

Evaluation of the Effect of Zinc supplementation on the Health Status of Hemodialysis Patients

A Thesis submitted for fulfillment of Master Degree in Pharmaceutical Science
(Clinical Pharmacy)

By

Rasha Roshdy Ibrahim El-kady

B. Pharm. Sci.

Demonstrator at Clinical Pharmacy Department
Faculty of Pharmacy
Ain Shams University

Under supervision of:

Prof. Dr. Nagwa Ali Sabri

Professor of Clinical Pharmacy and
Head of Clinical Pharmacy Department
Faculty of Pharmacy
Ain Shams University

Dr. Ahmed Mohamed Elsis

Assistant professor of Internal medicine & Nephrology
National Institute of Urology & Nephrology

Dr. Sara Mahmoud Zaki

Lecturer of Clinical Pharmacy
Clinical pharmacy department
Faculty of Pharmacy
Ain Shams University

Acknowledgement

I am deeply thankful to "Allah" by the grace of whom, this work was possible.

I would like to express my deep appreciation and gratitude to Prof. Dr.

Nagwa Ali Sabri, Head of Clinical Pharmacy Department – Faculty of Pharmacy - Ain Shams University, for her sincere help, valuable guidance and continuous support in completing this work,

I am very grateful to Dr. Ahmed Elseasi, Assistant Professor of Internal medicine and Nephrology – National Institute of Urology and Nephrology, for his great assistance, precious advice and valuable guidance in completing this work,

I'm greatly indebted to Dr. Sara Mahmoud Zaki, Lecturer of Clinical Pharmacy - Faculty of Pharmacy - Ain Shams University, for her kind help, guidance and follow up throughout the whole work. She spared no effort or time through this research

I would like to thank all members of Clinical Pharmacy Department, Faculty of Pharmacy - Ain Shams University, for their continuous support.

Many special thanks and deep gratitude for my Parents and family and best friends for their kind and sincere help, love and support during the progress of this work.

List of contents

Contents	Page
List of abbreviations	i
List of tables	iii
List of figures	iv
Abstract	vi
Key Words	viii
Review of literature	
Renal Failure	
I- Criteria for definition.	2
II- Staging.	2
III- Epidemiology.	4
IV- Etiology and risk factors.	5
V- Pathophysiology of chronic kidney disease.	7
VI- Chronic Kidney Disease progression.	8
VII- Clinical presentation.	9
VIII- Kidney function assessment.	9
IX- Complications of chronic kidney disease.	11
X- Management.	19
XI- Quality of life (QOL) of hemodialysis patients.	28
Zinc	
1. Trace minerals in Hemodialysis patients.	32
2. Zinc.	33
3. Zinc deficiency.	36
4. Zinc deficiency in hemodialysis patients.	37
Role of clinical pharmacist in CKD management.	38
Aim of the work	41

Patients and methods	42
Results	60
Discussion	85
Conclusion	92
Recommendations	93
Summary	94
References	97
Appendix	118
Arabic summary	i

