AUCTORES
Globalize your Research

Journal of Pharmaceutics and Pharmacology Research
Mohamed Zaeim Hafez Ahmed*

Open Access

Research Article

Protective effect of vitamin D and Doxorubicin on Induced Cardiotoxicity in Male Albino Rats

Ahmedel-Sayed Nour El-Deen [1], Ahmad Gadalla [1], Mohamed Zaeim Hafez Ahmed [1*], Elsayed Elagamy Elsayed Mohamed [2], Ramadan Hassan Ibrahim Thabet [2], Ahmed Mohamed Gad Allah [3], Ahmed Abdrabo Elshenawy Elsisi [2], Ahmed Abdel Nasser Ahmed Mohamed [2], Reda Mohamed Abdrabbou Fayyad [2], Ayman Sabry Yassin Al Sayed [4], Mohamed Nabil Mohamed [4], Mohammed Zain-Alabedeen [4] Abdelkareem, Rafik Elsayed Aboelhythim Ereba [2], Muhammad Abdelbaeth Hassan Elfiky [1]

[1] Physiology department, Faculty of medicine, Al Azhar University, Egypt.

[2] Pharmacology department, Faculty of medicine, Al Azhar University, Egypt.

[3] Forensic Medicine and Clinical Toxicology department, Faculty of medicine, Al Azhar University, Egypt.

[4] Microbiology department, Faculty of medicine, Al Azhar University, Egypt.

***Corresponding Author:** Mohamed Zaeim Hafez Ahmed, Physiology department, Faculty of medicine, Al Azhar University, Egypt.

Received date: April 24, 2023; **Accepted date:** May 05, 2023; **Published date:** May 15, 2023

Citation: Ahmedel-Sayed N. El-Deen, Ahmad Gadalla, Hafez Ahmed MZ, Elsayed Mohamed EE, Ibrahim Thabet RH, et al., (2023), Protective effect of vitamin D and Doxorubicin on Induced Cardiotoxicity in Male Albino Rats, *J. Pharmaceutics and Pharmacology Research*, 6(4); **DOI:** 10.31579/2693-7247/132

Abstract

Background & Objectives: Cardiotoxicity can manifest as abnormalities in the electrical representations of heart beats as seen by an electrocardiogram, such as a prolongation in the interval between the Q wave and T wave (QT interval), decreased amplitude of QRS complexes, a decrease in left ventricular ejection fraction, and cardiomyopathy. In order to assess the potential protective impact of vitamin D (Vit D) against the cardiotoxicity caused by doxorubicin (Dox).

Materials and Methods: The animal house of Al Azhar University sold 28 adult males Wistar albino rats of the local strain, weighing (850±150 g). Seven of them were housed in cages and given free access to food, water and kept in standardized settings with 12-hour light/dark cycles and 24-degree temperatures. Prior to the trial, they were held for two weeks to acclimatize to the new surroundings. The animals randomly divided into four groups of 7 rats in each, group 1(control group), group 2 (doxorubicin treated group), group 3 (Vit D pre-treated group), group 4 (Vit D and doxorubicin co-treated group).

Results: Serum calcium, creatine kinase, cardiac troponin concentration, and malondialdehyde levels in cardiac tissue significantly increased among group 2 (92.76±3.60, 95.51±5.13, 1.05±0.15, 9.70±0.82) than controls (14.07±0.84,15.44±0.65, 0.02±0.01, 6.09±0.27), group 3 (42.91±7.66, 38.60±2.47, 0.23±0.10, 7.93±0.33) and group 4 (30.29±2.06, 32.01±2.39, 0.37±0.09, 7.27±0.43), respectively (P<0.001).

Conclusion: The current results demonstrate that vitamin D supplementation in the diet, whether mild or moderate, was cardioprotective and didn't interfere with Dox ability to treat cancer in rats. It is reasonable to conduct more research to determine the ideal vitamin D dosages required to enhance heart functions without reducing Dox effectiveness.

Key words: cardiotoxicity; doxorubicin; male albino rates, Vitamin D supplementation

Introduction

Cardiotoxicity may manifest as irregularities in the electrical representations of heartbeats on an echocardiography (ECG), such as increase in the QT interval, a decrease in the amplitude of QRS complexes, and decrease in left ventricular ejection fraction, as well as cardiomyopathy **[1]**.

The anti-cancer drug doxorubicin (Dox) is extensively used in chemotherapy; however, the main side effects prevent it from being utilized in clinical settings is cardiotoxicity **[2]**.

The pathogenesis of Dox-induced cardiotoxicity involves the same to involve number of mechanisms, as oxidative stress-induced cell damage,

lipid peroxidation, mitochondrial damage, inflammation, and apoptosis. The elevation free radical production and oxidative stress are the main contributors to Dox-induced cardiotoxicity **[2]**.

