شبكة المعلومات الجامعية

التوثيق الإلكتروني والميكروفيلم

بسم الله الرحمن الرحيم

HANAA ALY

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جامعة عين شمس

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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

HANAA ALY

Faculty of Women for Arts, Science and Education

Biochemistry and Nutrition Department

Comparative Study between Saffron *(Crocus sativus L.)* and / or Turmeric *(Curcuma longa L.)* Extracts on D-galactose Deleterious Brain Effects in Rats.

Thesis

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قَالَ تَعَالَىٰ: ﴿وَقُلِ ٱعۡمَلُواْ فَسَيَرَى ٱللَّهُ عَمَلَكُمۡ
وَرَسُولُهُۥ وَٱلۡمُؤۡمِنُونَۖ وَسَتُرَدُّونَ إِلَىٰ عَٰلِمِ ٱلۡغَيۡبِ
وَٱلشَّهَٰدَةِ فَيُنَبِّئُكُم بِمَا كُنتُمۡ تَعۡمَلُونَ ١٠٥﴾

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Dedication

*This work is dedicated for **the soul of my grandmother,** may God bless her soul.*

*Also, I dedicate this work to my parents; who have raised me to be the person I am today. **My "father"** did not only raise and nurture me but also taxed himself dearly over the years for my education and intellectual development. **My "mother"** has been a source of motivation and strength during all hard moments. Thanks for their love, guidance and support that you have always given me.*

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Abstract

Saffron stigmas *(Crocus sativus L.)* and turmeric rhizomes (*Curcuma longa L.)* extracts are rich sources of active components with high medicinal value, so that this study designed to investigate the active chemical constituents and antioxidant capacities of saffron stigmas and turmeric rhizomes extracts (ESE and ETE), respectively. D- galactose deleterious brain effects as well as the role of ESE and ETE supplementation against D-galactose intoxication were evaluated on male rat's brain. Fifty-Five adult male Sprague-Dawley rats were divided into 5 groups; 10 rats each except D-galactose group that composed of 15 rats. Group (1) HCG: Healthy control group, rats were given distilled water orally and injected with normal saline subcutaneously (S.C) daily. Group (2) DGCG: D-galactose control group, rats were given distilled water daily orally and injected with D-galactose (250 mg/kg /day/S.C).; group (3-5): D-galactose intoxicated rats and supplemented with (30mg /kg body weight /daily orally) of ESE, ETE and (15mg /kg body weight /daily orally) from each extract, respectively for six weeks. Research results revealed that saffron and turmeric ethanolic extracts contain active chemical constituents including polyphenols and flavonoids that possess high antioxidant activity. Biochemical analysis of brain tissues documented that injection with D-galactose caused significant increase ($P \leq 0.05$) in oxidative stress parameters including [advanced glycation end products (AGEs), protein carbonyl group (PCG), malondialdehyde (MDA) and nitric oxide (NO) Levels], pro-inflammatory markers like [tumor necrosis factor alpha (TNF-α) and interleukin -6 (IL-6) levels] , epigenetic marker [$p16^{INK4a}$ content] as well as neural cell markers [metallothoenins (MTs) and serotonin (5-HT) levels].On the other hand D-galactose intoxication caused significant decrease ($P \leq 0.05$) in brain antioxidants as [total antioxidant capacity (TAC), reduced glutathione (GSH) level and catalase (CAT) activity] as well as brain acetylcholinesterase (AChE) activity. All these results were proved by the microscopic examination and immunohistochemical analysis of apoptotic and antiapoptotic

markers in brain tissues that revealed degenerative changes in cerebral cortex and hippocampus.

Oral administration of saffron and turmeric extracts alone or in combination decreased brain oxidants, pro-inflammatory markers, epigenetic marker and neural cell markers levels while increased the levels and activities of antioxidants as well as AChE activity associated with an improvement of brain microscopic examination and immunohistochemical analysis. The most significant improvements ($P \leq 0.05$) were recorded in the group that supplemented with both extracts. Study results proved that saffron and turmeric extracts active components were able to correct deleterious brain effects induced by D-galactose and using their mixture was more efficient in ameliorating brain toxicity than using each extract alone evidenced by biochemical analysis, microscopic examination as well as immunohistochemical determination of apoptotic and antiapoptotic markers in brain tissues.

It is advised to add saffron and turmeric to human foods and to prepare their extracts to be available for human beings due to their ability to preserve brain functions and structure as well as their potential to inhibit and retard brain aging and neuro-degeneration.

