Potential Cardioprotective Effects of Vitamin D on Cardiotoxicity induced by Doxorubicin

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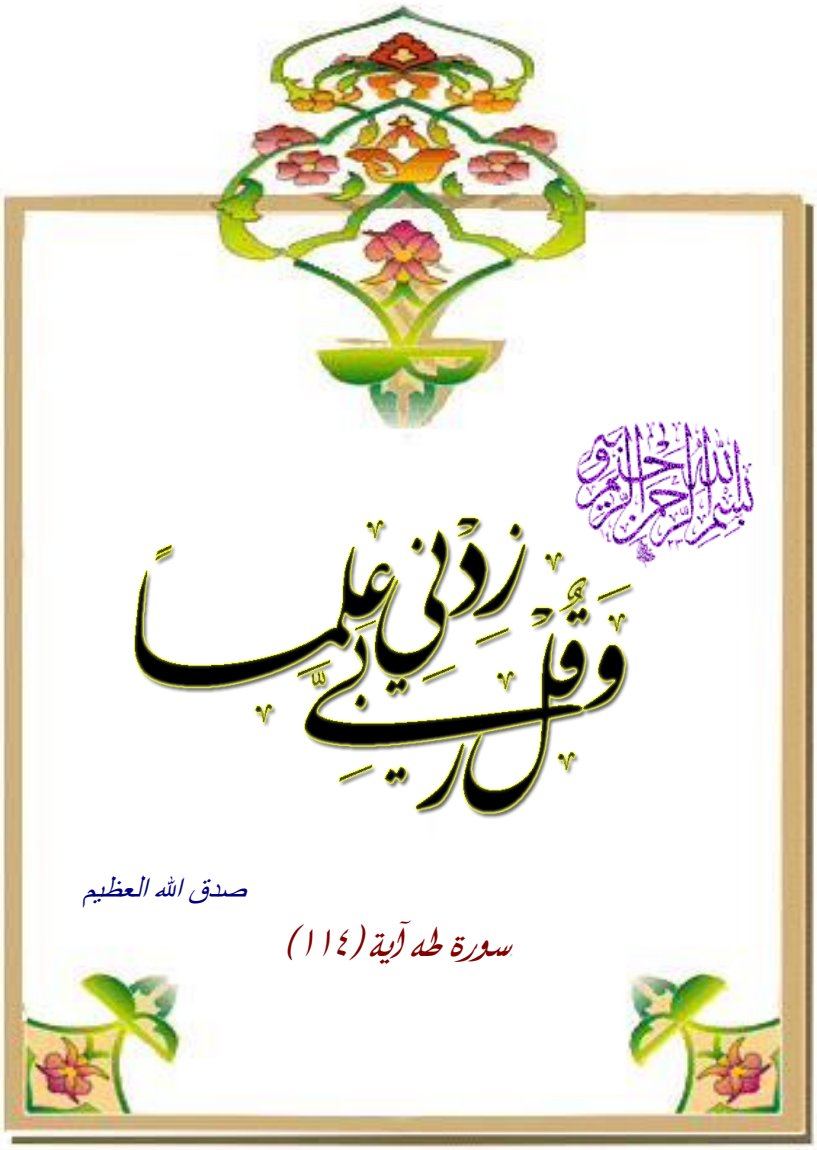

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List of Contents

Subject Page

Abstract

OBJECTIVES: The present study was conducted to evaluate the role of vitamin D on the acute cardiotoxicity induced by doxorubicin in albino rats and explore the possible underlying mechanism(s).

