply indebted to Dr. A.M. Rizk and Dr. F.M. Hammouds, Assistant Professors of the Pharmaceutical Sciences Laboratory, Estional Research Centre for their supervision, useful suggestions and for their indispensable help during the whole work and in the preparation of the thesis.

Grateful acknowledgement is expressed to the Mational Research Centre, for the facilities giving which made this work possible.

## CONTENTS

	Page
INTRODUCTION	1
AL! OF WORK	2
REVIEW OF LITERATURE	3
ASPHODELUS SPECIES	7
The Studied Species	9
MATERIAL	14
EXPERIMENTAL	
Chapter I	
ANTHRAQUINONES	
Anthraquinones	15
Isolation, Purification and Fractionation	17
Colour Reactions	20
Derivatives and Degradation Reactions	23
Chromatography of Anthraquinones	25
Spectra of Anthraquinones	27
Ultraviolet-Visible Spectra	27
Infrared Spectra	<b>2</b> 9
Nuclear Magnetic Resonance Spectra	33
Mass Spectra	34
Quantitative Determination of Anthraquinones.	<b>3</b> 8
Distribution and Seasonal Variation of	
Anthraquinones in Plant Parts	44
The Quinones of Family Liliaceae	50

- iii -	Page
A- Paper-Chromatography	<b>8</b> 8
B- Thin-Layer Chromatography	89
Isolation and Identification of Chrysophanol-8-	
mono-β-D-glucoside	90
Hydrolysis of the Glycoside	91
Investigation of C-glycosides	93
The Anthraquinones of Asphodelus microcarpus	94
a) Anthraquinones of the tubers of A.microcarpus.	94
Separation of the Free Anthraquinones by	
Column Chromatography	94
b) Anthraquinones of the leaves and Seeds of	
A. microcarpus	97
Two-Dimensional Thin-Layer Chromatography of the	
Anthraquinones in the Studied Asphodelus Species	97
Chapter II	
QUANTITATIVE DETERMINATION OF COUNTY COUNTY	
Quantitative Determination of the Anthraquinone	
Components in Asphodelus Species	99
Preparation of the Standard Curves	100
Procedure of the Quantitative Determination of	
Anthraquinones in Different Plant Parts	102
The Seasonal Variation of the Anthraquinones of	
tubers of Asphodelus microcarpus	106
DISCUSSION	109
SUMMARY	117
TO YOUTH WINT AND A	120

#### LIST OF TAPLES

- Table 1. Colour of hydroxyanthraquinones in alkaline solution.
- Table 2. The quinones of the family Liliaceae.
- Table 3. Properties of quinones isolated from family

  Liliaceae.
- Table 4. The properties of 1,8-dihydroxyanthraquinone and 1,3-dihydroxyanthraquinone.
- Table 5. The properties of naturally occurring bianthraquinones.
- Table 6. The R<sub>f</sub> value of the isolated anthraquinones and available authentic references.
- Table 7. The results obtained from the column chromatographic.

  fractionation of the total free anthraquinones of

  A. microcarpus.
- Table 8. The percentage of total free anthraquinones in the Egyptian Asphodelus species.
- Table 9. The absorption maxima of the anthraquinones.
- Table 10. The percentages of the anthraquinone components in the parts of Asphodelus species.
- Table 11. The percentage of anthraquinones in the tubers of

  A. microcarpus.
- Table 12. The seasonal variation of the anthraquinone component of A. microcarpus (tubers).

#### LIST OF FIGURES

- Fig. 1: Thin-layer chromatogram of total free anthraquinones of A. fistulogus.
- Fig. 2: Thin-layer chromatogram of the anthraquinone components No. VIII and IX.
- Fig. 3: Thin-layer chromatogram of the anthraquinone component No. IV and its decomposed product.
- Fig. 4: Ultraviolet spectrum of 1,8-dihydroxyanthraquinone (Isolated and Authentic).
- Fig. 5: Infrared spectrum of 1,8-dihydroxyanthraquinone (Isolated and Authentic.
- Fig. 6: N.M.R. Spectrum of 1,8-dihydroxyanthraquinone.
- Fig. 7: Mass spectrum of 1,8-dihydroxyanthraquinone.
- Fig. 8: Thin-layer chromatogram of the chrysophanol dianthraquinone and its cleavage product.
- Fig. 9: Ultraviolet spectrum of bichrysophanol.
- Fig. 10: Infrared spectrum of bichrysophanol.
- Fig. 11: Mass spectrum of bichrysophanol.
- Fig. 12: Thin-layer chromatogram of aloe-emodin (Isolated and Authentic).
- Fig. 13: Ultraviolet spectrum of aloe-emodin.
- Fig. 14: Infrared spectrum of aloe-emodin (Isolated and Authentic).

