Physiological and Histological Studies on The Heart of Male Albino Rats Exposed to Electromagnetic Field and The Protective Role of Silymarin and/orVitamin E.

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ABSTRACT

Aim of the work:This study aimed to determine the ameliorative effect of silymarin (SIL) and\or vitamin E (Vit.E) against changes induced by mobile phone radiation in the heart of male albino rats. **Matrerial and methods**:Total of 48 adult male albino rats were assigned for this study. The 1st group served as control (n=6), the 2nd group exposed to mobile phone generator radiation (900MHz) for 2hr/day 3days/week for two months, 3rd group (+ve control) supplemented with SIL, 4th group (+ve control) supplemented with Vit. E, 5th group (+ve control) supplemented with SIL and Vit.E, 6th group: exposed group supplemented with SIL, 7th group: exposed group supplemented with Vit.E and 8th group exposed group supplemented with SIL and Vit.E. Physiological ,histopathological and histochemical changes were studied.**Results**: Exposure to mobile phone causes increases in activities of CPK, CK-MB and LDH enzymes in serum and heart tissue and oxidative stress markers (MDA and H_2O_2),while antioxidants (CAT and GSH) were decreased in the heart tissue. Sodium (Na) and calcium (Ca) levels were decreased While, K level showed non-significant change in serum. Numerous histopathological changes were detected in the heart tissue of rats of the irradiated group with altered collagen fibres, polysaccharides in the cardiac muscle fibres of the exposed group. These changes manifested good amelioration in the exposed groups that supplemented with SIL and/or Vit.E.

Conclusion: Treatment of rats with SIL and/or Vit.E ameliorated the dangerous effect of mobile phone radiation occurred in the cardiac muscle fibres.

Key words: Mobile phone radiation - Albino rats - Heart -Silymarin - Vitamin E.

Introduction

Since mobile phones are generally held and used close to the body, they are considered as the main source of electromagnetic radiation (EMR) that any person is exposed to it. In fact, the whole body could act as an efficient antenna for absorption of EMR. Thus, the signals transmitted by a cell phone can reach all parts of the body and penetrate into the living tissues and influence the body at the cellular level.[1] It is possible to say that the deleterious effects of electromagnetic microwaves are generally exerted through elevation of body temperature [2,] creation of free radicals [3]and disruption of oxidant /antioxidant balance in various tissues exposed to mobile phone (MP) that has been shown in the experimental studies.[3]

The circulatory system is considered as the main connecting and feeding system of the body tissues, it is very sensitive and any malfunction of this system can create disturbances in all the body organs.[4]

Mobile phone induced heart tissue damage, this damage may be due to the mobile phones which are used in close proximity to the heart and therefore, EM radiation emitting mobile phone may be absorbed by the heart.[5]

Silymarin is found in the seeds of *Silybium marianum.* Silymarin is a standardized mixture of antioxidant flavonolignans. It is a free radical scavenger and a membrane stabilizer that prevents lipidperoxidation.[6]

El Banna *et al.*[7]demonstrated that extract of *Silybium marianum* have antioxidant activity, as it increases the activities of antioxidant enzymes superoxide dismutase (SOD), CAT and GSH.

Rao and Viswanath[8] reported that SIL protected the endogenous antioxidant enzymes, suppressed the neutrophil infiltration during ischemia-reperfusion and limited the infarct size in the heart, with concomitant reduction in serum and heart tissue MDA. Pretreatment

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with SIL also protected rat hearts from a further drop in mean arterial blood pressure during reperfusion and restored heart rate.

The protection by Vit.E may be due to the reduction of lipid peroxidation.Thus, Vit.E inhibited the EMF-induced tissue damage and supported the hypothesis that superoxide radicals are involved in its pathogenesis.Also, Vit.E caused significant increases of antioxidant enzymes which decreased in EMF exposed animals[9] thereby preventing lipid oxidation (LPO) and the initiation of oxidative tissue damage.[10.]

In biological systems affected by EMR, the mechanisms of tissues damage were thought to involve reactive oxygen species (ROS). With excessive free radical production and the resulting consumption of antioxidant, endogenous defense mechanisms could become insufficient. These free radicals lead to damage of large cellular molecules such as lipids, proteins and nucleic acid.[11]

Material and methods

Experimental design

A total of 48 male albino rats about 8-10 weeks old and weighing 110–120 grams. They were maintained on standard normal diet and water *ad libitum.*The animals were housed in clean cages and maintained under controlled conditions of temperature (25 ±1.5ºC) , light (12 hours light: 12 hours dark) and good ventilation.

Exposure system and application of electromagnetic field (EMF)

Albino rats (n=48) were divided into eight groups (n=6) .G1- the control group; G2- EMF group: rats exposed to 900 MHz (2hrs/day, 3times/week) for 2 months. G3- positive control received SIL only 70mg/kg b.wt. G4- positive control received Vit. E only 60mg/kgb.wt. G5-positive control received SIL and Vit. E. G 6- irradiated group plus SIL. G7- irradiated group plus Vit.E. G8- irradiated group plus SIL and Vit.E.

A specially designated electromagnetic field (EMF) exposure system, with a round plastic tube cage and a dipole exposure antenna was used and it produced EMF equals to mobile phone radiation with frequency equals 900 MHz[12] , with a specific absorption rate (SAR) equals 1.2 w/kg and constant power about 1.4 mW/cm (Holiday Industries Inc., UK).