List of abbreviations

ACE	Angiotensin Converting Enzyme
ACEI	Angiotensin Converting Enzyme Inhibitor
ACM	All Cause Mortality
ACR	Albumin to creatinine ratio
ADMA	Asymmetric dimethyl-arginine
AER	Albumin Excretion Rate
AFB	Acetate-free biofiltration
Ag II	Angiotensin II
AKI	Acute Kidney Injury
ALA	Aminolevulinic acid
APD	Automated Peritoneal Dialysis
ARB	Angiotensin II Receptor Blocker
BP	Blood Pressure
BSA	Bovine Serum Albumin
CAPD	Continuous Ambulatory Peritoneal Dialysis
CKD	Chronic Kidney Disease
CKD-BMD	Chronic Kidney Disease- Bone Mineral Disease
CKD-EPI	Chronic Kidney Disease Epidemiology Collaboration
Cu⁺⁺	Cupric ion
CUA	Calcific uremic arteriolopathy
CV	Cardiovascular
CVD	Cardiovascular Disease
CVM	Cardiovascular Mortality
DM	Diabetes Mellitus
DN	Diabetic Nephropathy
DNA	Deoxyribonucleic acid
DTRPs	Drug Therapy Related Problems
ECV	Extracellular volume
eGFR	estimated Glomerular Filtration Rate
ELISA	Enzyme Linked Immunosorbant Assay
EPO	Erythropoietin
ESA	Erythropoiesis-Stimulating Agent
ESCP	European Society of Clinical Pharmacy
ESRD	End Stage Renal Disease
ET-1	Endothelin-1
Fe⁺⁺	Ferrous ion
FSS	Fatigue Severity Scale
GFR	Glomerular Filtration Rate
GSH	Glutathione peroxidase
H₂O₂	Hydrogen Peroxide
Hb	Hemoglobin
HCl	Hydrochloric acid
HD	Hemodialysis
HDF	Hemodiafiltration
HF	Hemofiltration
HNS	Hypertensive Nephrosclerosis

HRP	Horseradish peroxidase
hs-CRP	high sensitive - C Reactive Protein
HTN	Hypertension
IDH	Intradialytic Hypertension
IQR	Interquartile Range
IL-6	Interleukin-6
KDIGO	Kidney Disease Improving Global Outcome
KDOQI	Kidney Disease Outcome Quality Initiative
LDL	Low Density Lipoprotein
LVH	Left Ventricular Hypertrophy
MDA	Malondialdehyde
MDRD	Modification of Diet in Renal Disease
MRI	Magnetic Resonance Imaging
NADPH	Nicotinamide adenine dinucleotide phosphate oxidases
NHP	Nottingham Health Profile
NK	Natural Killer
O₂⁻	Superoxide anion
OH	Hydroxyl radical
PD	Peritoneal Dialysis
PEM	Protein Energy Malnutrition
pmp	Per million population
PTH	Parathyroid Hormone
QOL	Quality of Life
RA	Renal Anemia
RAAS	Renin Angiotensin Aldosterone System
RBCs	Red Blood Cells
RDI	Recommended Daily Intake
RHDx	Regular hemodialysis duration
RO	Renal Osteodystrophy
ROS	Reactive Oxygen Species
RRT	Renal Replacement Therapy
RT	Renal Transplantation
SCr	Serum Creatinine
SD	Standard deviation
SF-36	Medical outcomes study short form health survey
SH	Sulphydal
sHPT	secondary Hyperparathyroidism
SLE	Systemic Lupus Erythematosus
SOD	Super Oxide Dismutase
TAC	Total Antioxidant Capacity
TMB	tetramethylbenzidine
TNF-α	Tumor necrosis factor - α
U.S.	United States
USRDS	United States Renal Data System
VSMC	Vascular Smooth Muscle Cell
WST-1	Water soluble tetrazolium salt
Zn	Zinc

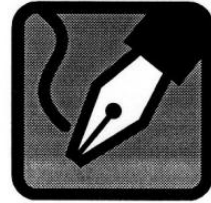
List of tables

Table	Page
Table (1): Staging of Chronic Kidney Disease based on Glomerular Filtration Rate.	3
Table (2): Staging of Chronic Kidney Disease based on Albuminuria.	3
Table (3): Etiology of ESRD in Egyptians.	6
Table (4): Risk factors for Chronic Kidney Disease.	6
Table (5): Risks and Benefits of peritoneal dialysis and hemodialysis.	25
Table (6): Baseline demographic data of the patients in the study groups	60
Table (7): Classification of ESRD causes in the study groups.	63
Table (8): Baseline laboratory investigations the study groups.	66
Table (9): Comparison between the study groups regarding serum zinc level before and after the study.	67
Table (10): Comparison between the study groups regarding serum MDA level before and after the study.	70
Table (11): Comparison between the study groups regarding serum TAC level before and after the study.	72
Table (12): Comparison between the study groups regarding serum SOD level before and after the study.	74
Table (13): Comparison between the study groups regarding serum hs-CRP level before and after the study.	76
Table (14): Comparison between the study groups regarding serum IL-6 level before and after the study.	78
Table (15): Comparison between the study groups regarding Hemoglobin level before and after the study.	80
Table (16): Comparison between the study groups regarding FSS before and after the study.	82

