

# **Phytochemical and Biological Study on *Schotia brachypetalae* Family Leguminosae (Fabaceae)**

A Thesis Submitted  
In Partial Fulfillment of the Requirements  
for the Degree of Master in Pharmaceutical Sciences  
(Pharmacognosy)

By

**Esraa Ashraf Ahmed El-Hawary**  
Bachelor of Pharmaceutical Sciences,  
Faculty of Pharmacy, Ain Shams University, 2010

**Under the Supervision of**

**Abdel Nasser B. Singab Ph.D**

Professor of Pharmacognosy  
Dean of Faculty of Pharmacy  
Ain Shams University

**Nahla A. Ayoub Ph.D**

Professor of Pharmacognosy  
Faculty of Pharmacy  
Ain Shams University

**Rola M. Labib Ph.D**

Lecturer of Pharmacognosy  
Faculty of Pharmacy  
Ain Shams University

Department of Pharmacognosy  
Faculty of Pharmacy  
Ain Shams University  
Abbasia, Cairo, Egypt  
2015

**To My Father “Dr Ashraf”, My  
Mother “Dr Nagwa” and My lovely  
Husband "Eng. Mohamed"**

**To the soul of my Grandmother  
and my Father-in-law**

### **Achnowledgement**

*I would like to express my deepest gratitude, sincere and profound appreciation to the following people who significantly contributed to the work done in this thesis:*

*I am deeply grateful to **Prof. Abdel Nasser B. Singab**, Professor of Pharmacognosy, Dean of Faculty of Pharmacy, Ain Shams University, for his advice, constant guidance, helpful suggestions and encouragement throughout this work. Thanks for his precious time and for his support, valuable advices and sincere comments. For being a role model in research work, for teaching us hard work in a practical way. Thanks for being a true leader and father for all of us.*

*I am deeply grateful to **Prof. Nahla A. Ayoub**, Professor of Pharmacognosy, Faculty of Pharmacy, Ain Shams University, for suggesting the research point, valuable comments during the course of this work, for her guidance, her knowledge and her marvelous treatment of all of us. She is a warm and kind person.*

*Thanks for both **Prof. Abdel Nasser B. Singab** and **Prof. Nahla A. Ayoub** for setting an example to what a dedicated professor, scientist and advisor should be. It has been an honour serving the department and a privilege working with each of them.*

*I am deeply grateful to **Dr. Rola M. Labib**, Lecturer of Pharmacognosy, Faculty of Pharmacy, Ain Shams University, for being such a role model in every part of the work, she was a continous source of advice, support and help. She is a friendly, warm and lovely person.*

*I am grateful to **Prof. Ashraf B. Naim**, Professor of Pharmacology, Faculty of Pharmacy, Ain Shams University, **Dr. Ahmed Essmat**, Lecturer of Pharmacology, Faculty of Pharmacy, Ain Shams University and **Ebrahim Abdel-Aziz**, master student pharmacology Department, Faculty of Pharmacy, Ain Shams University for hosting the evaluation of the anti-inflammatory assays.*

## Acknowledgement

Deep thanks goes to the Pharmacology Lab. Team in Germany, headed by **Prof. Michael Wink** for hosting the anti-oxidant assays.

I am also indebted to **Dr. Heba E. Handoussa**, Lecturer of Pharmacognosy, Faculty of Pharmacy, German University in Cairo, for her constructive cooperation during the solvation of LC/MS part of the Thesis, her continuous advice and deep knowledge.

I am thankful to **Dr. Ahmed H. El-Khatib**, Laboratory of Analytical and Environmental Chemistry, Department of Chemistry, Humboldt-Universitat zu Berlin for his help in LC/MS analysis, being helpful and friendly person.

My doctors; I am very thankful for every one of them for their continuous advising as a brother and sister not as my doctor. Dr Sherwit El-Ahmady, Dr Omayma El-Dahshan, Dr Mohamed Ashour, Dr Mohamed El-Shazly, Dr Sherif Ebada, Dr Eman Kamal, Dr Haidy Gad, Dr Fadia Salah, Dr Nada Mohamed and Dr Irini Ayoub.

My colleagues; for their cooperation, support and the friendship we share, Doaa, Mohammed, Ahmed, Naglaa, Sara, Mariam, Dina, Shimaa, Nouran, Heba, Mai, Amany, Aya and Maram.