The following methods are regarded to be the main ways that Dox, they are easily absorbed by cells and concentrate. Deoxyribonucleic acid (DNA) biosynthesis can be stopped by Dox ability to block DNA intercalation. This drug makes hydrogen bonds with guanine to create ternary Dox-DNA-topoisomerase II complexes, which activate DNA damage responses and ultimately cause cell death. This drug has a great affinity for ECG base pair sites **[3]**.

Vitamin D receptors are present in several tissues, including endothelial cells, vascular smooth muscle cells, and cardiomyocytes. Additionally, it has been shown to have an impact on inflammation, cell differentiation, and proliferation **[4]**.

Several human observational studies supported these experimental results raise the possibility that vitamin D or its equivalents may prove therapeutically advantageous in the prevention or treatment of cardiovascular disease **[5]**.

Furthermore, interleukin and C-reactive protein levels as well as congestive heart failure and low vitamin D levels all of them have been associated. Due to that vitamin D acts as a natural vascular defender factor. In fact, both hypertension, cardiac hypertrophy, enhanced renin

expression, and higher angiotensin II concentration are all signs of experimental vitamin D deficiency **[6]**.

Therefore, the aim of this study was to investigate whether vitamin D could provide cardiotoxicity protection against doxorubicin.

Materials and Methods

Animal Selection

A total of 28 mature male albino rats of the local strain (To avoid effect of feminine hormonal changes on the heart) from the Al Azhar University's animal house for 850±150 g. Seven of them were housed in a cage, and they given free access to food, water, and kept in standardised circumstances with 12-hour light/dark cycles and 24-degree temperatures. The university's animal ethics committee gave its approval to experimental methods (aproval ID, AZHPHY2021). Prior to the trial, they were held for two weeks to acclimate to the new surroundings.

Chemicals

Vitamin D (cholecalciferol) was administered in drops with a concentration of 2800 IU/ml, each containing 100 IU of Vitamin D, in DOX 50mg vials containing 25 ml of red solution, which were purchased from Pfizer Egypt Pharmaceutical Company (Vidrop, Medical Union Pharmaceuticals, Abu-Sultan, Ismailia, Egypt).

Experimental Procedures: All procedures were conducted at Al-Azhar University Assuit, Egypt.

Induction of cardiotoxicity: Doxorubicin, at a dose of 8.0 mg/kg, was administered once intraperitoneally after an adjustment time to cause cardiomyopathy. **[7]**

The animals were randomly divided into 4 groups of 7 rats in each:

Group 1(control group): standard control received intraperitoneal (i.p.) normal saline 5 ml/kg body weight for 4 weeks,

Group 2 (Doxorubicin treated group): the doxorubicin (DOX) group got a single intravenous dosage of DOX (25 mg/kg). **[8]**

Group 3 (Vit D Pre-treated group): Rats used as the pretreatment Vit D group received daily vitamin D supplements of 10,000 IU per 100 g of body weight for 4 weeks, followed by an intravenous injection of DOX (25 mg/kg).

Group 4 (Vit D and doxorubicin Co-treated group): Rats used in the doxorubicin plus vitamin D (DOX+Vit D) group received a single intravenous dose of DOX (25 mg/kg) and 10000 IU of vitamin D per 100 g of body weight each day for 4 weeks on the first day of the experiment.

***Collection of blood samples*:** Each rat had a tail vein removed, and fresh blood was drawn into tubes containing potassium oxalate and sodium fluoride. Measurements of cardiac troponin I, creatine kinase isoenzyme-MB (CK-MB) activity, and serum calcium were made using blood samples.

Biochemical parameters

Serum calcium: A Hitachi (model Z-5700) atomic absorption spectrophotometer graphite-furnace atomizer with modifications was used to measure the serum calcium level **[9].**

Serum creatine kinase: The serum CK-MB activity was determined spectrophotometrically in line with the suggested procedures using commercial kits obtained from spectrum diagnostics, Cairo, Egypt **[10].**

Serum level of cardiac troponin I: The serum level of cardiac troponin I (c-TnI) was measured using an enzyme linked immunosorbent assay (ELISA) kit produced by life diagnostics Inc. in accordance with the manufacturer's instructions **[10].**

Assessment of systolic blood pressure (SBP): measured in conscious rats using a non-invasive blood pressure monitor (tail-cuff plethysmograph) from (Ugo in Basile, Italy), in accordance with the standards specified by Irvine et al., **[11]**. On days 1 and 30, SBP measured one hour after the administration of drug. Back-to-back measurements of five were done. To calculate the value, the lowest three systolic blood pressure values were averaged**.[12]**

