بسم الله الرحمن الرحيم

MONA MAGHRABY

شبكة المعلومات الجامعية

التوثيق الالكتروني والميكروفيلم

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شبكة المعلومات الجامعية

التوثيق الإلكتروني والميكروفيلم

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

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تحفظ هذه الأقراص المدمجة بعيدا عن الغبار

MONA MAGHRABY

BIOCHEMICAL STUDIES ON NATURAL COMPONENTS AS ANTI-TOXIGENIC AGENTS

By

ALAA ALI KHALIL ALI OMAR

B.Sc. Agric. Sci. (Biochemistry), Fac. Agric., Zagazig Univ., 2009
M.Sc. Agric. Sci. (Biochemistry), Fac. Agric., Cairo Univ., 2015

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APPROVAL COMMITTEE

Dr. NAGAH EL-SHAHAT ALI ……..
Professor of Biochemistry, Fac. Agric., Ain Shams University

Dr. FOUAD ABDEL-REHIM AHMED ……..…
Professor of Biochemistry, Fac. Agric., Cairo University

Dr. SHERIF HELMY AHMED ……...
Professor of Biochemistry, Fac. Agric., Cairo University

Dr. YASMIN EMAM ABDEL-MOBDY ……
Associate Professor of Pesticides Toxicology, Fac. Agric., Cairo University

Date: 17 / 8 / 2021

SUPERVISION SHEET

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SUPERVISION COMMITTEE

Dr. SHERIF HELMY AHMED
Professor of Biochemistry, Fac. Agric., Cairo University

Dr. YASMIN EMAM ABDEL-MOBDY
Associate Professor of Pesticides Toxicology, Fac. Agric., Cairo University

Dr. HASSAN AHMED EL-SAYED AMRA (LATE)
Researcher Professor of Mycotoxins, Food Toxicology and Contaminants, NRC, Giza

Name of Candidate: Alaa Ali Khalil Ali Omar **Degree:** Ph.D

Title of Thesis: Biochemical Studies on natural components as anti-toxigenic agents

Supervisors: Dr. Sherif Helmy Ahmed
Dr. Yasmin Emam Abdel-Mobdy
Dr. Hassan Ahmed Amra

Department: Biochemistry **Branch:** Biochemistry

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ABSTRACT

Ziziphus spina-christi leaves (ZSC), Rhamnaceae, and *Euphorbia umbellate* latex (EU), Euphorbiaceae, considered being of the common potent natural. So, the aim of study was to carry out a bio-monitored investigation of extracts using *in vitro* assay.

Well-developed nanostructure incorporated with these extracts into a unique form with greater biocompatibility was investigated. The study demonstrated a combination of polysaccharide-based nanoparticles with extracts into a unique form which prepared by using two biocompatible polymers; alginate (ALG) and chitosan (CS). To investigate potential interactions between ALG/CS-NPs and the extract (ZSC/EU), physicochemical characterization was performed. The release profiles of phenolic compounds from ZSC/EU loaded ALG/CS-NPs were evaluated using quantification method.

ZSC/EU loaded ALG/CS-NPs was developed as a potent immunomodulatory natural extract with promising antimicrobial activity against multidrug resistant bacteria, antiviral activity against Coxsackie B3 virus, antioxidant activity against free radicals and oxidative stress, anti-inflammatory activity by inhibitory effect on lysosomal enzymatic activities and anti-proliferation activity against DNA oxidative damage (hepatocellular carcinoma cell line.

The results indicated that the novel agent has potent antioxidant activities and potent inhibitory effect on lysosomal enzymatic activities. Also, the novel agent had inhibitory effect; it caused cytotoxic effect by IC_{50} of 14.28 and 12.66 µg/ml for hepatocellular carcinoma cell line (HepG2) and breast adenocarcinoma cell line (MCF-7), respectively.

It was concluded that the novel agent had been reported to afford protection against oxidative stress due to their significant anti-inflammatory, antioxidant, antimicrobial, antiviral and anti-proliferation activities.

Kay words: *Ziziphus spina-christi*, *Euphorbia umbellate* (Pax) Bruyns, phenolic, flavonoids, Polysaccharide-Based Nanoparticles, polymers, Nanoformulations, anti-inflammatory activity, anti-proliferation activity, DPPH, MTT.

DEDICATION

I dedicate this work to whom my heartfelt thanks; to my grandfather; Khalil (late), my father; Ali (late) and my mother; Naeema (late), as well as to my brothers; Ragab, Mohamed, Omar, Khalil and El-Sayed, and my sister; Haniya for their patience, help and all the supports they lovely offered along the period of my post-graduation and I can't find adequate words to express my feeling towards them, so does my supervisor Prof. Dr. Hassan Amra (late). Also, I wish to express my feeling to my friend; Mohamed (late) and other friends for encouragement.