I would also like to thank **my dearest great parents Dr Ashraf El-Hawary and Dr Nagwa Shinkar who I really love and respect**, I am proud to be their daughter **and my brother, Ahmed and my sisters Aya and Asmaa**, as they suffered with me alot and for their continuous care, love and support and believing in me always; thank you very much really I love you so much.

I would like to thank also my second family **Aunt Azza, Amany and Ahmed** for their always presence, love and support.

Finally, I would like to thank my lovely husband, **Mohamed** for his encouragement, support, for being such a caring person and most of all his patience for which I am truly grateful. (وراء كل رجل عظيم امرأة... ووراء كل امرأة عظيمه رجل أعظم منها)

**Esraa Ashraf Ahmed El-Hawary**

**Cairo, 2015**

# List of Contents

|                                                                                                                               | Page |
|-------------------------------------------------------------------------------------------------------------------------------|------|
| <b>LIST OF FIGURES</b>                                                                                                        | i    |
| <b>LIST OF TABLES</b>                                                                                                         | iii  |
| <b>LIST OF ABBREVIATIONS</b>                                                                                                  | iv   |
| <b>INTRODUCTION</b>                                                                                                           | 1    |
| <b>AIM OF WORK</b>                                                                                                            | 3    |
| <b>REVIW OF LITERATURE</b>                                                                                                    |      |
| 1. Chemical review of genus <i>Schotia</i>                                                                                    | 4    |
| 2. Biological review of genus <i>Schotia</i>                                                                                  | 9    |
| <b>TAXONOMY</b>                                                                                                               | 11   |
| <b>MATERIALS, APPARATUS AND METHODS</b>                                                                                       |      |
| 1. Materials                                                                                                                  | 15   |
| 2. Apparatus                                                                                                                  | 17   |
| 3. Methods                                                                                                                    | 18   |
| <b>CHAPTER (1): Phytochemical investigation of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.</b>           |      |
| A. Phytochemical screening of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                                | 31   |
| B. Phytochemical investigation of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                            | 31   |
| Isolated Compounds from the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                                      |      |
| i. Identification of compound <b>1</b> : <b>Gallic acid</b>                                                                   | 33   |
| ii. Identification of compound <b>2</b> : <b>Myricetin-3-O-<math>\alpha</math>-L-<sup>1</sup>C<sub>4</sub>-rhamnoside</b>     | 34   |
| iii. Identification of compound <b>3</b> : <b>Quercetin-3- O-<math>\alpha</math>-L-<sup>1</sup>C<sub>4</sub>-rhamnoside</b>   | 35   |
| C. Determination of total Phenolic and Flavonoid contents of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond. | 37   |
| D. LC/ESI/MS <sup>n</sup> analysis of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                        | 37   |
| E. Discussion                                                                                                                 | 78   |
| <b>CHAPTER (2): Biological investigation of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.</b>              |      |
| A. Anti-inflammatory activity of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                             | 79   |
| B. Anti-oxidant activity of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                                  | 86   |
| C. Discussion                                                                                                                 | 92   |
| <b>CHAPTER (3): Standardization of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.</b>                       | 93   |
| <b>General summary</b>                                                                                                        | 95   |
| <b>Conclusion and Recommendation</b>                                                                                          | 99   |
| <b>References</b>                                                                                                             | 100  |
| <b>Arabic summary</b>                                                                                                         |      |