Chemicals

Vitamin E is used as alpha-tocopherol acetate. It is purchased from Sigma Company, given orally at a dose level of 60 mg/kg. b.wt dissolved in corn oil.[13]

Silymarin purchased from South-Egypt Drug Industries Company (SEDICO) was given orally by gastric tube in a dose of 70 mg/kg.b.wt[14] dissolved in distilled water.

Preparation of samples and biochemical analysis:

Animals of all groups were sacrificed at the end of the experiment. Blood samples were collected, serum obtained by centrifugation at 3000 rpm for 10 min. For the assessment of heart enzymes as creatine phosphokinase (CPK)[15] Creatine kinase (CK-MB) [16.] Lactate dehydrogenase (LDH). [17] and some electrolytes as Sodium (Na), Potassium (K) and calcium (Ca). Part of heart tissue was homogenized in 0.1M phosphate buffer and centrifuged at 4000 rpm for 15 min in a cooling centrifuge and the supernatant was pipetted into plastic tubes for determination antioxidants such as catalase (CAT) enzyme,[18] Glutathione (GSH).[19] Oxidative stress biomarkers, lipid peroxidation was evaluated by measuring malondialdhyde (MDA) [20]and hydrogen peroxide (H_2O_2).[21]

Histological and histochemical techniques

Cardiac muscle fibres were immediately excised and fixed in 10% neutral formalin. Paraffin sections (5µm in thickness) were prepared for processing the histological and histochemical studies. For general histology, sections were stained with Harris' haematoxylin and eosin.[22] polysaccharides were detected by using periodic acid Schiff's (PAS) reagent[23] and collagen fibres were stained by using Mallory's trichrome stain.[24]

Statistical analyses

Data are represented as means, standard error of (SE) percentage of change. Significant differences between the mean values were statistically analyzed using simple one way analysis of variance **(SPSS program, version 16, Duncan'smultiple range test)** for the

significant interrelation between the various groups.[25]

RESULTS

Tables (1and 2) showed that SIL and/or Vit.E didn't affect CPK, CK-MB and LDH activities in serum and heart tissue in comparison with the normal control. It was obvious that CPK,CK-MB and LDH activities in serum and heart tissue increased significantly ($P<0.05$) in rats exposed to EMF for two months in comparison with the normal control group.However, the enzyme activities decreased in the irradiated groups supplemented with SIL and/or Vit.E in comparison with that obtained in the irradiated group.

Table (3) showed that SIL and/or Vit.E didn't affect the CAT enzyme activity, GSH, MDA and H_2O_2contents in heart tissue. The CAT activity and GSH content decreased significantly ($P<0.05$) in rats exposed to EMF for two months in comparison with the normal control group.Whereas, CAT activity and GSH contents in heart tissue increased in the irradiated group supplemented with SIL and/or Vit.E in comparison with the irradiated group. While, MDA and H_2O_2 increased in heart tissue of rats that exposed to EMF when compared to the control group. Administration of SIL and/or Vit.E to the exposed rats manifested good amelioration in CAT activity, GSH, MDA and H_2O_2 contents.

The exposed group to EMF showed a significant low Na and Ca levels in serum.While, K level showed non-significant change in serum. The exposed group supplemented with SIL and/or Vit.E investigated good amelioration in the levels of these parameters (Table 4).

1-The histopatological study

Several histopathological changes were observed in heart tissue of the irradiated group (R); these changes include: distorted cardiac muscle fibres with deeply stained nuclei (pyknotic) (P) and highly thickened and elongated arterial wall which contained haemolysed blood cells (Figure 2). Somewhat normal appearance of heart tissue can be detected in the exposed group that supplemented with SIL (RS) (Figure3). Irregular distribution of nuclei of myocytes of cardiac tissue appeared in the exposed group supplemented with Vit.E (RE) (Figure 4). Somewhat normal appearance of cardiac muscle fibres was realized due to co-administration of SIL and Vit.E to the exposed group (RSE) (Figure 5).

Collagen fibres: normal distribution of collagen fibres was observed in heart tissue of a rat of the control group (Figure 6). Highly increased collagen bundles were observed in the highly distorted cardiac tissue of R group (Figure 7). Somewhat normal appearance of collagen fibres in the cardiac muscle of groups RS, RE and RSE (Figures 8,9,10).

2-The histochemical changes

Polysaccharides-Normal distribution of PAS +ve materials was detected in heart tissue of a rat of the control group (Figure 11). Depleted PAS +ve materials were demonstrated in the ruptured cardiac muscle fibres (➔) of a rat of group R. Highly widened endomysium were negatively stained (Figure 12), moderately stained PAS +ve materials were noticed in the cardiac muscle fibres of a rat of R group, but elongated and thickened arterial wall acquired deep staining affinity (Figure 13). Somewhat normal content of PAS+ve materials were detected in groups RS, RE and RSE **(**Figures14,15,16).

DISCUSSION

Exposure to a magnetic field poses a risk for cardiovascular morbidity and mortality.[26] Among workers in the electrical industry, statistically significant differences were found between magnetic field exposure and arrhythmia-dependent and acute myocardial infarcts-dependant death.[27]

With regard to the effect of EMF on the cardiovascular system, EMF might interfere with work of cardiac pacemakers and other implantable medical devices like cardioverter defibrillators.[28] Mobile phones were reported to cause a rise of blood pressure of 5-10 mm Hg each time of exposure and it was suggested that mobile phone could induce constrictive effect on the blood vessels.[29] Moreover, increased fetal and neonatal heart rate and decreased cardiac output were found during subjecting pregnant women to mobile phones.[30]

Cardiac markers are biomarkers measured to evaluate heart function. They are often discussed in the context of myocardial infarction, but other conditions can lead to an elevation in the cardiac marker level. Most of the early markers identified were enzymes and as a result, the term "cardiac enzymes" is sometimes used.[31.]