Electrocardiogram (ECG): Rats got 50 mg/kg sodium pentobarbital ip injection at the conclusion of the experiment **[13]**. Rats given general anaesthesia before being placed supine on a board, where the ECG continuously recorded using industry-standard lead II (right forelimb to left hind limb), which is artifact-free. Needles electrodes inserted into the pads of each rat's paws and connected to a Biocare ECG 101 (Shenzhen Biocare Electronics Co., Ltd., China). The ECG used to determine the length and amplitude of the Pwave, QRS complex, and ST segment alterations **[14]**

Measurement of MDA, SOD and GSH in cardiac tissue

The cardiac tissues were subsequently immersed in cold saline (1:10 w/v), then homogenised using a machine. The levels of superoxide dismutase (SOD), malondialdehyde (MDA), and glutathione (GSH) in cardiac tissues then determined by centrifuging the supernatant at 3000 rpm **[15].**

Statistical analysis

Following the analysis of variance (ANOVA) test, the data were examined using the least significant difference (LSD) post hoc test in Statistical Package for the Social Sciences (SPSS) 23. The information was converted to mean standard deviation (SD). A value is considered significant level when $P \leq 0.05$..

Results

A total of 28 male albino rats included in our study, it was discovered that the serum calcium levels of group 2 significantly higher (92.76±3.60) than control group (14.07±0.84), group 3 (42.91±7.66), and group 4 (30.29±2.06) (P 0.001), **(Table 1).**

Serum calcium	Mean ±SD	Range	F	P value	95% CI	
					Lower	Upper
Normal control group (N=7)	14.07±0.84	12.90-15.20	421.245	**<0.001 ***	13.29	14.85
Doxorubicin treated group (N=7)	92.76±3.60	97.80-87.90			89.43	96.08
Vit D Pre-treated group (N=7)	42.91±7.66	33.70-52.40			35.83	50.00
Vit D and doxorubicin Co-treated group (N=7)	30.29±2.06	27.20-33.10			28.38	32.19
Post Hoc P (Mean Difference)	*P1<0.001*(78.69), P2<0.001*(28.84), P3<0.001*(16.21), P4<0.001*(49.84), P5<0.001*(62.47), P6<0.001*(12.63)*					

Table 1: Serum calcium among the studied groups of rats

F: One way ANOVA test. *****Significant. **CI:** Confidence interval for Mean.
P1: normal control group compared doxorubicin treated group.
P2: normal control group compared vit D Pre-treated group.
P3: normal control group compared vit D and doxorubicin Co-treated group.
P4: doxorubicin treated group compared vit D Pre-treated group.
P5: doxorubicin treated group compared vit D and doxorubicin Co-treated group.
P6: vit D Pre-treated group compared vit D and doxorubicin Co-treated group.

Additionally, serum creatine kinase levels substantially higher in group 2 (95.51±5.13) than in the control group (15.44±0.65), group 3 (38.60±2.47), and group 4 (32.01±2.39) groups (P<0.001) groups. (**Table2)**

Serum creatine kinase (CK-MB) (IU/L)	Mean ±SD	Range	F	P value	95% CI	
					Lower	Upper
Normal control group (N=7)	15.44±0.65	14.66-16.40	880.608	**<0.001***	14.84	16.04
Doxorubicin treated group (N=7)	95.51±5.13	89.30-102.70			90.77	100.26
Vit D Pre-treated group (N=7)	38.60±2.47	35.60-42.60			36.31	40.89
Vit D and doxorubicin Co-treated group (N=7)	32.01±2.39	29.60-35.70			29.81	34.22
Post Hoc P (Mean Difference)	*P1<0.001*(80.08), P2<0.001*(23.16), P3<0.001*(16.58), P4<0.001*(56.91), P5<0.001*(63.50), P6=0.001(6.59)*					

Table 2: Serum creatine kinase among the studied groups of rats

F: One way ANOVA test. *****Significant. **CI:** Confidence interval for Mean.
P1: normal control group compared doxorubicin treated group.
P2: normal control group compared vit D Pre-treated group.
P3: normal control group compared vit D and doxorubicin Co-treated group.
P4: doxorubicin treated group compared vit D Pre-treated group.
P5: doxorubicin treated group compared vit D and doxorubicin Co-treated group.
P6: vit D Pre-treated group compared vit D and doxorubicin Co-treated group.