- Fig. 15: Thin-layer chromatogram of the anthraquinone component No. VII and its cleavage product.
- Pig. 16: Ultraviolet spectrum of anthraquinone component No. VII.
- Pig. 17: Infrared spectrum of anthraquinone component No. VII.
- Fig. 18: Thin-layer chromatogram of chrysophanol (Isolated and Authentic).
- Fig. 19: Ultraviolet spectrum of chrysophanol.
- Fig. 20: Infrared spectrum of chrysophanol (Isolated and Authentic).
- Fig. 21: Infrared spectrum of anthraquinone component
  No. IX.
- Fig. 22: Ultraviolet spectrum of anthraquinone component No.
- Fig. 23: Mass spectrum of anthraquinone component No. IX.
- Fig. 2: Thin-layer chromatogram of the anthraquinone component No. IX and its cleavage product.
- Fig. 25: Mass spectrum of the acetate derivative of anthraquinone component No. IX.
- Fig. 26: Thin-layer chromatogram of the acetate derivative of anthraquinone component No. IX and its cleavage product.
- Fig. 27: Thin-layer chromatogram of the anthraquinones of

  A. fistulosus.

- Pip. 28: Paper chromatography of anthraquinone alycosides of A. fistulosus.
- Fig. 29: Thin-layer chromatogram of anthrequinone glycosides of A. fistulosus.
- Fig. 30: Thin-layer chromatogram of the isolated anthraquinone glycoside and its aglycone.
- Fig. 31: Paper chromatography of the sugar hydrolysate of the anthraquinone glycoside.
- Fig. 32: Thin-layer chromatogram of the anthraqui... ones of Asphodelus species.
- Fig. 33: Two-dimensional thin-layer chromatogram of the anthraquinones of the leaves of A. fistulosus.
- Fig. 34: Two-dimensional thin-layer chromatogram of the anthraquinones of the seeds of A. fistulosus.
- Fig. 35: Two-dimensional thin-layer chromatogram of the anthraquinones of the leaves of A. microcarpus.
- Fig. 36: Two-dimensional thin-layer chromatogram of the anthraquinones of the seeds of A. microcarpus.
- Fig. 37: Two-dimensional thin-layer chromatogram of the anthraquinones of the tubers of A. microcarpus (collected in April).
- Fig. 38: Standard calibration curve of anthraquinones.
- Fig. 39: Two-dimensional thin-layer chromatogram of the anthraquinones of the tubers (collected in Januar)

#### INTRODUCTION

Most of the Egyptian flors, which comprises about 2000 species, are desert or semi-desert plants. The majority of these plants are still not yet investigated. Our attention in the Pharmaceutical Sciences Laboratory, N.R.C. is especially drawn to the flora of the desert, and in particular drug plants as well as plants of economic potentialities. Meanwhile, the study of plants liable to cause toxicity to grazing animals is always emphasized.

The plants studied, in this work, viz. Asphodelus fistulosus and A. microcarpus are widely distributed in the Egyptian desert; the former is widely distributed in the New Valley while the later grows all over the Mediterranean Costal region.

The genus Asphodelus belongs to a family rich in medecinal plants. It includes several genera which were reported (1) as remedies in folk medicine; their value in this respect has been attributed to various constituents. These constituents may be either alkaloids as colchicine in Colchicum spp. (2), anthraquinones as in Aloe spp. (3), volatile substances as in Allium spp. (4) and glycosides as in Urginea spp. (5,6), or any other constituents.

- 2 -

#### AIM OF TORK

The recent invertigation, carried out in this laboratory, on tubers of <u>Asphodelus microcargus</u>, showed that it contained a fair quantity of cuthraquinones. The fact that the other common species i.e. <u>A. fistulosus</u> was not investigated, in addition to the incomplete study of <u>A. microcarpus</u>, lead the author to carry the present work. This work deals with the following:

- 1- The study of the anthraquinone constituents of A. fistulosus.
- 2- The identification of the anthraquinone constituents (other than those reported) of A. ricrocarpus.
- 3- A qualitative and quantitative comparison of the anthraquinones in the different parts of the two species (leaf, seed, tuber).
- 4- A study of the seasonal variation of the anthraquinones of A. microcarpus (tubers) in order to select the best time of the year to collect the tubers.

# REVIEW OF LITERATURE

#### REVIEW OF LITERATURE

Klein and Pollauf (7) recroded evidence for the presence of colchicine in <u>Asphodelus albus</u> Willd, but this was later refuted by Santavy (8).

Levie and Marini (9) in their search for national resources of cellulose for paper pulp stated that

A. ramosus, on dry basis, gave cellulose 55 %. Rovesti (10) reported on the composition of A. ramosus and discussed the possibilities of producing alcohol from it.

Arrazola (11) found that hydrolysis of asphodel-inulin, the non-cellulosic polysaccharides of the tuberous root stocks of  $\underline{A}$ . albus and  $\underline{A}$ . microcarpus, gives fructose which supports the growth of the yeast <u>Torula utilis</u>.

Arrazola et al. (12) stated that the roots of Willd A. albus and A. microcarpus were saccharified, yielding a juice containing 17.18 % sugars, mostly levulose with some dextrose.

A process for alcohol production from Asphodelus tubers was reported by Guzzi and Prunotto (13).

Tappi (14) isolated, from the anthers of A. albus xanthophyll, xanthophyll ejoxids, flavoxanthin and f-carotene III in addition to other carotenoids.

Leyte (15) reported on the composition (total protein, digestible protein, fat, crude fibre, N-free extract and ash) of A. microcarpus and its possible utilization as animal food.

Khan et al. (16) studied the composition of the oil of A. fistulosus seeds. The oil contains myrestic, palmitic, steeric, oleic and linoleic; the unsaponifiable matter contains fucosterol and an unidentified yellow substance.

Van Rheede van Ouitshoorn (17) detected anthraquinones (aloe-emodin, nataloe-emodin and chrysophanol) in several genera of the <u>Filiaceae</u>, including <u>A. albus</u> which contains chrysophanol and aloe-emodin as free anthraquinones; citric and malic acids were also detected.

Sased et al. (18) reported on the amino acids composition of the proteins of A. <u>fistulosus</u> and suggested this protein as a possible human food source.

Colin and Neyro (19) found laevulosan, sucrose, a reducing sugar and an amorphous glycoside in tubers of