The present study showed that CPK, CK-MB and LDH enzyme activities increased in the heart tissue in EMF exposed group. However, the enzyme activities manifested good amelioration in the exposed group that supplemented with SIL and/or Vit.E.

Development of various heart diseases and daily exposure with EMF hypothesized an association between exposure to the magnetic fields and acute cardiovascular disease (CVD).[32]They demonstrated that the mobile phone may influence heart rate variability by changing autonomic balance.

Lipid peroxidation was believed to be an important cause of destruction and damage to cell membranes and has been shown to be a contributing factor to the development of oxygen radicals-mediated tissue damage. Oxidative stress in the heart tissue was associated with a significant increase in the activity of CPK and LDH common characteristic toxicity released to the blood stream from damaged hear tissue.[33]

The present study revealed that the exposed group of rats that supplemented with SIL and/or Vit.E showed good amelioration in the levels of CPK, CK-MB and LDH in heart tissue. Since the activities of these enzymes nearly returned to the normal levels compared to the control group.

Indeed, many antioxidative plants and their isolated components have been reported to possess cardioprotective activity in the experimental models of myocardial ischemia-reperfusion injury.[34] Oxidative stress is one of the mechanisms implicated in the pathogenesis of ischemic-reperfusion injury. There were comprehensive experimental and clinical evidences that antioxidants attenuate myocardial infarction.[35]

It have been reported that SIL had a cardioprotective activity noticed in ameliorating the activities of serum marker enzymes, ie, LDH, CK-MB and CPK against myocardial ischemia-reperfusion-induced myocardial injury in rats.[8] The protective effect of this extract may be attributed to presence of flavonoid compounds and their antioxidant effects and free radical scavenging properties.[6]

Vitamin E is considered as a mean of correcting plasma antioxidant status and attenuating the cardiovascular disease that accompanied kidney failure.[36]

Vitamin E could prevent or delay coronary heart disease (CHD) which comes from several sources. An *in vitro* study have found thatVit.E inhibits oxidation of low-density lipoprotein (LDL) cholesterol.[37] They added that Vit.E might also help to prevent the formation of blood clots that could lead to a heart attack or venous thromboembolism .

Antioxidants and free radicals

Results of the present study showed a significant decrease in CAT activity and GSH levels in heart tissues of the EMF (900MHz) exposed group. However, MDA and H_2O_2 were increased in heart tissues of the same group. Supplementation of SIL and/or Vit.E to exposed rats showed good amelioration in the levels of these parameters.

Reduced GSH as well as parallel elevation in MDA in this study may be due to oxidative stress and impairment of antioxidant defense mechanisms in EMF exposed rat[38] attributed the elevated utilization of antioxidant system as an attempt to detoxify the generated free radical by radiation. The depletion in GSH levels after exposure to EMF radiation noted in the present study in the blood serum, liver and heart tissues may be due to the reaction of GSH with free radicals resulting in the formation of GSSG.[39] Moreover, the availability of GSH can also be limited by deficiency in synthesis, enhanced efflux, or inefficient reduction of GSSG. In the normal condition, GSH is restored by synthesis, but in the irradiated animals, normal synthesis and/or repair is disrupted due to damage to DNA and membranes.[40]

El Banna*et al.* [7] demonstrated that extract of *Silybium marianum* have a significant hepatoprotective and antioxidant activity and may be useful for patients who suffer from

liver diseases, as it increases the activities of antioxidant enzyme SOD, CAT and GSH levels. The protective effect of this extract may be due to flavonoid compounds and their antioxidant effects and free radical scavenging properties.

The present data are agree with the result of **Abd El Rahman *et al.***[41] who reported that SIL,Vit.E and their co-administration in rats exposed to EMF (900 MHz) for 2 months showed increase in oxidative stress identified by increases in serum MDA, associated with decreases in SOD and CAT activities and GSH content.

Electrolytes

Electrolytes are dissolved in different compartments of body fluid including the serum of the blood, inside the cells (intracellular) and outside the cells (extracellular).[42]

Increased serum potassium levels in the present results is supported by the work done by **Dindic *et al.*** [43] who demonstrated that wistar rats exposed to EMF by a mobile phone (900MHz) 2h for 3 months showed high serum potassium concentration in comparison with the control group. Also, agree with the work done by **Sokolovic*et al.*** [44] who stated that serum potassium concentration was significantly higher in the exposed rats to 900MHz for 3 months, while the concentration of sodium did not differ in the exposed group. The authors stated that hyperkalemia could be the possible systemic marker of impaired cells membrane fluidity and increased permeability.

Increased potassium serum concentration in EMF-exposed rats may be an indicator of cellular membranes damage. Probably, this is the consequence of increased membrane per meability and potassium leaking, induced by oxidative damage or by impaired function of ions channels.[45]

The observed hypocalcaemia associated with exposure to EMF occurred as a result of alteration of intracellular signaling pathways resulted from radiofrequency (RF) radiation exposure through changes in Ca^{2+} permeability across cell membranes.[46]It has been reported that calcium positive ions strengthen cell membranes because they bind the negatively-charged phospholipid molecules and that electromagnetic radiation could lead to the replacement of calcium with monovalent ions that weakens the membrane and makes it more likely to tear and form pores.[47] Thus, the observed hypocalcemia in this study might be one of the mechanisms by which EMF interacts with the biological tissues.