Moreover, serum cardiac troponin concentration substantially higher in group 2 (1.05±0.15) compared to control group (0.02±0.01), group 3 (0.23±0.10), and group 4 (0.37±0.09), (P<0.001) **(Table 3) and (Figure 1).**

Serum level of cardiac troponin I (ng/ml)	Mean ±SD	Range	F	P value	95% CI	
					Lower	Upper
Normal control group (N=7)	0.02±0.01	0.01-0.03	138.035	**<0.001***	0.01	0.02
Doxorubicin treated group (N=7)	1.05±0.15	0.88-1.24			0.92	1.18
Vit D Pre-treated group (N=7)	0.23±0.10	0.10-0.38			0.13	0.32
Vit D and doxorubicin Co-treated group (N=7)	0.37±0.09	0.25-0.52			0.29	0.46
Post Hoc P (Mean Difference)	*P1<0.001*(1.03), P2=0.001(0.21), P3<0.001*(0.35), P4<0.001*(0.82), P5<0.001*(0.68), P6=0.012(0.15)*					

Table 3: Serum level of cardiac troponin I among the studied groups of rats

F: One way ANOVA test. *****Significant. **CI:** Confidence interval for Mean.
P1: normal control group compared doxorubicin treated group.
P2: normal control group compared vit D Pre-treated group.
P3: normal control group compared vit D and doxorubicin Co-treated group.
P4: doxorubicin treated group compared vit D Pre-treated group.
P5: doxorubicin treated group compared vit D and doxorubicin Co-treated group.

P6: vit D Pre-treated group compared vit D and doxorubicin Co-treated group.

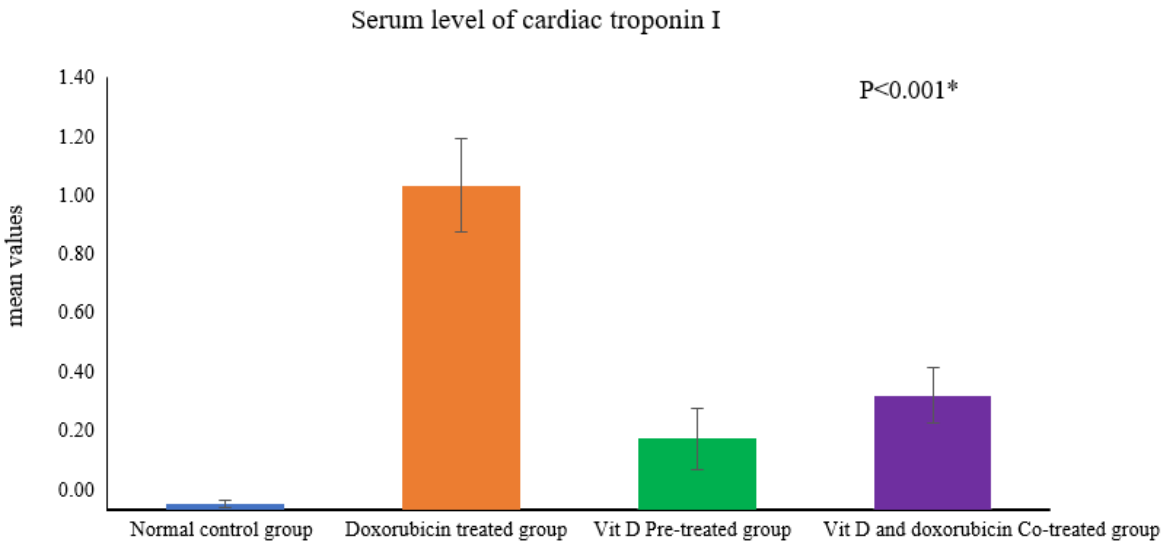

Figure 1. Distribution of Serum level of cardiac troponin I among the studied groups of rats**.**

While, malondialdehyde levels in cardiac tissue considerably higher in group 2 (9.70±0.82) compared to control group (6.09±0.27), group 3 (7.93±0.33) and group 4 (7.27±0.43), (P<0.001). Superoxide dismutase and glutathione levels in cardiac tissue considerably lower in group 2 compared to the normal control, group 3, and group 4 (P< 0.001) **(Table 4).**

