سامية محمد مصطفى

شبكة المعلومات الجامعية

بسم الله الرحمن الرحيم

سامية محمد مصطفى

شبكة المعلومات الجامعية

شبكة المعلومات الجامعية

التوثيق الالكتروني والميكروفيلم

شبكة المعلومات الجامعية

سامية محمد مصطفى

جامعة عين شمس

التوثيق الإلكتروني والميكروفيلم

قسم

نقسم بالله العظيم أن المادة التي تم توثيقها وتسجيلها علي هذه الأقراص المدمجة قد أعدت دون أية تغيرات

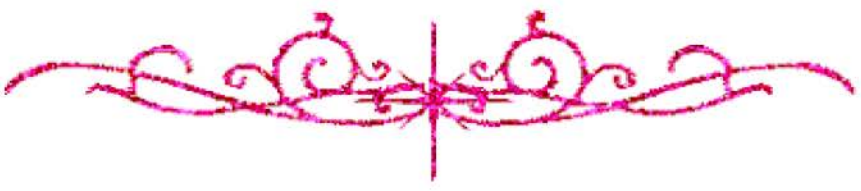
يجب أن

تحفظ هذه الأقراص المدمجة بعيدا عن الغبار

سامية محمد مصطفى

شبكة المعلومات الجامعية

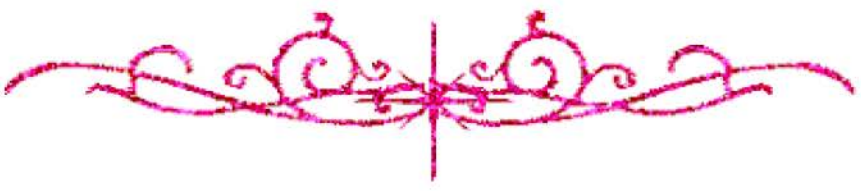
بعض الوثائق الأصلية تالفة



سامية محمد مصطفى

شبكة المعلومات الجامعية

بعض الوثائق الأصلية تالفة



سامية محمد مصطفى

شبكة المعلومات الجامعية

بالرسالة صفحات لم ترد بالأصل

BIOTECHNOLOGICAL STUDIES ON MEDICINAL PLANT *PLECTRANTHUS BARBATUS* ANDREWS

By

DINA SAYED MOHAMED MAHMOUD

B.Sc., Agric. Sc. (Biotechnology), Cairo Univ. (2012)

A Thesis Submitted in Partial Fulfillment

Of

The Requirements for the Degree of

MASTER OF SCIENCE
in
Agricultural Sciences
(Genetics)

Department of Genetics

Faculty of Agriculture

Ain-Shams University

2019

Approval Sheet

BIOTECHNOLOGICAL STUDIES ON MEDICINAL PLANT *PLECTRANTHUS BARBATUS* ANDREWS

By

DINA SAYED MOHAMED MAHMOUD

B.Sc., Agric. Sc. (Biotechnology), Cairo Univ. (2012)

This Thesis for Master Degree has been approved by:

Dr. Magda Ali Mahmoud El-Enany .....

Associate Researcher Professor of Genetics, National Research Center, Giza, Egypt.

Dr. Mohamed Abdel-Salam Rashed .....

Professor Emeritus of Genetics, Faculty of Agriculture, Ain Shams University

Dr. Lamyaa Mostafa Kamal Sayed .....

Associate Professor of Genetics, Faculty of Agriculture, Ain Shams University

Dr. Eman Mahmoud Fahmy .....

Professor Emeritus of Genetics, Faculty of Agriculture, Ain Shams University

Date of Examination: 20 / 2 / 2019

BIOTECHNOLOGICAL STUDIES ON MEDICINAL PLANT *PLECTRANTHUS BARBATUS* ANDREWS

By

DINA SAYED MOHAMED MAHMOUD

B. Sc., Agric. Sc. (Biotechnology), Cairo Univ. (2012)

Under the supervision of:

Dr. Eman Mahmoud Fahmy

Professor Emeritus of Genetics, Department of Genetics, Faculty of Agriculture, Ain Shams University (principal supervisor).

Dr. Lamyaa Mostafa Kamal Sayed

Associate Professor of Genetics, Department of Genetics, Faculty of Agriculture, Ain Shams University.

Dr. Mohamed Ismail Gamal Elden Diab

Associate Researcher Professor of Biotechnology, Tissue Culture Unit, Department of Plant Genetic Resources, Desert Research Center .

ABSTRACT

Dina Sayed Mohamed: Biotechnological studies on medicinal plant *Plectranthus barbatus* Andrews. Unpublished M.Sc. Thesis, Department of Genetics, Faculty of Agriculture, Ain Shams University, 2019

This study aimed at introducing the new species *Plectranthus barbatuse* Andrews (*Coleus forsckoli*) belonging to family: *Lamiaceae* to the Egyptian flora for its medicinal importance due to the presence of active ingredient forskolin. The conservation of *P.barbatuse* through *in vitro* micro-propagation of stem nods showed that the two cytokinin TDZ and Kin gave the best number of roots and shoot length, respectively. Production of forskolin using callus culture was inducted in both leaf and stem node explants with 2,4-D auxin, while negative results appeared in the control. For the extraction and quantitative estimation of forskolin by HPLC, the forskolin content in leaf callus on MS medium containing 2mg/L 2,4-D recorded the highest value (467.76 mg/g) in the fifth subculture. Moreover, the highest forskolin content in stem callus on MS medium containing 0.5mg/L 2,4-D was 751.96 mg/g in the fifth subculture. Using elicitor feeding in leaf callus gave the best forskolin content with sucrose or casine, while in stem callus, casine only gave the best forskolin content. The relative gene expression analysis using quantitative real time-PCR (qRT-PCR) in the previous stage was detected by using primer pairs for three *Cytochrome 450 genes*. *P450c4* and *P450c6* showed up-regulated expression in stem callus than leaf callus, while *P450c7* gave up-regulated expression in leaf callus than stem callus compared with the control (mother plant). These results confirmed the importance of Cytochrome *P450* genes expression in stem and leaf.

Keywords: *Plectranthus barbatuse* Andrews, Forskolin, Micropropagation, Callus culture, HPLC, Gene expression, Quantitative real time (RT-PCR), *CytochromeP 450* genes

ACKNOWLEDGEMENT

First and foremost thanks to "Allah", the most merciful for guiding me and giving me the strength to complete this work.

I would like to express my deep gratitude to **Professor Dr. Eman M. Fahmy**, Professor Emeritus of Genetics, Faculty of Agriculture, Ain Shams University for her supervision guidance, care, sincere help, constructive criticism, great encouragement and review of the manuscript.

I would like also to express my deepest gratitude and sincere thanks to **Dr. Lamyaa Mostafa Kamal Sayed,** Associate Professor of Genetic, Faculty of Agriculture, Ain Shams University for her continuous supervision, constructive suggestions, supplying me with all the necessary facilities and review of the manuscript.

My sincere appreciation to **Dr. Mohamed Ismail Gamal Elden Diab**, Associate Researcher Professor of Biotechnology, Tissue Culture Unit, Department of Plant Genetic Resources, Desert Research Center for his kind support, valuable supervision, suggesting the problem, kind guidance, great help and review of the manuscript.

Great appreciation is also extended to all the staff members of Plant Genetic Resources Department, Desert Research Center and of Genetics Department, Faculty of Agriculture, Ain Shams University.

CONTENTS

LIST OF TABLES