The relationship between intracellular Ca^{2+} and the oxidative stress may be a complex process. Oxidative stress may change cytosolic Ca^{2+} concentration in cells that subsequently activate further production of ROS.[48]RF-EMW may also alter intracellular calcium homeostasis by acting on plasma membrane calcium channels.[49]

The changes in serum Na^{+}, K^{+} and Ca^{++} can be explained by radiofrequency radiation (RF) which might alter intracellular signaling pathways through changes in ionic distribution and membrane fluidity or change Ca^{2+} permeability across cell membranes.[50] RF could also alter the conformational energy of glycoproteins in the cell membrane to open Ca^{2+} channels.[51]These changes could cause pathophysiological changes in the brain such as tumorigenesis ansd neural degeneration. Also, **Mortazavi *et al.***[52]reported that RF radiation from mobile phones could alter intracellular signaling pathways through changes in Ca^{++} permeability across cell membranes and cellular calcium levels**.**

Administered of *Silybum marianum* oil (SMO) orally which is rich in essential fatty acids, phospholipids, sterols, and Vit.E for 7 weeks, significantly attenuated the D-galactose induced liver mitochondrial dysfunction by improving the activities of Na^{+}-K^{+}-ATPase, Ca^{2+}-Mg^{2+}-ATPase, membrane potential and membrane fluidity.[53]

Histopathological and histochemical studies.

The histopathological and histochemical changes in the heart tissue

In the present study photomicrographs showed distorted cardiac muscle fibres of the irradiated rats with deeply stained nuclei (pyknotic) and highly thickened and elongated arterial wall which contained haemolysed RBCs in the exposed rat group.

The present results come in agreement with the work of **Khaki andKhaki**[54] who

demonstrated that heart ventricular sections from rat group that exposed to EMF of 50 Hz showed increased dark brown stained muscle fiber nuclei.Also, with the work done by **Mohamed and Emam**[55]who showed highly widened endomysium and degenerated muscle fibres with loss of striations with bizarre distribution of nuclei in mother's heart of irradiated pregnant rats that exposed to 2Gy gamma rays in the 7^{th} or 14^{th} day of gestation.

The pyramidal cells in the brain of newly born mice exposed to mobile phone radiation 900-1800 MHz for 4 months underwent an obvious phase of degeneration, haemorrhage, gliosis, spread of apoptotic cells and increased vacuolization.[56]

Results of the present study come in disagreement with the opinion of [27]**Soker*et al*.** who exposed rats for 14 days 3 hr/ day to 2.5 gauss levels. They did not observe any differences in the cardiac muscles of the control and irradiated group by using electromicrographs.

Degenerated areas in the cardiac tissue observed in the present study could be considered as a reactive change that may be related to the inhibitory effect on the vascular smooth muscles which induced relaxation and consequent vasodilatation. This result is supported by **Melamed *et al*.**[57] who reported that this vasodilatation and increased vascular permeability should lead to loss of fluid from the blood. So, the vessels were engorged with blood cells with consequent slowing down of the blood stream which would result in degeneration and necrosis in the cardiac tissues.

Damaged cardiac tissue observed in the present study may be due to increased lipid peroxidation (MDA) and decreased GSH and CAT levels. In this respect **El-Habit *et al*.** [58] and **Saada and Azab**[59] reported that the histological damage might result from an increase in the process of lipid peroxidation and a decrease in the activity of antioxidant enzymes with the consequent damage of cellular membranes.

The apoptotic cell death plays a pivotal role in the development of heart failure.[60]They used genetically modified mice and clearly indicated a direct, causal relation between levels of apoptosis and the progression towards advanced heart failure

In the present study administration of SIL to the exposed group showed somewhat normal appearance of the cardiac tissue, but nuclei of some myocytes were hypertrophied. Administration of Vit.E showed irregular distribution of nuclei of myocytes of cardiac tissue. However, co-administration of SIL and Vit.E showed somewhat normal appearance of the cardiac muscle fibres.

Collagen fibres

The present study showed highly increased collagen bundles in the distorted cardiac tissue of the exposed group.

The present investigation is supported by the work done by **Mohamed and Emam**[55] who detected increased collagen fibres in most of cardiac muscle tissues of the pregnant rats exposed to 2Gy γ-rays on day 7 or day 14 of gestation when compared to the control group.

Increased collagen fibres post-irradiation were detected in the different tissues as described by several authors.[61] **George *et al*.** [62] suggested that decreased synthesis of collagenolytic enzymes might contribute to further accumulation of collagen.

Treatment of rat with SIL and/or Vit.E showed somewhat normal appearance of collagen fibres in the cardiac muscle fibres.

The present results come in agreement with the work done by **Mohamed**[63] who reported that SIL and Vit.E could ameliorating the increased collaged fibres in the lung tissue of rats that exposed to mobile phone radiation 900MHz.

Polysaccharides

The present study investigated depleted PAS +ve materials in the ruptured cardiac muscle fibers of rats of the irradiated group with highly widened endomysium which were negatively stained. .

The present investigation comes in agreement with the result of **Mohamed and Emam.**[55] They revealed that reduced staining affinity of PAS +ve materials was detected in the maternal cardiac tissue of pregnant rats that exposed to 2Gy of γ-rays on day 7 or day 14 of gestation when compared to the control group.

Decreased PAS +ve materials may be due to failure of the tissue to synthesize or store

glycogen and may be also a result of degeneration observed in the endomysium. Decreased glycogen content post-irradiation exposure was noticed in studies of many authors.[64]

However, results of the present investigation come in disagreement with the result of **Gorczynska and Wegrzynowicz**[65] who noticed increased glycogen content in the tissues post-irradiation. They stated that this increase in PAS+ materials may be due to increased cortisol which usually leads to an accumulation of glycogen in the tissue**.**

The present result comes in disagreement with the result of **Mohamed**[63] who found high increase of the PAS +ve materials in the thickened alveolar septa and walls of the bronchioles and blood vessels in the lung tissue of rats exposed to 900MHZ for 2 months.