Variable	Mean ±SD	Range	F	P value	95% CI	
					Lower	Upper
MDA in cardiac tissue (nmol/gprotein)						
Normal control group (N=7)	6.09±0.27	5.70-6.50	61.549	**<0.001***	5.83	6.34
Doxorubicin treated group (N=7)	9.70±0.82	8.70-11.10			8.94	10.46
Vit D Pre-treated group (N=7)	7.93±0.33	7.50-8.40			8.23	7.50
Vit D and doxorubicin Co-treated group (N=7)	7.27±0.43	6.80-8.10			6.80	8.10
Post Hoc P (Mean Difference)	*P1<0.001*(3.61), P2<0.001*(1.84), P3<0.001*(1.19), P4<0.001*(1.77), P5<0.001*(2.43), P6=0.024(0.66)*					
SOD in cardiac tissue (IU/mg protein)						
Normal control group (N=7)	106.14±6.44	98.00-115.00	84.575	**<0.001***	100.19	112.10
Doxorubicin treated group (N=7)	50.86±6.52	42.00-61.00			44.83	56.88
Vit D Pre-treated group (N=7)	77.14±7.73	66.00-89.00			69.99	84.30
Vit D and doxorubicin Co-treated group (N=7)	65.14±6.23	58.00-78.00			59.38	70.90
Post Hoc P (Mean Difference)	*P1<0.001*(55.29), P2<0.001*(29.00), P3<0.001*(41.00), P4<0.001*(26.29), P5=0.001(14.29), P6=0.003(12.00)*					
GSH in cardiac tissue (µg/g wet wt)						
Normal control group (N=7)	219.57±10.91	207.00-234.00	88.015	**<0.001***	209.48	229.66
Doxorubicin treated group (N=7)	145.00±7.59	137.00-157.00			137.98	152.02
Vit D Pre-treated group (N=7)	175.71±8.52	164.00-188.00			167.84	183.59
Vit D and doxorubicin Co-treated group (N=7)	160.86±8.84	152.00-178.00			152.68	169.03
Post Hoc P (Mean Difference)	*P1<0.001*(74.57), P2<0.001*(43.86), P3<0.001*(58.71), P4<0.001*(30.71), P5=0.003(15.86), P6=0.005 (14.86)*					

Table 4: Oxidative stress indicators in cardiac tissue among the studied groups of rats.

MDA: Malondialdehyde. **SOD:** Superoxide dismutase. **GSH:** Glutathione.
F: One way ANOVA test. *****Significant. **CI:** Confidence interval for Mean.
P1: normal control group compared doxorubicin treated group.
P2: normal control group compared vit D Pre-treated group.
P3: normal control group compared vit D and doxorubicin Co-treated group.
P4: doxorubicin treated group compared vit D Pre-treated group.
P5: doxorubicin treated group compared vit D and doxorubicin Co-treated group.
P6: vit D Pre-treated group compared vit D and doxorubicin Co-treated group.

Discussion

Multiple cancer types can be successfully treated with doxorubicin (Dox), including triple negative breast cancer (TNBC). However, the use of Dox is constrained because of its cardiotoxic adverse effects, which are caused by increase of oxidative stress **[16]**. Cardiomyopathy, a prolonged QT interval, a smaller QRS complex, a decrease left ventricular ejection fraction, and long delayed between Q and T waves are a few examples of abnormalities in the electrical representations of heartbeats on ECG that indicator of cardiotoxicity presence **[17]**.

Dox is converted into a reactive metabolite by the enzymes as NADPH quinone oxidoreductase (NQO1), antioxidant equilibrium in cells which is altered, and reactive oxygen species (ROS) **[18].**

Therefore, novel strategies have been explored to lower oxidative stress in Dox users and to improve treatment outcomes without compromising the anticancer potential. The use of antioxidants might to have no effect or might even modestly improve chemotherapy results **[19]**.

Reduced Dox-induced reactive oxygen species (ROS) levels may lessen cardiotoxic side effects during TNBC treatment, also use of antioxidants during chemotherapy may benefit healthy tissues and minimize the chance of cancer recurrence. Vitamin D and its active metabolites have an antioxidant effect by activating NRF2-dependent antioxidant signalling. This may inhibit the growth of cancerous cells **[20]**. So, this study was performed to see whether vitamin D may counteract the cardiotoxicity caused by doxorubicin.

In our study, serum calcium was considerably higher in doxorubicin-treated group compared to other studied groups. Dysregulation of intracellular Ca2+ homeostasis is one of the prominent mechanisms behind Dox's harmful effects on mitochondria. By inhibiting SERCA2a transcription, among other mechanisms by Dox affects Ca2+ homeostasis, Ca2+ uptake into the sarcoplasmic reticulum (SR) is hampered **[21]**. In this concern **Awad et al., [10]** found that Dox can bind to and activate the ryanodine receptor (RYR2), causing the release of Ca2+ from the SR into the cytosol. In fact, after Dox intoxication, both cytosolic and mitochondrial Ca2+ markedly increased. In the past, the overabundance of Ca2+ in both the cytoplasm and mitochondria contributed to the cardiotoxicity of Dox **[22]**.

A recent study by Lee et al. (1) reported that, low vitamin D supplementation may decrease cardiotoxic adverse effects during Dox treatment without lowering Dox efficacy. Dietary vitamin D supplementation increased plasma vitamin D levels. Mice exposed to Dox (10 mg/kg) developed cardiotoxicity after receiving two weekly injections, despite the fact that vitamin D treatment prevented cardiotoxic adverse effects in vivo as demonstrated by ECG markers such as ejection percent, stroke volume, and fractional shortening. Additionally, 4-hydroxynonenal (4-HNE) and C-Myc induced compensated cardiac hypertrophy (C-MYC) levels were decreased, also rats receiving vitamin D with Dox (10mg/kg) had much greater survival rates compared those receiving Dox (10 mg/kg) alone. **[23]**.

