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بسم اللّه الرحمن الرحيم

مركز الشبكات وتكنولوجيا المعلومات

قسم التوثيق الإلكتروني

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التوثيق الإلكتروني والميكروفيلم

قسم

نقسم باللّه العظيم أن المادة التي تم توثيقها وتسجيلها

علي هذه الأقراص المدمجة قد أعدت دون أية تغيرات

Ain Shams University
Faculty of Pharmacy
Department of Pharmacology and Toxicology

Study of the Modulatory Effect of Amiloride on Doxorubicin Induced Neurotoxicity in Rats

A Thesis
submitted in partial fulfillment of the requirements for
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(Pharmacology & Toxicology)

By

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In the name of Allah, The Most Gracious and The Most Merciful, Peace and blessings be upon our Prophet Mohammed and his good followers till the Day of Judgement.

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Cognitive impairment or "chemobrain" is a troublesome adverse effect which had been increasingly reported by cancer patients after doxorubicin (DOX) chemotherapy. Notably, Hypertension, a very common comorbidity in cancer patients, could pose a greater risk for negative cognitive outcomes. Amiloride (AML) is an antihypertensive, potassium-sparing diuretic that has been proven to be neuroprotective in different experimental models; this can be attributed to its ability to inhibit different ion transporters such as $Na^{+/}H^{+}$ exchanger (NHE), which upon excessive activation can result in intracellular cationic overload, followed by oxidative damage and cellular death. Accordingly, this study was designed to investigate the potential neuroprotective effect of AML against DOX-induced chemobrain and to elucidate possible underlying mechanisms. Briefly, Histopathological examination and neurobehavioral testing (Morris water maze, Y maze and passive avoidance test) showed that AML co-treatment (10mg/kg/day) markedly attenuated DOX (2mg/kg/week)-induced neurodegeneration and memory impairment after 4 weeks of treatments. We found that DOX administration up-regulated NHE expression and increased lactic acid content in the hippocampus which were markedly opposed by AML. Moreover, AML mitigated DOX-induced neuroinflammation and decreased hippocampal tumor necrosis factor-α level, nuclear factor kappa-B, and cyclooxygenase-2 expression. Additionally, AML counteracted DOX-induced hippocampal oxidative stress as indicated by normalized malondialdehyde and glutathione levels. Furthermore, AML halted DOX-induced hippocampal apoptosis as evidenced by decreased caspase-3 activity and lower cytochrome c immunoexpression. Our results in addition to the previously reported antitumor effects of AML and its ability to mitigate cancer resistance to DOX therapy could point toward possible new repositioning scenarios of the diuretic AML especially regarding hypertensive cancer patients.

KEY WORDS: Doxorubicin, Amiloride, Hypertension, Chemobrain, Na^{+}/H^{+} exchanger

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LIST OF ABBREVIATIONS

Abbreviation	*Term*
°C	Degree Celsius
µl	Micro liter
µg	Micro gram
µmol	Micro mole
A_{sample}	Sample absorbance
$A_{standard}$	Standard absorbance
Ag	Antigen
AML	Amiloride
ANOVA	Analysis of variance
ApoA-1	Apolipoprotein A-1
ASIC	Acid sensing ion channel
ATP	Adenosine-5'-triphosphate
Aβ	Amyloid beta protein
BBB	Blood–brain barrier
BSA	Bovine serum albumin
Ca^{2+}	Calcium
CA	Cornu Ammonis
CAT	Catalase
cm	Centimeter
CNS	Central nervous system
COX-2	Cyclooxygenase- 2
CSF	Cerebrospinal fluid
Cu	Cupper
DTNB	5,5′-Dithiobis(2-nitrobenzoic acid)
DNA	Deoxyribonucleic acid
DOX	Doxorubicin
ECL	Enhanced Chemiluminescence
EEG	Electroencephalogram
ELISA	Enzyme linked immunosorbent assay
ENaC	Epithelial sodium channel
FDA	Food and drug administration
Fe	Ferrous
g/gm	Gram
GSH	Glutathione reduced
H^{+}	Hydrogen
hr	Hour

Abbreviation	*Term*
H&E	Hematoxylin and Eosin
H_2O	Water
H_2O_2	Hydrogen peroxide
HRP	horseradish peroxidase
i.p.	Intraperitoneal injection
IHC	Immunohistochemical
IL-6	Interleukin- 6
IL-1β	Interleukin- 1 Beta
IRP-1	Iron Regulatory Protein-1
K^+	Potassium,
Kg	Kilogram
LPS	lipopolysaccharide
M	Molar
MDA	Malondialdehyde
mg	Milligram
min	Minute
ml	Milliliter
mM	Millimolar
mmol	Millimole
MnSOD	Manganese Superoxide Dismutase
MPP+	1-methyl-4-phenylpyridinium
MPTP	Mitochondrial Permeability Transition Pore
MWM	Morris water maze
Na^+	Sodium
Na^+-K^+-ATPase	Sodium Potassium ATP pump
NF-κB	Nuclear factor-kappa b
NHE	Sodium/Hydrogen Exchanger
NCX	Sodium/Calcium Exchanger
ng	nanogram
nm	Nanometer
nmol	Nanomole
O.D	Optical density
PA	passive avoidance
PAGE	Polyacrylamide gel electrophoresis
PBS	Phosphate buffer saline
pHi	intracellular pH
PPE	Palmar-Plantar Erythodysesthesia
PVDF	Polyvinylidene Fluoride
pNA	Para -nitro aniline

Abbreviation	*Term*
R.T	Room temperature
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
s/sec	Second
SAP	Percent of spontaneous alternation
SD	Standard deviation
SDS	Sodium dodecyl sulfate
SOD	Superoxide dismutase
TAE	Total number of arm entries
TEMED	Tetramethylethylenediamine
TBA	Thiobarbaturic acid
TBS	Tris buffered saline
TBST	Tris Buffered Saline-Tween
TMB	Tetramethylbenzidine
TNF-α	Tumor necrosis factor-alpha

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