In the present study treatment of the exposed group with SIL and/or Vit.E showed somewhat normal content of PAS +ve materials in cardiac muscle fibres.

Administration of SIL increased the cytoplasmic glycogen granules compared to cisplatin group, where granules appeared moderately stained. However, the marked reduction in glycogen stores induced by CDDP was improved after pretreatment with SIL.[66]

Silymarin and Vit.E ameliorated the increase in PAS+ materials that appeared in lung of rats that exposed to EMF 900 MHz.[63]

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Table (1): Serum CPK, CK-MB and LDH enzymes activities in the control and different treated groups.

Groups / Parameters	Normal control	Positive control			Treatment			
		SIL	Vit.E	SIL+ Vit.E	Irrad.	Irradi.+ SIL	Irrad.+ Vit.E	Irrad.+ SIL + Vit.E
CPK) (U/L)	**68.4± 3.80**	**65.36± 3.91**	**64.95± 3.99**	**62.86± 3.87**	**123.70± 3.83a**	**77.80± 3.60b**	**74.50± 3.66b**	**72.30± 3.83b**
% of changes vs normal cont. % of changes vsirrad.group		-4.44	-5.04	-8.10	+80.85	+13.74 -37.11	+8.92 - 39.77	+5.70 - 41.55
CK-MB (U/L)	**28.70± 1.40**	**24.67± 1.50**	**26.41± 1.46**	**25.6± 1.44**	**60.41± 1.60a**	**36.37± 1.43ab**	**39.95± 1.51ab**	**32.40± 1.52b**
% of changes vs normal cont. % of changes vsirrad.group		-14.00	-7.98	-10.80	+110.4	+26.72 -39.79	+39.2 -33.87	+12.89 -46.37
LDH(U/L)	**216.33± 4.90**	**215.14 .±4.82**	**211.2± 4.78**	**213.8± 4.84**	**318.60± 4.86a**	**240.30± 4.92ab**	**235.7± 4.79ab**	**233.± 4.81b**
% of changes vs normal cont. % of changes vsirrad.group		-0.55	-2.37	-1.17	+47.27	+11.08 -24.58	+8.95 -26.02	+7.71 -26.7%

All results represent M±SE.

a : significant in comparison with normal control group. **b** : significant in comparison with irradiated group.

Table (2): Heart tissue CPK, CK-MB" and LDH enzyme activities in the control and different treated groups.

Groups / Parameters	Normal control	Positive control			Treatment			
		SIL	Vit.E	SIL+ Vit.E	Irrad.	Irradi.+SIL	Irrad.+ Vit.E	Irrad. +SIL +Vit.E
CPK (U/g.tissue)	**80.21± 4.45**	**79.38± 4.67**	**77.61± 4.34**	**74.53± 4.38**	**185.56± 4.46a**	**120.48± 4.61ab**	**123.03± 4.44ab**	**119.58± 4.51ab**
% of changes vs normal cont. % of changes vsirrad.group		-0.34	-1.75	-0.41	+131.37	+50.22 -35.07	+53.40- -33.70	+49.10 - 35.56
CK-MB (U/g.tissue)	**26.15± 1.15**	**30.35± 1.14**	**27.96± 1.16**	**23.45± 1.13**	**41.03 ± 1.18a**	**32.26± 1.14ab**	**32.06± 1.11ab**	**31.08± 1.15ab**
% of changes vs normal cont. % of changes vsirrad.group		+16.06	+6.92	-10.33	+56.90	+23.37 -21.37	+32.75 -21.13	+18.85 - 24.25
LDH (U/g.tissue)	**253.8± 4.60**	**241.98± 4.70**	**240.73± 4.50**	**246.58± 4.60**	**338.53± 4.55a**	**261.98± 4.40b**	**257.7± 4.60b**	**256.87± 4.70b**
% of changes vs normal cont. % of changes vsirrad.group		-4.66	-5.15	-2.84	+33.30	+3.22 - 22.61	+1.54 -23.88	+1.21 -24.12

All results represent M±SE.

a : significant in comparison with normal control group. **b** : significant in comparison with irradiated group.

Table (3): Heart tissue CAT activity, GSH, MDA and H_2O_2 contents in the control and different treated groups.