The higher levels of 4-HNE, NADPH and quinone acceptor oxidoreductase (NQO) expression were consistent with elevated cardiac ROS levels induced by 10 mg/kg Dox. In contrast to rats just receiving Dox (10 mg/kg), and those received vit D had reduced levels of 4- HNE and NQO1, which may prevent 10 mg/kg Dox from producing ROS. NQO1 suppression has been shown to boost sensitivity to chemotherapeutic medicines like Dox **[24].** After vitamin D administration, there was an increase in the phosphorylation of DRP1 at S637, a hallmark for mitochondrial fission inhibition. The stop of treatment with vitamin D, the Dox-induced increase in C-MYC expression in cardiac tissue, supporting earlier research that showed vitamin D can reduce C-MYC in cancer models **[25]**. The oxidation of polyunsaturated fatty acids by ROS to generate 4-HNE in combination with dox-induced C-MYC expression may make cardiac dysfunction worse.

In the study by Lee et al., (1), vitamin D improved cardiac function without interfering with the effect of 10 mg/kg Dox on the reduction in TNBC tumor volume. Rats that received vitamin D supplements had improved cardiac health, greater survival rates, and higher body weights than Dox-treated mice who didn't get vit D. Rats receiving vit D supplements also saw greater declines in heart function and survival. Mice administered Dox with 6 mg/kg showed less cardiac adverse effects, but given vit D supplements, their tumors were reduced. The amount of vit D used in this study (10,000 IU/kg) is far lower than typical vit D supplement dosages; a 75 kg adult would receive 662 IU **[26]**.

The serum cardiac troponin I concentration in the current study was significantly higher in the doxorubicin-treated group than in the studied group. In a study by **Reagan et al. [27]** discovered that chronic doxorubicin-induced cardiotoxicity resulted in cardiac damage at all dosages (1, 2, or 3 mg/kg/wk.). Both the incidence and the mean magnitude of cardiac troponin (cTn) signals frequently increased with increasing dose and/or longer treatment duration. The cTnT and cTnI signals correlated in the samples under investigation. Serum cTn results following a 2- or 4-week recovery period also demonstrated the continuation of cardiac injury beyond the treatment period, which consistent with the established aetiology of doxorubicin cardiomyopathy. This finding was consistent with **Chaudhari et al**. **[28]** finding GO and KEGG pathway analyses demonstrated that Dox exposure lowered the expression of genes involved in cardiac contraction and pathways related with cardiomyopathies in a preferential way. In contrast to these different gene-specific effects, exposed cardiomyocytes' cTnT and cTnI proteins underwent significant, long-lasting changes in their shape and topographical organization. The most notable ones were cytoplasmic rearrangements of cTns, where the usual bundling patterns of the cell were changed to a random accumulation that persisted throughout the entire trial. Additionally, problems with cells' contractility and beating patterns. Similar alterations in human cardiomyocytes treated with Dox have been previously reported, however without a thorough examination of the morphology of cTns. Overall, these data indicates that Dox-induced morphological and maybe functional changes in human cardiomyocytes have long-lasting effect and as a result of their persistence, lead to their delayed malfunctioning **[39]**.

In the present study, malondialdehyde levels in cardiac tissue were considerably higher in the doxorubicin-treated group compared to other groups. While, the doxorubicin-treated group had significantly lower levels of superoxide dismutase and glutathione in comparison to other groups. **Zhao et al. [30]** has also shown a connection between myocardial oxidative stress and the underlying mechanisms of Dox-induced cardiotoxicity. During evolution, cells create an inherent antioxidant system to reduce ROS levels and boost cell viability. GSH, GSH-Px, and SOD are the most prominent antioxidant enzymes. The elevation of SOD, GSH, and GSH-Px levels in vivo and the downregulation of ROS levels in vitro demonstrated that Dossin protected Dox-induced oxidative damage to the myocardium **[31]**. In addition, Dossin significantly reduced MDA, an end-product of lipid hydroperoxide as well as one biomarker. On the same line, **Alam et al.**, **[32]** found that, the reduced amount of GSH in the heart was a result of Dox treatment. Groups C and D that received thymoquinone experienced a drop in MDA level and an increase in GSH level. GSH deficiency may impede cell defence, which could result in tissue damage.