DESIGN: The present study was performed on forty- eight adult male rats randomly assigned to four groups, each group has (N = 12): Group I : Normal control rats.Group II Doxorubicin- treated rats, received doxorubicin i.p. inj at dose of 15 mg / kg b.w.and sacrificed 24 hours after the injection. Group III : Vitamin D pretreated- Doxorubicin intoxicated rats, rats of this group were pretreated (i.p. injected) with vitamin D for three successive days as well as the day of doxorubicin injection., rats of this group will be sacrificed 24 hours after treatment with doxorubicin. Group IV: Vitamin D treated - rats. Rats of this group were injected by vitamin D I.P.in a dose of 10,000/100g b.w., for four days. On the day of sacrifice, blood pressures were measured and rats were subjected to ECG recording. Blood sample were collected in heparinized tubes and the separated plasma was subjected to the measurement of calcium, troponin I as well as CK-MB. Thereafter, the chest cage was opened, the heart was removed washed in saline and dried by filter paper and preserved at -80°c for subsequent determination of MDA, SOD & GSH in cardiac tissue.

RESULTS: The results of the present study revealed that doxorubicin treated rats showed a significant bradycardia, prolongation of PR and QTc interval in the ECG recording as well as prolongation of QRS wave, also both the systolic and diastolic blood pressures were elevated compared to the control group. The plasma level of calcium was significantly decreased while that of troponin I & CPK significantly increased in the doxorubicin treated rats compared to control rats. Regarding the cardiac tissue, the levels of both MDA and SOD were significantly elevated, while GSH level was significantly decreased in the doxorubicin treated rats compared to the control rats. The results of Vitamin D pretreated-doxorubicin treated rats showed a normalization of QTc interval, blood pressure and the plasma level of troponin I and CK-MB as well as the cardiac tissue levels of MDA, SOD and GSH, compared to the control group. The plasma calcium level was significantly increased in the vitamin D pretreated-doxorubicin intoxicated rats compared to the control rats. Regarding the vitamin D treated rats, the plasma calcium level showed a significant increase.

Doxorubicin provoked an inflammatory responses as indicated by the increased expression of nuclear factor kappa B (NF-kB), and this response was abolished by the vitamin D supplementation. Moreover, doxorubicin resulted in an increased apoptotic tissue damage by increasing the expression of Bax, cytochrome c and caspase 3 activity.Vitamin D pretreatment abolished the acute apoptotic actions induced by doxorubicin.

CONCLUSION: Vitamin D could be considered as a potent cardioprotective agent against the acute doxorubicin-induced cardiotoxicity via suppressing the oxidative stress, preventing the inflammatory reactions and antagonizing the apoptotic damage, as well as normalizing the hypocalcemic response induced by doxorubicin.

List of Abbreviations

ACE	Angiotensin-converting enzyme
Ang II	Angiotensin II
ANP	Atrial natriuretic peptide
Bax	bcl-2-like protein 4
Bcl-2	B-cell lymphoma 2
BNP	Brain natriuretic peptides
BP	Blood pressure
Bpm	Beats per minute
CASPASES	Cysteine Aspartyl-specific Proteases
CAT	Catalase
CHF	Congestive heart failure
CK-MB	Creatinine kinase MB
CV	Cardiovascular
CVD	Cardiovascular disease
DBP	Vitamin D binding protein
DOX	Doxorubicin
DOX-Fe	Doxorubicin-iron complexes
ECG	Electrocariography
FGF	Fibroblast growth factor (23)
G6PDH	Glucose-6-phosphate dehydrogenase ()
GPx	Glutathione peroxidase
H & E	Hematoxylin and Eosin
H2O2	Hydrogen peroxide
HF	Heart failure
HR	Heart rate
IFCC	International Fedration of clinical chemistry.
Ip	Intraperitoneal
IU	International unit
LDL	low density lipoprotein
MDA	Malonyl dialdeyde
NBT	Nitroblue tetrazolium

List of Abbreviations (Cont.)