Groups / Parameters	Normal control	Positive control			Treatment			
		SIL	Vit.E	SIL+ Vit.E	Irrad.	Irradi.+ SIL	Irrad.+ Vit.E	Irrad.+ SIL + Vit.E
CAT) (U/ g. tissue)	**58.5± 1.80**	**58.6± 1.79**	**59.06± 1.78**	**62.08± 1.83**	**40.7± 1.82a**	**55.71 1.81b**	**56.73± 1.78b**	**56.85± 1.82b**
% of changes vs normal cont. % of changes vs irrad.group		+0.17	+0.96	+6.12	-30.43	-4.77 +36.88	-3.03 +39.30	-2.82 +39.68
GSH (mmol/ g. tissue)	**15.12± 0.50**	**16.20± 0.53**	**16.49± 0.49**	**17.01± 0.48**	**10.7± 0.52a**	**13.23± 0.49b**	**13.88± 0.49b.**	**14.06± 0.51b**
% of changes vs normal cont. % of changes vs irrad.group		+7.14	+9.06	+12.50	-29.23	-12.50 +23.65	-8.20 +29.7	-7.01 +31.40
MDA (nmol/ g. tissue)	**16.1± 0.80**	**15.98± 0.76**	**15.97± 0.69**	**15.94± 0.77**	**28.05± 0.71a**	**19.83± 0.68b**	**18.43± 0.65b**	**17.80± 0.76b**
% of changes vs normal cont. % of changes vs irrad.group		-24.10	-6.21	-0.62	+74.20	+23.17 -29.30	+14.40 -34.30	+11.18 -36.57
H2O2 (U/ g. tissue)	**0.75± 0.09**	**0.68± 0.07**	**0.63± 0.08**	**0.62± 0.08**	**2.12± 0.078a**	**1.20± 0.081ab**	**1.30± 0.071ab**	**0.91± 0.06b**
% of changes vs normal cont. % of changes vs irrad.group		-9.33	-16.00	-17.33	+182.66	+ 60.00 -43.40	+73.33 -38.63	+21.33 -57.08

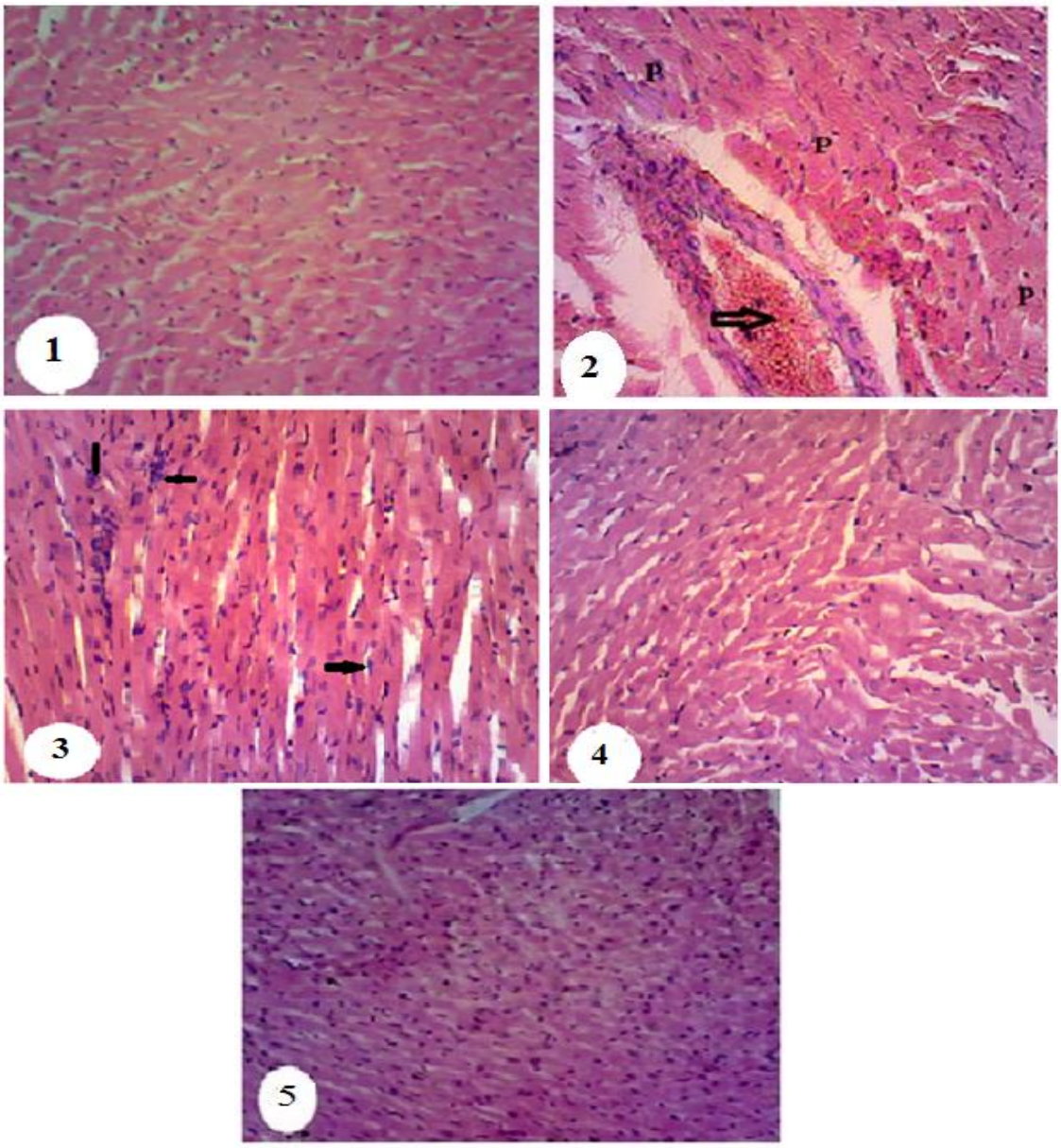
All results represent M±SE.
a : significant in comparison with normal control group.
b : significant in comparison with irradiated group.