List of Abbreviations

Abbreviation	Full term
3-NP	Nitropropionic acid
4-AP	4 aminophenazone
5-HT	5-hydroxytryptamine
5-LOX	5- lipooxgenase
8-OHdG	8-hydroxy-2-deoxyguanosine
AChE	Acetyl choline esterase
ACR	Acrylamide
AD	Alzheimer's disease
AGEs	Advanced glycation end products
$AlCl_3$	Aluminum chloride
ALP	Alkaline Phosphatase
ALT	Alanine aminotransferase
AMPK	5' AMP-activated protein kinase
ANOVA	Analysis of variance
APAP	Acetaminophen
AST	Aspartate aminotransferase
ATP	Adenosine triphosphate
Aβ	Amyloid β-peptide
Bax	Bcl -2 associated X protein
BBB	Blood brain barrier
Bcl -2	B-cell lymphoma 2
BDMC	Bisdemethoxycurcumin
BMI	Body mass index
BUN	Blood urea nitrogen
CA1	Current area 1
CA2	Carbonic anhydrase 2
CAT	Catalase
PCG	Protein carbonyl group
CCl_4	Carbon tetrachloride
Cd	Cadmium
CD4	Cluster of differentiation 4

CDNA	Complementary DNA
CE	Catechin equivalent
CHD	Coronary heart disease
CK-MB	Creatine kinase-MB
CNS	Central nervous system
COX-2	Cyclooxygenase-2
CPA	Cyclophosphamide
Cr	Creatinine
CUR	Curcumin
Cys-C	Cystatin C
Cyt C	Cytochrome C
D.H_2O	Distilled water
Da	Dalton
DA	Dopamine
DAB	Diaminobenzidine tetrachloride
DEN	Diethyl nitrosamine
DEPC	Diethylpyrocarbonate
D-Gal	D-galactose
DHBS	3, 5-Dichloro -2-hydroxybenzene sulfonic acid
DM	Diabetes mellitus
DMC	Demethoxycurcumin
DNA	Deoxyribonucleic acid
DPPH	1, 1-diphenyl-2-picryl hydrazyl
DTNB	5, 5` dithiobis (2 - nitrobenzoic acid)
EDTA	Ethylenediaminetetraacetic acid
ELISA	Enzyme linked immunosorbent assay
ERK	Extracellular-regulated kinase
ESE	Ethanolic saffron extract
ETC	Electron transport chain
ETE	Ethanolic turmeric extract
FAβ	β-amyloid fibrils
Ft	Feet tall
GAE	Gallic acid equivalent

GGT	Gamma-glutamyl transferase
GLUT2	Glucose transporter 2
GLUT3	Glucose transporter 3
GLUT4	Glucose transporter 4
GPx	Glutathione peroxidase
GSH	Reduced glutathione
GST	Glutathione-S-transferase
H&E	Hematoxylin and Eosin
H_2O_2	Hydrogen peroxide
Hb	Hemoglobin
HbA1c	Hemoglobin A1c
HLD	High lipid diet
HO-1	Heme oxygenase-1
HPLC	High Performance Liquid Chromatography
HRP	Horseradish Peroxidase
Hyp	Hydroxyproline
ICV	Intra-cerebroventricular
IL-6	Interleukin 6
iNOS	Inducible nitric oxide synthase
LD50	Lethal dose
LDH	Lactate dehydrogenase
LDL	Low-density lipoprotein
Li_2CO_3	Lithium carbonate
LSD	Least significant difference
MAO	Monoamine oxidase
MCAO	Middle cerebral artery occlusion
MDA	Malondialdehyde
MES	Maximum electroshock
MI	Myocardial infarction
MMLV-RT	Moloney Murine Leukemia Virus Reverse Transcriptase
MS	Multiple sclerosis
MTs	Metallothenesis
Na_2CO_3	Sodium carbonate

NADPH	Nicotinamide adenine dinucleotide phosphate
NEDA	N –(1 – naphthyl) ethylenediamine
NF-kB	Nuclear factor – Kb
NFT	Neurofibrillary tangles
NMDAR	N-Methyl- D-aspartate receptor
NO	Nitric oxide
NO_2	Nitrogen dioxide
NOs	Nitric oxide synthase
NRC	National Research Council
NSAIDs	Non-steroidal anti-inflammatory drugs
O_2^-	Superoxide anion
OD	Optical density
OH^-	Hydroxide ions
$ONOO_2$	Peroxy nitrite
P300-HAT	Histone acetyltransferase p300
PARP-1	Poly ADP ribose polymerase
PBS	Phosphate buffered saline
PCOS	Polycystic ovary syndrome
PD	Parkinson's disease
PGE2	Prostaglandin E2
PGI2	Prostacyclin I2
PKB	Phosphorylase Protein kinase B
PPAR	Peroximose proliferator– activated receptor
PPm	Parts per million
PTZ	Pentylene tetrazol
PUFAs	Polyunsaturated fatty acids
Qpcr	Quantitative polymerase chain reaction
RBCs	Red blood cells
RCR	Respiratory control ratio
RNA	Ribonucleic acid
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
RQ	Relative quantification
S phase	Synthesis Phase

S. D	Standard deviation
SGLT-1	Sodium-dependent glucose transporters type 1
SN	Substantia nigra
SOD	Superoxide dismutase
SPSS	Statistical Package for Social Science
STZ	Streptozocin
TAC	Total antioxidant capacity
TBA	Thiobarbituric acid
TCA	Trichloroacetic acid
TE	Trolox equivalent
TMB	3, 3′, 5, 5′-Tetramethylbenzidine
TMT	Trimethyltin
TNF-α	Tumor necrosis factor alpha
UDP-galactose	Uridine diphosphate galactose
UDP-glucose	Uridine diphosphate glucose
UK	United Kingdom
UV	Ultra violet
VCAM-1	Vascular cell adhesion molecule 1
VLDL	Very-low-density lipoprotein
WHO	World Health Organization

List of Content