## List of Figures

| Figure                                                                                                                                                                                           | Page |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------|
| 1 Photograph of <i>Schotia brachypetala</i> Sond. (A) tree, (B) leaves                                                                                                                           | 11   |
| 2 Compound <u>1</u> Gallic acid                                                                                                                                                                  | 33   |
| 3 <sup>1</sup> H-NMR spectrum of compound <u>2</u> : Myrecitin-3- <i>O</i> - $\alpha$ -L- <sup>1</sup> C <sub>4</sub> -rhamnoside                                                                | 34   |
| 4 <sup>1</sup> H-NMR spectrum of compound <u>3</u> : Quercetin-3- <i>O</i> - $\alpha$ -L- <sup>1</sup> C <sub>4</sub> -rhamnoside                                                                | 36   |
| 5 (A) LC/ESI/MS <sup>n</sup> (-ve) spectrum of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond., showing mass to charge ratio ( <i>m/z</i> ) and retention time values           | 40   |
| (B) LC/ESI/MS (+ve) spectrum of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond., showing mass to charge ratio ( <i>m/z</i> )                                                    | 40   |
| 6 LC/ESI/MS <sup>n</sup> spectrum of Fraction (III) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond., showing mass to charge ratio ( <i>m/z</i> ) and retention time values   | 44   |
| 7 LC/ESI/MS <sup>n</sup> spectrum of Fraction (IV) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond., showing mass to charge ratio ( <i>m/z</i> ) and retention time values    | 48   |
| 8 LC/ESI/MS <sup>n</sup> spectrum of subfraction (F1) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond., showing mass to charge ratio ( <i>m/z</i> ) and retention time values | 51   |
| 9 LC/ESI/MS <sup>n</sup> spectrum of subfraction (F2) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond., showing mass to charge ratio ( <i>m/z</i> ) and retention time values | 55   |
| 10 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 253 (Daidzein)                                                                                                                                      | 63   |
| 11 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 285(Luteolin aglycone)                                                                                                                              | 63   |
| 12 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 315 (Isorhamnetin)                                                                                                                                  | 63   |
| 13 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 343 (Galloyl quinic/epiquinic acid isomer)                                                                                                          | 64   |
| 14 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 431(Kaempferol-3- <i>O</i> -rhamnoside)                                                                                                             | 64   |
| 15 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 441 [(epi) catechin gallate]                                                                                                                        | 64   |
| 16 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 447 (Quercetin-3- <i>O</i> -rhamnoside)                                                                                                             | 65   |
| 17 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 449 (Myrecitin-3- <i>O</i> - $\alpha$ -arabinopentoside)                                                                                            | 65   |
| 18 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 463 (Quercetin-3- <i>O</i> -hexoside)                                                                                                               | 66   |
| 19 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 477 (Quercetin-3- <i>O</i> -glucouronide)                                                                                                           | 66   |
| 20 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 493 ( <i>O</i> -Caffeoyl- <i>O</i> -galloyl-hexoside)                                                                                               | 66   |
| 21 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 519 (Isorhamnetin acetyl glucoside)                                                                                                                 | 67   |
| 22 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 521(Petunidin-3- <i>O</i> -acetyl glucoside)                                                                                                        | 67   |
| 23 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 567 (Phloretin xyloglucoside)                                                                                                                       | 67   |
| 24 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 583 (Kaempferol derivative)                                                                                                                         | 68   |
| 25 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 585 (Quercetin-3- <i>O</i> -(2"- <i>O</i> -galloyl)-pentoside)                                                                                      | 68   |
| 26 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 593 (Kaempferol-3- <i>O</i> -rutinoside)                                                                                                            | 69   |
| 27 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 599 (Quercetin-hexose protocatechuic acid isomer)                                                                                                   | 69   |
| 28 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 601 (Myrecitin-3- <i>O</i> -(2"- <i>O</i> galloyl)-pentoside isomer)                                                                                | 69   |
| 29 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 615 (Quercetin-3- <i>O</i> -(2"- <i>O</i> -galloyl)-hexoside isomer)                                                                                | 70   |
| 30 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 625 (Petunidin-3- <i>P</i> -coumaroyl-glucoside).                                                                                                   | 70   |
| 31 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 627 (Myrecitin-3- <i>O</i> -rutinoside)                                                                                                             | 70   |
| 32 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 635 (Trigalloyl hexose isomer)                                                                                                                      | 71   |
| 33 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 638 (Tricin-7- <i>O</i> -neohesperidoside)                                                                                                          | 71   |
| 34 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 737 (Procyanidin dimer hexoside).                                                                                                                   | 71   |
| 35 LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 751 [(epi) catechin-peonidin-3- <i>O</i> -glucoside]                                                                                                | 72   |

