

شبكة المعلومات الجامعية التوثيق الإلكتروني والميكروفيلو

بسم الله الرحمن الرحيم





MONA MAGHRABY



شبكة المعلومات الجامعية التوثيق الإلكتروني والميكروفيلو



شبكة المعلومات الجامعية التوثيق الالكتروني والميكروفيلم



MONA MAGHRABY



شبكة المعلومات الجامعية التوثيق الإلكترونى 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



يجب أن

تحفظ هذه الأقراص المدمجة بعيدا عن الغبار



MONA MAGHRABY



Faculty of Women for Arts, Science& Education Zoology Department

Effect of *Nigella sativa* on silver nanoparticles induced hepato-nephrotoxicity in *Clarias* gariepinus

Thesis

Submitted in partial fulfillment for the requirements for the degree of Master of Science in Zoology

By

Nermein Gamal Andrawes Georgy

B.Sc. of science, Zoology department,

Faculty of Women for Arts, science& Education

Ain Shams University

Supervision

Prof. Dr. Afaf Abd El- Galeel Abo Nour

Prof. Dr. Adel Abdel-Aleem Shaheen

Professor of Physiology, Zoology Department,
Faculty of Women for Arts, Science &
Education

Ain Shams University

Professor of fish diseases and management.

Department of aquatic animals diseases and management

Faculty of Veterinary Medicine

Benha university

Assoc. Prof. Heba Salah Mohamed Hamed

Associate professor of Fish physiology, Zoology Department,
Faculty of Women for Arts, Science & Education
Ain Shams University

2021

<u>DEDICATION</u>

To my Kind

Parents

To my

Faithful Brother

Kyrissos Gamas

Qualifications

Name : Nermein Gamal Andrawes

Scientific Degree: B.Sc.

Department : Zoology

College : Faculty of women for Arts, Science

& Education

University : Ain Shams University

Graduation year : 2013

Courses

Courses studied by the candidate in partial fulfillment of the requirements for the degree of M.Sc.

- 1-Physiology
- 2- Histology
- 3- Immunology
- **4- Toxicology**
- 5- Ethics of scientific research
- 6- Writing scientific research



Ain Shams University
Faculty of Women for Arts,
Science& Education
Zoology Department

APPROVAL SHEET

Name: Nermein Gamal Andrawes Georgy

Title: Effect of Nigella sativa on silver nanoparticles

induced hepato-nephrotoxicity in Clarias

gariepinus.

Scientific Degree: M.Sc.

Supervisors

Prof. Dr.

Prof. Dr. Adel Abdel-Aleem Shaheen

Afaf Abd El- Galeel Abo Nour

Professor of Physiology, Zoology Department, Faculty of Women for Arts, Science & Education Ain Shams University Professor of fish diseases and management.

Department of aquatic animals diseases and management.

Faculty of Veterinary Medicine

Benha university

Assoc. Prof. Heba Salah Mohamed Hamed

Associate professor of fish physiology,

Zoology Department,

Faculty of Women for Arts,

Science & Education

Ain Shams University

2021

ACKNOWLEDGMENT

Thanks my God, who gives me Knowledge and ability to finish this work.

I wish to express my deepest gratitude to:

Prof. Dr. Afaf Abd El- Galeel Abo Nour, Professor of Physiology, Zoology Department, Faculty of Women for Arts, Science & Education, Ain Shams University, for her supervision, kind encouragement, support in hard time and she suggested the plan to perfect work.

Prof. Dr. Adel Abdel-Aleem Shaheen, Professor of fish Diseases and Management, Department of Aquatic Animals Diseases and Management, Faculty Veterinary Medicine Benha University, for supervision, guidance, kind cooperation and his brilliant ideas to complete this work.

Assoc. Prof. Heba Salah Mohamed Hamed, Associate professor of Fish physiology, Zoology Department, Faculty of Women for Arts, Science & Education, Ain Shams University, much thanks from my heart for her great efforts, kind support, guidance, correction and her endurance for a long time.

My cordial thanks to my family for their support, kind encouragement donation and love.

ABSTRACT

Effect of *Nigella sativa* on silver nanoparticles induced hepato-nephrotoxicity in *Clarias* gariepinus

Silver nanoparticles (Ag-NPs) is effectively used in varied aquaculture applications. This study aimed to determine LC₅₀ of (Ag-NPs) in African catfish (*Clarias gariepinus*) as well as the protective role of *Nigella sativa* (NS) against Ag-NPs toxicity. Fish were divided into 4 groups of triplicates as follows: group I: was showed as control, group II: was fed on 3% NS inclusion diet, group III: was exposed to 50 mg/L Ag-NPs and group IV exposed to 50 mg/L Ag-NPs and fed on 3% NS for 30 days.

At the end of experiment blood collected for biochemical analysis. As well as liver and kidney tissues of catfish were taken for oxidative enzymes, DNA analysis and histopatholgical studies.

The results exhibited that exposed of catfish with 50 mg/L Ag-NPs lead to a marked increase in liver enzymes Alkaline phosphatase (ALP), alanine amino transferase (ALT) Aspartate amino transferase (AST). Also, these increases were indicated in kidney activities (urea, creatinine, and uric acid). Levels of total proteins, albumin and globulin were markdly increased. However, A/G ratio decreased. Also, glucose, cortisol and lactate dehydrogenase (LDH) levels were significantly increased. and total lipids levels (cholesterol and triglyceride) were increased. On the other and, African catfish exposed to Ag-NPs showed significant (p<0.05) elevation in the level of Lipid peroxidation (LPO) of liver and kidney tissues. These results indicated that the levels of reduction glutation (GSH), catalase (CAT), superoxide dismutase (SOD), and total antioxidant capacity (TAC) increased compared to the control fish after exposure to 50 mg/L Ag-NPs for 30 days.