NE	Norepinephrine
NF-Kb	Nuclear Factor kappa- B
NSAIDs	Nonsteroidal anti-inflammatory drugs.
O_2^-	Superoxide anion radical
OD	Optical density
PI3K	Phosphatidylinositol-3 kinase
PKC	Protein kinase C),
PMS	Phenazine methosulphate
PTH	Parathyroid hormone.
RAS	Renin-Angiotensin System
ROS	Reactive oxygen species
RXR	Retinoid X receptor
SD	Standard deviation
SOD	Superoxide dismutase,
TBA	Thiobarbituric acid reagent
TBARS	Thiobarbituric acid reactive substance
TBS	Tris-buffered saline
TCAA	Trichloroacetic acid
TNFR	Tumor necrosis factor receptor
UV	Ultraviolet (UV)
VDR	Vitamin D receptors
VDRE	Vitamin D response elements

List of Tables

List of Figures

List of Figures (Cont.)

List of Figures (Cont.)

1. Doxorubicin

The anthracyclines are a group of antibiotics being highly effective against a spectrum of malignancies including both hematological and solid tumors. Doxorubicin (DOX) is an anthracyline drug first extracted from *Streptomyces peucetius var. caesius* in the 1970's and routinely used in the treatment of several cancers including breast, lung, gastric, ovarian, thyroid, non-Hodgkin's and Hodgkin's lymphoma, multiple myeloma, sarcoma, and paediatric cancers **(Cortes-Funes and Coronado,2007).**

1.1. Chemistry

DOX belongs to the group of anthracycline antibiotics that consists of the tetracyclic quinoid aglycone adriamycinone (14-hydroxydaunomycinone) linked to the amino- sugar daunosamine. The chemical name for adriamycin is therefore the following: (7S : 9S)-9-hydroxyacetyl-4-methoxy-7,8,9,10-tetrahydro-6,7,9, ll-tetrahydroxy-7-0-(2′3′6′-trideoxy-3′-amino-α-L-lyxohexopyranosyl) -5,12-naphthacenedione. The chemical structure of Doxorubicin hydrochloride is ahown in (Fig. 1)

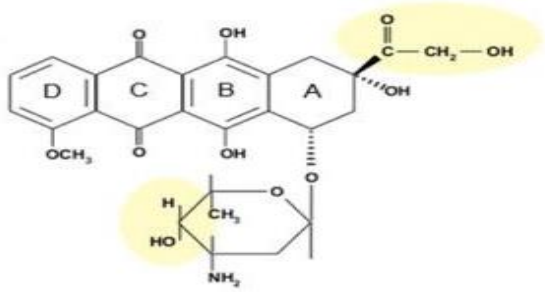

Fig. (1): Chemical structure of Doxorubicin (DOX) ***(Torres and Simic, 2012)***

Doxorubicin hydrochloride: Molecular formula is $C_{27}H_{29}NO_{11}$.HCl.; Molecular weight is 579.99 and CAS number, 25316-40-9. It is soluble in water and isotonic sodium chloride and slightly soluble in methanol. It is an orange red hygroscopic crystalline powder.

1.2. Pharmacodynamics

The mechanisms of DOX cytotoxicity in cancer cells is complex including: (i) DNA cross-linking - DNA alkylation and inhibition of both DNA replication and RNA transcription also (ii) it inhibits topoisomerase II and (iii) induces free radicals generation, leading to DNA damage & lipid peroxidation**. (Minotti et al., 2004a).**

1.2.1. *DNA Intercalation*

The anti-cancer activity of anthracyclines is likely due to their intercalation into DNA, which may disrupt replication and transcription of genomic DNA and lead to the death of cancer cells **(Tewey et al., 1984)**

Intercalation into DNA leading to inhibition of macromolecular synthesis was the first mechanism described for cytotoxicity of anthracyclines **(Marco et al., 1975).** The rather strong binding of daunorubicin and DOX to DNA has been characterized extensively **(Chaires et al., 1982 and Chaires et al 1996)**. Considering this and also taking into account that the DNA in cells does not occur naked but as chromatin, it seems unlikely that DNA intercalation is the only or most essential pathway of anthracycline cytotoxicity.