|    | <b>Figure (continued)</b>                                                                                                                                                                              | <b>Page</b> |
|----|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------|
| 36 | LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 850 (Procyanidin trimer)                                                                                                                                     | 72          |
| 37 | LC/ESI/MS <sup>n</sup> peak at <i>m/z</i> 631 (Myrecitin-3- <i>O</i> -(2''- <i>O</i> -galloyl)-hexoside isomer)                                                                                        | 72          |
| 38 | (A) (B) (C) (D) (E) Compounds identified from LC/ESI/MS <sup>n</sup> spectra of <i>Schotia brachypetala</i> Sond. methanolic leaf extract, fraction(III), fraction(IV) and subfractions F1 and F2      | 73          |
| 39 | Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on the level of PGE <sub>2</sub> in carrageenan-induced rat paw edema model                                        | 81          |
| 40 | Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on the ear disc weight in croton oil-induced rat ear edema model                                                   | 83          |
| 41 | Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on MPO activity in croton oil-induced rat ear edema model                                                          | 83          |
| 42 | Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on TNF- $\alpha$ level in croton oil-induced rat ear edema model                                                   | 84          |
| 43 | Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. on histopathological changes in the croton oil-induced ear edema experiment                                                     | 85          |
| 44 | <i>C. elegans</i> stress resistance under juglone treatment                                                                                                                                            | 87          |
| 45 | Effect <i>Schotia brachypetala</i> Sond. methanolic leaf extract on ROS levels in <i>C. elegans</i> using DCFD assay                                                                                   | 88          |
| 46 | Micrographs show a representative worm treated with the extract 50 $\mu$ g/ml (B), 100 $\mu$ g/ml (C), 200 $\mu$ g/ml (D) and a representative worm from the control group (E)                         | 88          |
| 47 | Effect of <i>Schotia brachypetala</i> Sond. extract on <i>hsp16.2::GFP</i> expression in transgenic <i>C. elegans</i> strain (TJ375 <i>hsp 16.2::GFP(gplsI)</i> under juglone-induced oxidative stress | 89          |
| 48 | Micrographs show a representative worm treated with three different concentrations 50 $\mu$ g/ml (B), 100 $\mu$ g/ml (C), 150 $\mu$ g/ml (D) and a representative worm from the control group (E)      | 90          |
| 49 | Effect of <i>Schotia brachypetala</i> extract on DAF-16 subcellular localization in transgenic <i>C. elegans</i> strain (TJ356)                                                                        | 91          |
| 50 | Micrographs show a representative worm for cytosolic (B), intermediate (C) and nuclear (D) pattern                                                                                                     | 91          |
| 51 | Standard calibration curve of gallic acid                                                                                                                                                              | 93          |

## List of Tables

| Table                                                                                                                                                                                                          | Page |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------|
| 1 Chemical structure, names and occurrence of phytoconstituents of genus <i>Schotia</i>                                                                                                                        | 5    |
| 2 Results of phytochemical screening of methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                                                                                                           | 31   |
| 3 Results of column chromatographic fractionation of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond.                                                                                          | 32   |
| 4 Results of column chromatographic analysis of Fraction III                                                                                                                                                   | 33   |
| 5 Chromatographic and spectral data for compound <u>2</u>                                                                                                                                                      | 34   |
| 6 Chromatographic and spectral data for compound <u>3</u>                                                                                                                                                      | 35   |
| 7 Tentative identification of compounds detected in the methanolic leaf extract of <i>Schotia brachypetala</i> Sond. by LC/ESI (-ve)/MS <sup>n</sup> and LC/ESI (+ve)/MS                                       | 38   |
| 8 Tentative identification of compounds detected in fraction (III) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond. by LC/ESI (-ve)/MS <sup>n</sup> and LC/ESI (+ve)/MS                     | 42   |
| 9 Tentative identification of compounds detected in fraction (IV) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond. by LC/ESI (-ve)/MS <sup>n</sup> and LC/ESI (+ve)/MS                      | 46   |
| 10 Tentative identification of compounds detected in subfraction (F1) of fraction (IV) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond. by LC/ESI (-ve)/MS <sup>n</sup> and LC/ESI (+ve)/MS | 49   |
| 11 Tentative identification of compounds detected in subfraction (F2) of fraction (IV) of the methanolic leaf extract of <i>Schotia brachypetala</i> Sond. by LC/ESI (-ve)/MS <sup>n</sup> and LC/ESI (+ve)/MS | 53   |
| 12 Summary of the identified compounds by LC/ESI/MS <sup>n</sup>                                                                                                                                               | 57   |
| 13 Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on rat paw volume in carrageenan-induced rat paw edema model                                                            | 80   |
| 14 Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on the level of PGE <sub>2</sub> in carrageenan-induced rat paw edema model                                             | 80   |
| 15 Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on rat disc weight in croton oil-induced rat ear edema model                                                            | 82   |
| 16 Effect of methanolic leaf extract of <i>Schotia brachypetala</i> Sond. (SB extract) on MPO activity in croton oil-induced rat ear edema model                                                               | 83   |
| 17 <i>In vitro</i> antioxidant activity of <i>Schotia brachypetala</i> Sond. methanolic leaf extract                                                                                                           | 86   |
| 18 The antioxidant effect of <i>Schotia brachypetala</i> Sond. extract against ROS formation <i>in vivo</i>                                                                                                    | 88   |
| 19 The effect of <i>Schotia brachypetala</i> Sond. extract on <i>hsp16-2::GFP</i> expression                                                                                                                   | 90   |