Table (4): Serum Sodium (Na), Potassium (K) and Calcium(Ca) levels in the control and different treated groups**.**

Groups / Parameters	Normal control	Positive control			Treatment			
		SIL	Vit.E	SIL+ Vit.E	Irrad.	Irradi.+ SIL	Irrad.+ Vit.E	Irrad.+ SIL+ Vit.E
Na(mg/dl)	**140.26± 0.24**	**139.83± 0.33**	**139.5± 0.25**	**139.6± 0.26**	**135.38± 0.25a**	**139.1± 0.26ab**	**138.9± 0.24ab**	**140.2± 0.23a**b
% of changes vs normal cont. **% of changes vs irrad.group**		-0.31	-0.54	-0.47	-3.48	-.906 +2.77	-0.97 +2.60	-0.04 +3.56
K(mg/dl)	**4.4± 0.32**	**4.36 0.27**	**4.20± 0.30**	**3.80 0.2**	**4.9± 0.32**	**4.35± 0.18**	**4.06± 0.16**	**4.38± 0.2**
% of changes vs normal cont. **% of changes vs irrad.group**		-0.91	-4.55	-13.64	+11.36	-1.14 -11.22	-7.730 -17.14	-0.45 -10.61
Ca(mg/dl)	**9.88± 0.11**	**9.79± 0.14**	**9.77± 0.13**	**9.99± 0.12**	**7.9± 0.15ab**	**8.88± 0.13ab**	**9± 0.14ab**	**9.25± 0.15ab**
% of changes vs normal cont. **% of changes vs irrad.group**		**-0.91**	**-1.12**	**+ 1.11**	-20.04	-10.12 +12.41	-8.91 +13.92	-6.38 +17.09

All results represent M±SE.
a : significant in comparison with normal control group.
b : significant in comparison with irradiated group.

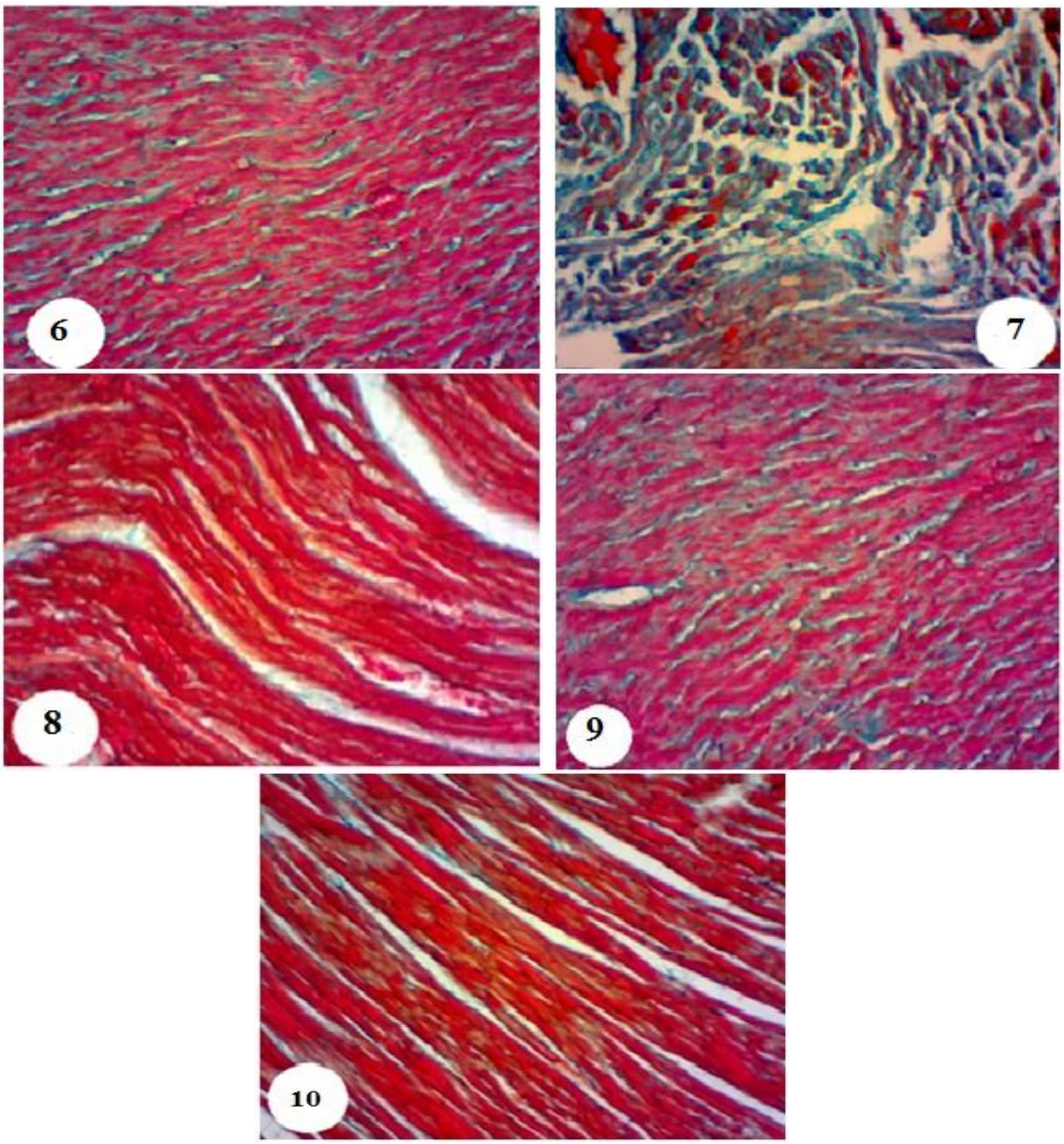
1-The histopatological study



1-The histopatological study

Figs.(1-5) Photomicrographs showing heart tissue of the control and treated groups (H&Ex200). **1**-Showingwelldeveloped cardiac muscle fibres of the control group. **2** -Showingdistorted cardiac muscle fibres of a rat of the irradiated group (R) with deeply stained nuclei (pyknotic) (P) and highly thickened and elongated arterial wall which contains haemolysed blood cells (→). **3**-Showing somewhat normal appearance of the cardiac tissue of group RS, but nuclei of some myocytes are hypertrophied (→). **4**- Showing irregular distribution of nuclei of myocytes of cardiac tissue of group RE (→). 5-Showing somewhat normal appearance of cardiac muscle fibres of a rat of group RSE.