Conclusion

The current results demonstrate that vitamin D supplementation in the diet, whether mild or moderate, was cardioprotective and didn't interfere with Dox ability to treat cancer in rats. It is reasonable to conduct more research to determine the ideal vitamin D dosages required to enhance heart functions without reducing Dox effectiveness.

References

1. Lee KJ, Wright G, Bryant H, Wiggins LA, Dal Zotto VL, Schuler M, Malozzi C, Cohen MV, Gassman NR. (2021). Cytoprotective effect of vitamin d on doxorubicin-induced

cardiac toxicity in triple negative breast cancer. International journal of molecular sciences. Jul 12;22(14):7439.

2. Gül SS, Aygün H. (2018). Cardioprotective effect of vitamin D and melatonin on doxorubicin-induced cardiotoxicity in rat model: an electrocardiographic, scintigraphic and biochemical study. The European Research Journal.
3. Podyacheva EY, Kushnareva EA, Karpov AA, Toropova YG. (2021). Analysis of models of doxorubicin-induced cardiomyopathy in rats and mice. A modern view from the perspective of the pathophysiologist and the clinician. Frontiers in pharmacology.Jun 3;12:670479.
4. Vaidya A, Forman JP. (2012). Vitamin D and vascular disease: the current and future status of vitamin D therapy in hypertension and kidney disease. Current hypertension reports. Apr;14(2):111-119.
5. Gardner DG, Chen S, Glenn DJ.(2013). Vitamin D and the heart. American Journal of Physiology-Regulatory, Integrative and Comparative Physiology. Nov 1; 305(9):R969-977.
6. Manucha W, Juncos LI. (2017). The protective role of vitamin D on the heart and the kidney. Therapeutic Advances in Cardiovascular Disease. Jan;11(1):12-19.
7. Sergazy S, Shulgau Z, Fedotovskikh G, Chulenbayeva L, Nurgozhina A, Nurgaziyev M, Krivyh E, Kamyshanskiy Y, Kushugulova A, Gulyayev A, Aljofan M. (2020). Cardioprotective effect of grape polyphenol extract against doxorubicin induced cardiotoxicity. Scientific reports. Sep 7;10(1):1-2.
8. Todorova VK, Beggs ML, Delongchamp RR, Dhakal I, Makhoul I, Wei JY, Klimberg VS. (2012). Transcriptome profiling of peripheral blood cells identifies potential biomarkers for doxorubicin cardiotoxicity in a rat model. PloS one. Nov 27; 7(11):e48398.
9. Godinho AF, Trombini TV, Oliveira EC. Effects of elevated calcium on motor and exploratory activities of rats. Brazilian journal of medical and biological research. 2002; 35:451-457.
10. Awad HH, El-Derany MO, Mantawy EM, Michel HE, Mona M, El-Din RA, El-Brairy AI, El-Demerdash E.(2021). Comparative study on beneficial effects of vitamins B and D in attenuating doxorubicin induced cardiotoxicity in rats: Emphasis on calcium homeostasis. Biomedicine & Pharmacotherapy. Aug 1; 140:111679.
11. Irvine RJ, White J, Chan R.(1997). The influence of restraint on blood pressure in the rat. Journal of pharmacological and toxicological methods. Nov 1;38(3):157-162.
12. Kamel MM, El-Farouk LO, Osman AS, Khorshid OA, Shabrawy-Abdo ME. Comparative study of the protective effect of metformin and sitagliptin against doxorubicin-induced cardiotoxicity in rats. Clinical Pharmacology and Biopharmaceutics. 2017; 6:174.
13. Mohamed AS, Hosney M, Bassiony H, Hassanein SS, Soliman AM, Fahmy SR, Gaafar K. (2020). Sodium pentobarbital dosages for exsanguination affect biochemical, molecular and histological measurements in rats. Scientific Reports. Jan 15;10(1):1-3.
14. Zaafan MA, Zaki HF, El-Brairy AI, Kenawy SA.(2013). Protective effects of atorvastatin and quercetin on isoprenaline-induced myocardial infarction in rats. Bulletin of Faculty of Pharmacy, Cairo University. Jun 1;51(1):35-41.
15. Zhao L, Tao X, Qi Y, Xu L, Yin L, Peng J. (2018). Protective effect of dioscin against doxorubicin-induced cardiotoxicity via adjusting microRNA-140-5p-mediated myocardial oxidative stress. Redox biology. Jun 1; 16:189-198.