## List of Abbreviations

|                        |                                                                       |
|------------------------|-----------------------------------------------------------------------|
| ABTS <sup>+</sup>      | [2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)]              |
| CC                     | column chromatography                                                 |
| cm                     | centimeter                                                            |
| CMC-Na                 | carboxymethyl cellulose sodium                                        |
| 2D                     | two dimensional                                                       |
| Da                     | dalton                                                                |
| DCF                    | 2', 7'- dichlorofluorescein                                           |
| DCFDA                  | 2', 7'- dichlorofluorescein diacetate                                 |
| DMSO                   | dimethyl sulfoxide                                                    |
| Dil                    | diluted                                                               |
| DPPH <sup>•</sup>      | (2, 2-diphenyl-1-picrylhydrazyl)                                      |
| DW                     | dry weight                                                            |
| ELISA                  | enzyme-linked immunosorbent assay                                     |
| ESI/LC/MS <sup>n</sup> | electrospray ionization- liquid chromatography-mass–mass spectrometry |
| g                      | gram                                                                  |
| GFP                    | green fluorescence protein                                            |
| <sup>1</sup> H-NMR     | proton nuclear magnetic resonance                                     |
| HPLC                   | high performance liquid chromatography                                |
| hsp                    | heat shock protein                                                    |
| Hz                     | hertz                                                                 |
| <i>J</i> value         | coupling constant                                                     |
| Kg                     | kilogram                                                              |
| L                      | liter                                                                 |
| <i>m/z</i>             | mass to charge ratio                                                  |
| mg                     | milligram                                                             |
| MPO                    | myeloperoxidase                                                       |
| NSAID                  | non-steroidal anti-inflammatory drugs                                 |
| PC                     | paper chromatography                                                  |
| PGE <sub>2</sub>       | prostaglandin E <sub>2</sub>                                          |
| PPC                    | preparative paper chromatography                                      |
| ppm                    | part per million                                                      |
| R <sup>2</sup>         | regression coefficient                                                |
| RBCs                   | red blood cells                                                       |
| ROS                    | reactive oxygen species                                               |
| RSD                    | relative standard deviation                                           |
| R <sub>t</sub>         | retention time                                                        |
| S.D.                   | standard deviation                                                    |
| TNF- $\alpha$          | tumor necrosis factor $\alpha$                                        |
| UV                     | ultraviolet                                                           |
| $\delta$               | chemical shift                                                        |
| $\lambda$              | wave length                                                           |
| $\mu$ l                | microliter                                                            |

# Introduction

### Introduction

During the last few decades, there has been an increasingly intensified search for biologically active compounds of natural origin. Over this period, a worldwide interest in drugs derived from plants has been developed to produce a daily increase of public demand of these drugs for treatment of broad range of diseases. Many of these drugs possess multiple biological effects as anticancer, antiviral, antihyperglycemic or immuno-modulating agents (Dahanukar *et al.* 2000).

Phenolic compounds are found in both edible and non-edible plants and they have been reported to have a lot of biological effects. Crude plant extracts rich in phenolics are of increasing interest for biological and phytochemical investigations. Flavonoids and other phenolics have been suggested to play a preventive role in cancer development and heart diseases (Dahanukar *et al.* 2000).