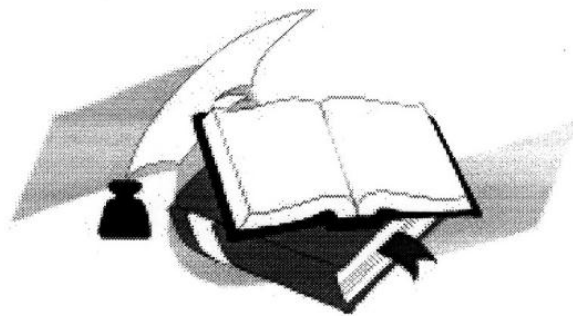
List of figures

Figure	Page
Figure (1): Prediction of chronic kidney Disease prognosis by glomerular filtration rate and albuminuria Categories.	9
Figure (2): The multifactorial pathogenesis of renal anemia.	13
Figure (3): Pathophysiology of cardiovascular Complications in Chronic Kidney Disease.	15
Figure (4): Pathogenesis of hyperphosphatemia and abnormalities of mineral and bone metabolism in chronic kidney disease.	17
Figure (5): Algorithm for management of anemia using iron and ESA therapy.	23
Figure (6): The process of diffusion and ultrafiltration	26
Figure (7): Factors contributing to fatigue in ESRD.	30
Figure (8): Detailed flow diagram of the study.	43
Figure (9): Fatigue Severity Scale	47
Figure (10): Principal of SOD assay kit	52
Figure (11): IL-6 ELISA test principal	56
Figure (12): Serial dilutions of the IL-6 standard.	57
Figure (13): Age in the study groups	61
Figure (14): Hemodialysis duration in the study groups	62
Figure (15): Co-morbidities in both study groups.	64
Figure (16): Concurrent medications administered in both groups.	65
Figure (17): Boxplot of serum zinc level in study groups before and after the study.	68
Figure (18): Boxplot of percent change of zinc level in both groups.	69
Figure (19): Boxplot of serum MDA level in study groups before and after the study.	71

Figure (20): Boxplot of serum TAC level in study groups before and after the study.	73
Figure (21): Median serum SOD level in study groups before and after the study.	75
Figure (22): Median serum hs-CRP level in study groups before and after the study.	77
Figure (23): Median serum IL-6 level in study groups before and after the study.	79
Figure (24): Median Hemoglobin level in study groups before and after the study.	81
Figure (25): Median Fatigue Severity Scale in study groups before and after the study.	83
Figure (26): Safety of zinc supplement in group 1 patients.	84



Abstract



Abstract

Introduction:

Dialysis has improved the life expectancy of End Stage Renal Disease (ESRD) patients, yet the mortality rate still remains disappointingly high. Oxidative stress and inflammation together with their complications such as cardiovascular diseases, protein-energy wasting and erythropoietin-resistant anemia are prevalent in hemodialysis (HD) patients and increase their mortality. The prevalence of zinc (Zn) deficiency in patients with HD is about 40–78%, which has been found to predispose them to oxidative stress, inflammation and anemia. Fatigue is one of the most common symptoms experienced by dialysis patients, with its prevalence ranging from 60% to 97%. Zinc supplementation appears to ameliorate fatigue in chronic fatigue syndrome patients.

Aim of the work:

The aim of this study was to evaluate the role of zinc as an adjunctive therapy on oxidative stress, inflammation, anemia and fatigue in hemodialysis patients.