Collagen fibres

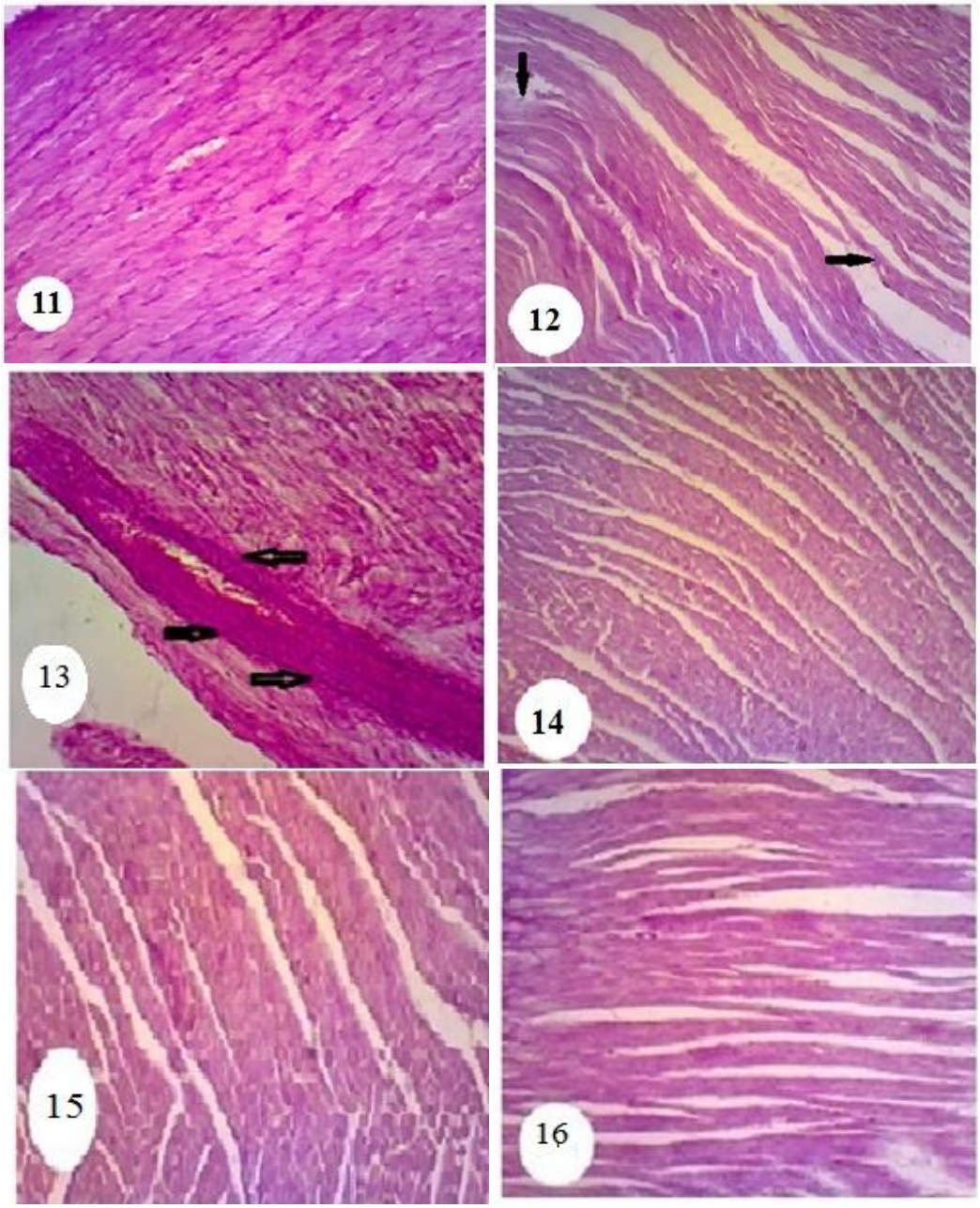


Collagen fibres

Figs.(6-10) Photomicorgraphs showing distribution of collagen fibres in heart of the control and treated groups(Mallory's trichrome, x200).6-Heart of the control group showing normal distribution of collagen fibres in the cardiac muscle fibres. 7- **R** group showing highly increased collagen bundles in the highly distorted cardiac muscle fibres. 8-10.group **RS, RE and RSE** showing somewhat normal appearance of collagen fibres in the cardiac muscle fibres.

2-<u>The histochemical changes</u>

2a-Polysaccharides



<u>Histochemical changes</u>

<u>Polysaccharides</u>

Figs.(11-16) Photomicrgraphs showing distribution of PAS +ve materials in liver tissue of rats of the control and treated groups **(PAS x200). (11)-**Showing normal distribution of PAS +ve materials in liver tissue of rats of the control group. (**12)-**Showing depleted PAS +ve materials in the ruptured cardiac muscle fibres (⟞) of a rat of R group. Notice that highly widened endomysium appeared negatively stained. (**13)-**Showing moderately stained PAS +ve materials in the cardiac muscle fibres of R group, but elongated and thickened arterial wall acquired deep staining affinity (➔of the heart tissue of group **R.**

(14-16)showing somewhat normal content of PAS+ve materials in the cardiac muscle fibres of groups RS,RE and RSE.