16. Cappetta D, De Angelis A, Sapio L, Prezioso L, Illiano M, Quaini F, Rossi F, Berrino L, Naviglio S, Urbanek K.(2017). Oxidative stress and cellular response to doxorubicin: a common factor in the complex milieu of anthracycline cardiotoxicity. Oxidative medicine and cellular longevity. Oct 18;2017.
17. Coppola C, Rienzo A, Piscopo G, Barbieri A, Arra C, Maurea N.(2018). Management of QT prolongation induced by anti-cancer drugs: target therapy and old agents. Different algorithms for different drugs. Cancer treatment reviews. Feb 1;63:135-143.
18. Singh K, Bhori M, Kasu YA, Bhat G, Marar T.(2018). Antioxidants as precision weapons in war against cancer chemotherapy induced toxicity–Exploring the armoury of obscurity. Saudi Pharmaceutical Journal. Feb 1;26(2):177-190.
19. Nakai K, Fujii H, Kono K, Goto S, Kitazawa R, Kitazawa S, Hirata M, Shinohara M, Fukagawa M, Nishi S.(2014). Vitamin D activates the Nrf2-Keap1 antioxidant pathway and ameliorates nephropathy in diabetic rats. American journal of hypertension. Apr 1;27(4):586-595.
20. Gorini S, De Angelis A, Berrino L, Malara N, Rosano G, Ferraro E.(2018). Chemotherapeutic drugs and mitochondrial dysfunction: focus on doxorubicin, trastuzumab, and sunitinib. Oxidative medicine and cellular longevity. Oct;2018.
21. Agustini FD, Arozal W, Louisa M, Siswanto S, Soetikno V, Nafrialdi N, Suyatna F.(2016). Cardioprotection mechanism of mangiferin on doxorubicin-induced rats: focus on intracellular calcium regulation. Pharmaceutical Biology. Jul 2;54(7):1289-1297.
22. Zeekpudsa P, Kukongviriyapan V, Senggunprai L, Sripa B, Prawan A.(2014). Suppression of NAD (P) H-quinone oxidoreductase 1 enhanced the susceptibility of cholangiocarcinoma cells to chemotherapeutic agents. Journal of Experimental & Clinical Cancer Research. Dec;33(1):1-3.
23. Willems PH, Rossignol R, Dieteren CE, Murphy MP, Koopman WJ.(2015). Redox homeostasis and mitochondrial dynamics. Cell metabolism. Aug 4;22(2):207-218.
24. Salehi-Tabar R, Nguyen-Yamamoto L, Tavera-Mendoza LE, Quail T, Dimitrov V, An BS, Glass L, Goltzman D, White JH.(2012). Vitamin D receptor as a master regulator of the c-MYC/MXD1 network. Proceedings of the National Academy of Sciences. Nov 13;109(46):18827-18832.
25. Pludowski P, Holick MF, Grant WB, Konstantynowicz J, Mascarenhas MR, Haq A, Povoroznyuk V, Balatska N, Barbosa AP, Karonova T, Rudenka E.(2018). Vitamin D supplementation guidelines. The Journal of steroid biochemistry and molecular biology. Jan 1; 175:125-135.
26. Rainville C, Khan Y, Tisman G.(2009). Triple negative breast cancer patients presenting with low serum vitamin D levels: a case series. Cases journal. Dec;2(1):1-5.
27. Reagan WJ, York M, Berridge B, Schultze E, Walker D, Pettit S.(2013). Comparison of cardiac troponin I and T, including the evaluation of an ultrasensitive assay, as indicators of doxorubicin-induced cardiotoxicity. Toxicologic pathology. Dec;41(8):1146-1158.
28. Chaudhari U, Nemade H, Gaspar JA, Hescheler J, Hengstler JG, Sachinidis A. (2016). MicroRNAs as early toxicity signatures of doxorubicin in human-induced pluripotent stem cell-derived cardiomyocytes. Archives of toxicology. Dec;90(12):3087-3098.
29. Zhang YW, Shi J, Li YJ, Wei L. (2009). Cardiomyocyte death in doxorubicin-induced cardiotoxicity. Archivum immunologiae et therapiae experimentalis. Dec;57(6):435-445.
30. Zhao L, Tao X, Qi Y, Xu L, Yin L, Peng J. (2018). Protective effect of dioscin against doxorubicin-induced cardiotoxicity via adjusting microRNA-140-5p-mediated myocardial oxidative stress. Redox biology. Jun 1; 16:189-198.

31. Maejima Y., Kuroda J., Matsushima S., Ago T., Sadoshima J.(2011). Regulation of myocardial growth and death by NADPH oxidase. J. Mol. Cell Cardiol. 50:408–416.
32. Alam MF, Khan G, Safhi MM, Alshahrani S, Siddiqui R, Sivagurunathan Moni S, Anwer T. (2018). Thymoquinone ameliorates doxorubicin-induced cardiotoxicity in swiss albino mice by modulating oxidative damage and cellular inflammation. Cardiology Research and Practice. Apr 1;2018.

DOI: 10.31579/2688-7517/132