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Alaa Ali Khalil Ali Omar

ABBREVIATIONS, EXPRESSIONS AND SYMBOLS

AA	Antioxidant activity or ascorbic acid
ACP	Acid phosphatase
ALG	Alginate
Amox	Amoxicillin
ANOVA	Analysis of variance
ANT	Adenine nucleotide translocator
BHA	Butylated hydroxyanisole
BHT	Butylated hydroxytoluene
CC_{50}	Cytotoxic concentration 50
CNS	Central nervous system
CPE	Cytopathic effect
CS	Chitosan
CFIDS	Chronic fatigue immunodeficiency syndrome
DA	Degree of acetylation
DDT	Dammarane-type triterpenoids
DEMC	Diethylmethyl chitosan
DMEC	Dimethylethyl chitosan
DMEM	Dulbecco's Modified Eagle's Medium
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic Acid
DOX	Doxorubicin
DPPH	2, 2-diphenyl-1-picrylhydrazyl or 1, 1-diphenyl-2- picryl- hydrazyl
DTT	Dithiothreitol
EDTA	Ethylene di-amine tetra acetic acid
EI	Electron ionisation
ELISA	Enzyme-linked immunosorbent assay
EU	*Euphorbia umbellate*
FA	Fraction of acetylation
FBS	Fetal bovine serum
FRAP	Ferric reducing antioxidant power
FT-IR	Fourier-transform infrared spectroscopy
G	α-L-gluronic acid
GAE	Gallic acid equivalent
β-GAL	β-galactosidase
GAE	Gallic acid equivalent
GC/MS	Gas Chromatography–Mass Spectrometry
GIT	Gastrointestinal tract
β-GLU	β-glucuronidase

HBSS	Hanks´ Balanced Salt Solution
HP	Heliobacter pyroli
HPLC	High performance liquid chromatograph
HRP	Horseradish Peroxidase
HRS	Hydroxyl radical scavenging activity
ICAD	Inhibitor of caspase-activated DNase
LD_{50}	Lethal dose
M	β-(1-4)-D-mannuronic
MDA	Malondialdehyde
mETC	Mitochondrial electron transport chain
MIC	Minimum inhibitory concentration
MMP	Mitochondrial membrane potential
MPS	Myofascial pain syndrome
mtROS	Mitochondria ROS
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide
MW	Molecular weight
$NADP^+$	Nicotinamide adenine dinucleotide phosphate
β-NAG	N-Acetyl-β-glucosaminidase
ND	Not detected
NMR	Nuclear Magnetic Resonance
NO	Nitric oxide
NPDDs	Nanoparticulate drug delivery system
NPs	Nanoparticles
NRF2	Nuclear factor
OD	The optical density
8-OHdG	8-hydroxy-2-deoxyguanosine
OFRs	Oxygen free radicals
OS	Oxidative stress
PA	Pattern of acetylation
PBP	The penicillin binding protein
PBS	Phosphate buffer saline
PEC	Polyelectrolyte complex
PEO	Poly ethyl oxide
PDA	Potato Dextrose Agar
PM	Physical mixture
PTs	Pentacyclic triterpenes
PUFAs	Polyunsaturated fatty acids
PVA	Poly vinyl alcohol
PVP	Poly vinyl pyrrolidone
QE	Quercetin equivalents
RDA	The Recommended Dietary Allowance
RNS	Reactive nitrogen species

ROS	Reactive oxygen species
RSA	Radical scavenging activity
RT	Retention time
SBs	Strand breaks
SCGE	Single cell gel electrophoresis
SD	Standard deviation
SEM	Scanning electron microscope
SMs	Secondary metabolites
SPSS	Statistical package for social sciences
STPP	Sodium tripolyphosphate
TAC	Total antioxidant capacity
TAE	Tannic acid equivalent
TBA	Thiobarbituric acid
TBARS	Thiobarbituric acid reactive substances
TCA	Trichloroacetic acid
$TCID_{50}$	Tissue Culture Infectious Dose
TEC	Triethyl chitosan
TEM	Transmission electron microscopy
TFC	Total flavonoids content
TLC	Thin-layer chromatography
TMC	N-trimethyl chitosan chloride
TMS	Tetramethylsilane
TNF- α	Ttumor necrosis factor alpha
TNF- β	Ttumor necrosis factor beta
TP	Total proteins
TPC	Total phenolic compounds
UV	Ultraviolet
VDAC	Voltage-dependent anion channel
WHO	World health organization
XRD	X-ray diffraction
ZSC	*Zizyphus spina-cristi*

CONTENTS

CONTENTS (continued)