Furthermore, percentage of DNA damage in both liver and kidney tissue of African catfish exposed to 50 mg/L Ag-NPs significantly increased.

Histological study in hepatic and renal tissues of African catfish represented more damage and alternation in tissues that exposed to 50 mg/L Ag-NPs for 30 days compare to control group and treatment group with 3% NS. It is concluded that the feeding of African catfish on a (NS) enriched diet minimized the destructive impacts of Ag-NPs toxicity and restored the abovementioned variable near to the control group. The current investigation has revealed that dietary NS has antagonistic functions against Ag-NPs toxicity in African catfish.

Key words: Silver nanoparticles

Clarias gariepinus

Nigella Sativa

Biochemistry

DNA fragmentation

Histopathology.

CONTENTS

Subjects P	age No.
List of tables	i
List of figures	vi
List of abbreviations	ii
Introduction	1
Aim of work	4
Review of literature	5
 Silver nanoparticles Clinical signs and behavioral effect Ag-NPs and liver function Ag-NPs and kidney functions Ag-NPs and total proteins Ag-NPs and stress indicators Ag-NPs and Lactated dehydrogenase enzyme Ag-NPs and lipid parameters Ag-NPs and oxidative stress and Anti-oxidants enzyme 1-Lipid peroxidation (LPO) 2-Reduced glutathione (GSH) 3-Catalase enzyme (CAT) 4-Superoxide dismutase (SOD) 5-Total Anti-oxidant Capacity (TAC) 	5 6 7 8 8 9 11 11 yme 12 12 13 14 14 15
Ag-NPs and DNA FragmentationAg-NPs and histopathological changeNigella sativa	16 17 18
Materials and methods	21
Materials	21
- Experimental fish	21

-Management -Chemicals Methods	21 22 23
 Preparation and characterization of silver nanoparticles Ag-NPs 	22
 Determination of 96 hours half lethal concentration (LC₅₀) of silver nanoparticles 	23
Preparation of dietExperimental design	23 24
Clinical investigation and post mortem examinationBiochemical studies	24 25
G- Determination of serum albumin H- Determination of serum globulin I- Determination of serum glucose J- Determination of serum cortisol K- Determination of serum LDH enzyme L- Serum lipogram - Determination of serum cholesterol	25 25 25 25 25 26 26 26 26 26 26 26 26
 Oxidative stress and antioxidant enzymes Determination of Lipid peroxidation (LPO) 	27 27
- Determination of Reduction glutathione (GSH)	27
<i>j</i> , , , , , , , , , , , , , , , , , , ,	27
 Determination of Superoxide dismutase (SOD) Determination of Total anti-oxidant capacity (TAC) 	27

_	Agrose gel electrophoresis for DNA fragmentation	28
-	Histopathological studies	29
-	Statistical analysis	29
Resu	llts	30
-	96 hours half lethal concentration (LC ₅₀) of silver	
	nanoparticles in Clarias gariepinus	30
_	Clinical signs and postmortem finding	33
-	Results of serum biochemical analysis	33
	1- Serum liver enzymes	33
	A- Serum Alkaline phosphatase (ALP)	33
	B- Serum Alkaline amino transferase (ALT)	33
	C- Aspartate amino transferase (AST)	34
	2- Serum kidney function	37
	A- Serum urea	37
	B- Serum creatinine	37
	C- Serum uric acid	37
	3- Serum immune function	40
	A- Serum total protein	40
	B- Serum albumin	40
	C- Serum globulin	40
	D- Serum A/G ratio	40
	4- Serum stress indicators	43
	A- Serum glucose	43
	B- Serum cortisol	43
	5- LDH enzyme	46
	6- Serum total lipids	46
	A- Cholesterol	46
	B- Triglycerides	46
	7- Oxidative stress and antioxidant enzymes	49
	A- Lipid peroxidation (LPO)	49
	B- Reduced glutathione (GSH)	49
	C- Catalase enzyme (CAT)	49
	D- Superoxide dismutase (SOD)	49
	E- Total antioxidant capacity (TAC)	50
	8- DNA fragmentation	50
	9- Histopathological change	57

Discussion		71
-	Half lethal concentration (LC ₅₀)	72
-	Clinical and behavioral change	73
-	Biochemical parameters	74
	1- Serum ALP, ALT and AST level	74
	2- Serum urea, creatinine and uric acid	76
	3- Serum protein, albumin and globulin level	77
	4- Serum glucose level	78
	5- Serum cortisol level	79
	6- Serum LDH level	81
	7- Serum cholesterol and triglycerides level	82
-	Oxidative stress and antioxidant enzyme	83
	 Liver and kidney LPO content 	84
	2- Liver and kidney GSH content	85
	3- Liver and kidney CAT content	86
	4- Liver and kidney SOD content	87
	5- Liver and kidney TAC content	88
-	DNA fragmentation	89
-	Histological changes	90
-	Nigella sativa	94
Conclusion		95
Sumn	nary	96
References		99
A rahi	ic summary	1