Among the famous plant families which include genera that embraces phenolic-rich species, Leguminosae (Fabaceae) occupies a distinguishable situation. This is due to the fact that many of its plants are capable of synthesizing and accumulating high percentage of phenolics. Fabaceae is a large family, distributed in tropical and subtropical regions. The family is considered the second largest family among the flowering plants, behind Asteraceae, with 730 genera and over 19,400 species. It is a large family of trees, shrubs and herbs bearing bean pods distributed throughout the world.

Leguminosae is one of the most important families from an economic point of view. The seeds are rich in starch and protein, used as widespread source of food as in various beans (Rendle 1959).

Legumes, broadly defined by their unusual flower structure, podded fruit, and the ability of 88% of the species examined to date to form nodules with rhizobia. In addition to traditional food and forage uses, legumes can be milled into flour, used to make bread, doughnuts, tortillas, chips, spreads, and extruded snacks or used in liquid form to produce milks, yogurt, and infant formula. Pop beans, licorice (*Glycyrrhiza glabra*), and soybean candy provide novel uses for specific legumes. Legumes have been used industrially to prepare biodegradable plastics, oils, gums, dyes, and inks. Galactomannan gums derived from *Cyamopsis* spp. and *Sesbania* spp. are used in sizing textiles and paper, as a thickener, and in pill formulation (Graham and Vance 2003).

Many legumes have been used in folk medicine. Isoflavones from soybeans and other legumes have more recently been suggested both to reduce the risks of cancer and to lower serum cholesterol. Soybean and soyfood phytoestrogens have been suggested as possible alternatives to hormone replacement therapy for postmenopausal women (Graham and Vance 2003).

*Schotia brachypetala* Sond. tree is indigenous to South Africa (Watt and Breyer-Brandwijk 1932, Brenan 1967). *Schotia* was named after Richard Schott and *brachypetala* means having short petals in Greek and refers to the flowers which are unique among *Schotia* species in that the petals are partially or completely reduced to linear filaments. The beauty of the flower is in brightly coloured sepals, stamens and pedicels. The flower produces copious amounts of nectar which overflows and drips or weeps (Germishuizen *et al* 2006).

Traditionally, the bark decoction was used to strengthen the body, treat dysentery and diarrhea, nervous and heart conditions, flu symptoms and an emetic in excessive beer drinking, the bark is also used for dyeing giving reddish brown or red colour. Its roots were also used to treat diarrhea and heartburn. The seeds are edible after roasting (Du *et al.* 2014). The timber is of good quality suitable for furniture making and flooring blocks and said to be excellent for all kinds of wagon wood (Germishuizen *et al* 2006).

# Aim of Work

### Aim of Work

To the best of our knowledge, there was little information concerning *Schotia brachypetala* Sond., cultivated in Egypt. It was therefore, found interesting to subject the extract of the leaves of entitled plant to an intensive biological and phytochemical investigations.

Pilot study on the methanolic leaf extract of *Schotia brachypetala* Sond. using 2D paper chromatography showed a complicated mixture of polyphenolics, this directed the present study to investigate in-depth the phenolics of the leaf extract. Besides evaluation of the biological activity of the extract by determining its anti-inflammatory and anti-oxidant activities. Our work strategy included:

1. Phytochemical investigation of the methanolic leaf extract of *Schotia brachypetala* Sond. The study included: phytochemical screening and phytochemical investigation of the methanolic leaf extract in an effort to fractionate and isolate phenolic compounds. Identification of the isolated compounds using spectroscopic methods e.g. UV,  $^1\text{H-NMR}$  and LC/ESI/MS<sup>n</sup> spectrometry. Also, it will enable the correlation between the biological activity and the chemical constituents whenever possible. Together with determination of the total phenolic and flavonoidal content of the methanolic leaf extract of *Schotia brachypetala* Sond.
2. Biological investigation of the methanolic leaf extract of *Schotia brachypetala* Sond. It included: screening for the anti-inflammatory activity in two experimental models, also screening for the anti-oxidant activity in different *in-vitro* and *in-vivo* models.
3. Standardization of the methanolic leaf extract of *Schotia brachypetala* Sond. using HPLC analysis.