Patients and methods:

Patients on long-term hemodialysis with lower than normal serum zinc concentration ($< 80 \mu\text{g/dL}$) were randomized in to 2 groups; group 1 received 25 mg/day oral Zn supplement ($n = 28$) and group 2 received placebo ($n = 24$) for twelve weeks. Serum Zn, malondialdehyde (MDA), total antioxidant capacity (TAC), superoxide dismutase (SOD) activity, high sensitive-CRP (hs-CRP), interleukin-6 (IL-6) and hemoglobin (Hb) were assessed before and after the supplementation period. Quality of life was assessed using Fatigue Severity Scale (FSS) at baseline and at the end of the study.

Results:

The results obtained showed that there was a significant increase in both serum Zn and Hb levels from baseline values in patients of group1 with ($p<0.001$) and ($p=0.002$) respectively. Also these levels were significantly different from post-study levels of both Zn and Hb in patients of group 2 with ($p<0.001$) and ($p=0.005$) respectively. Moreover, there were significant changes in serum MDA and TAC levels in group1 at the end of the study ($p< 0.05$) without significant change between the two groups. There were no significant changes in SOD activity within groups or between the two groups at the end of the study.

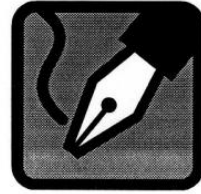
Concerning serum levels of inflammatory marker, it was found Zn supplementation did not affect serum hs-CRP or in IL-6 levels with no significant difference within or between both groups. Using FSS, there were a significant decrease in fatigue in group 1 ($p = 0.001$) with a significant difference between the two groups in percent change ($p < 0.001$) at the end of the study.

Conclusions:

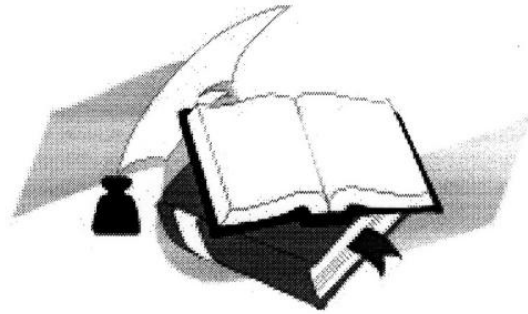
From the results found it was concluded that, zinc supplementation at dose of 25 mg/day for 12 weeks ameliorates low serum Zn and Hb levels, improve fatigue and may reduce oxidative stress in Zn deficient HD patients suggesting that Zn supplementation may be considered an adjunctive treatment in HD to reduce morbidity and mortality risk factors. Zinc supplementation at dose of 25 mg/day for 12 weeks does not ameliorate inflammation in these patients.

Key Words

Anemia – Chronic kidney disease – End Stage Renal Disease – Fatigue – Hemodialysis – Hemoglobin – Inflammation – Oxidative stress – Zinc supplementation



Review of Literature



Renal Failure

Kidney failure is classified into two broad categories: acute kidney injury and chronic kidney disease. The basic difference between them is the rate of disease progression, which is rapid for acute kidney injury (usually within days or weeks) and gradual for chronic kidney disease (usually in the range of years) (**Chawla et al., 2014**).

Both acute and chronic kidney disease can lead to complete loss of kidney function, causing the patient to depend on renal replacement therapy either dialysis or transplantation (**Tammen et al., 2014**).

Acute Kidney Injury (AKI):

It is characterized by abrupt decline in glomerular filtration rate (GFR). Many definitions for AKI are present with the most popular one is the elevation of serum creatinine > 0.3 mg/dL over a baseline creatinine below 2 mg/dL (**Kellum et al., 2011**).

Chronic Kidney Disease(CKD):

It is a life-threatening condition characterized by progressive and irreversible loss of kidney function. The growing inability of the kidneys to adequately clear the blood of waste products finally results in the implementation of dialysis (or kidney transplant) in order to prevent azotemia, systemic organ damage and death (**Lopez-Novoa et al., 2010**).

The Kidney Disease Improving Global Outcome (KDIGO) has defined CKD as:

Abnormalities in kidney structure or function persist for 3 months or longer, with implications for health (**KDIGO, 2013**).