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Effect of estrogen depletion induced by vinylcyclohexene diepoxide on the ecg in the female rats

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***Abstract*— Vinylcyclohexene Diepoxide (VCD) kills small pre-antral follicles, especially primordial and primary follicles, The bioactivation of VCD in the ovary results in accelerated atresia of these early-stage follicles. This focused follicular destruction disrupts the ovarian reserve, which leads to ovarian failure. The aim of the study includes effect of VCD on the level of estrogen, And evaluating of alterations in ECG parameters associated with estrogen depletion . rats randomly divided into two groups,the first set as control group ,in the second group the rats where received vinylcyclohexene (VCD) 80 mg/kg BW/daily for 2 weeks as a VCD group. In the current study there was a significant (p≤0.05) decrease in the level of estrogen in the serum (17.40±0.17) in the VCD group compared to the control group (10.22±0.41) . the results of this study showed a significant decreased (p ≤ 0.05) in waves of aterial depolarization (P) (1.49 ± 0.09), ventricular depolarization (QRS)(6.22 ± 0.38),ventricular repolarization (T)(1.474 ± 0.18) time in the VCD group as compare with control group (p)(4.65 ± 0.35) ,QRS (16.56 ± 1.2),T (2.544b ± 0.1) increase in voltage of waves 0.052 ± 0.002 MV (P) 0.04 ± 0.004 (QRS) in the VCD group compare to control group 0.02 ± 0.001 (p), 0.024 ± 0.002 (QRS) while there was no significant difference in T wave between control (0.06 ±0.001) and VCD group (0.07 ± 0.004) . there is a decrease in the level of estrogen in the VCD group that was injected with the VCD, and this depletion causes an increase in the heart voltage.**

***Keywords* — ECG, female rats, estrogen depletion ,VCD**

INTRODUCTION

Estrogen is an essential hormone that is crucial for preserving physiological equilibrium, especially in the cardiovascular and reproductive systems. (1) oestrogen levels drop precipitously with ageing or the start of menopause, resulting in pathological alterations such osteoporosis and cardiovascular diseases.

By diminishing ovarian follicles in female rats, 4-vinylcyclohexene diepoxide (VCD) is employed in experimental studies to imitate this condition. Which cause a decrease in oestrogen production that is similar to the menopause experienced by humans. Studying the physiological and molecular processes linked to oestrogen shortage is made easier with the help of this model. (2)

Heart dysfunction, including changes in electrocardiogram (ECG) parameters that raise the risk of cardiac rhythm problems, has been associated with oestrogen shortage. The loss of estrogen's protective effects on cardiac ion channels and vascular function is partially responsible for these alterations. However, it is still unclear exactly how oestrogen receptors (ERα and ERβ) affect these cardiac outcomes, especially in models of chemically induced hormone depletion. (3)

The electrical activity produced by the heart is simply recorded in an electrocardiogram (ECG). An example of ECG signals from a typical cardiac cycle is provided. For a number of biological uses, including heart rate measurement, cardiac rhythm analysis, abnormality diagnosis, emotion detection, and biometric identification, the ECG is a useful non-invasive instrument. The detection of cardiovascular illnesses is one of the main areas where ECG analysis is necessary (4).

An ECG's waves make it possible to distinguish between a normal and abnormal cardiac beat. The P wave, which indicates the depolarisation of the atrial muscles, is the first deflection visible on an ECG. Ventricular depolarisation is represented by the QRS complex, while ventricular repolarisation is represented by the subsequent T wave (5). The study's objectives include determining how VCD affects oestrogen levels, Assessing changes in ECG parameters linked to oestrogen deficiency using VCD

MATERIALS AND METHODS

The animals were placed in good condition in special plastic cages.In terms of temperature around (30 ±5 C°) , ventilation and the light system was 12 hrs per day (**6**)

A total of 16 adult female rats, they were placed in the animal house of veterinary medicine college of kerbala university

,rats were randomly and equally divided into two groups , each group contain 8 rats received normal saline intramuscular (IM) as control group. In the second group animals received 80 mg\ kg BW daily of VCD (Macklin-china) (IM) for 2 weeks according to method reported by (**7**).

Electrocardiogram (ECG) recordings were performed on animals without anesthesia. The animals were prepared by shaving their fur and placing them on a custom-designed wooden board. Modified electrodes were attached to their limbs after applying a conductive gel to facilitate electrical impulse transmission. The animals were calmed by xylazine (Dutch farm international-holland) repeatedly before electrode placement to prevent fear or panic. Recordings followed Einthoven's Triangle method, using settings of 10 mm/mV amplification, 25 mm/s voltage, and a speed of ½ second. Results were plotted on specialized graph paper divided into 1 mm squares (vertical: 1 mm = 0.1 mV; horizontal: 1 mm = 0.04 seconds). The P wave represented atrial depolarization, the QRS complex indicated ventricular depolarization, and the T wave reflected ventricular repolarization. Atrial repolarization was not recorded due to weaker atrial muscle signals. (8,9).

Ethical approve

This investigation was conducted in the physiology department ,university of karbala ,collage of veterinary medicine under reference number UOK.VET.PH.2024.098

STATISTICAL ANALYSIS

Statistical analysis was done using graphpad prism software 8.0,using the T-test method to measure the differences between the two different groups **(10).**

RESULT & DISCUSSION

In the current study there was a significant ($p \leq 0.05$) decrease in the level of estrogen in the serum in the VCD group compared to the control group as a result of ovarian failure**.**

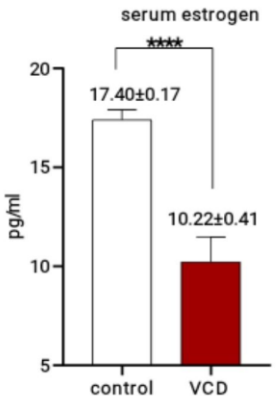

Figure 1. The effect of VCD injected on level of serum estrogen to induced ovarian failure in rats

In the current study there was a significant ($p \leq 0.05$) decrease in waves of (P, QRS and T) time in the VCD group as compare with the control group .also In the current study there was a significant ($p \leq 0.05$) increase in voltage of waves (P and QRS) in the VCD group as compare with the control group while there was no significant difference in T wave between control group and VCD group as shown in table (1)

Table 1. effect of ovarian failure induced by 80 mg daily /2weeks Im injected g/of VCD in the P wave, QRS wave and T-wave time and voltage in rats

Parameter	Control group	VCD group
P wave (ms)	4.65 ± 0.35 A	1.49 ± 0.09 B
QRS wave (ms)	16.56 ± 1.2 A	6.22 ± 0.38 B
T wave (ms)	2.544b ± 0.1 A	1.474 ± 0.18 B
P wave (MV)	0.02 ± 0.001 B	0.052 ± 0.002 A
QRS wave (MV)	0.024 ±0.002 B	0.04 ± 0.004 A
T wave (MV)	0.06 ±0.001 A	0.07 ± 0.004 A

Capital letter in the same row represents a significant deference

VCD destroyed small pre-antral follicles, especially primordial and primary follicles, but has no effect on larger, more developed follicles, according to research. The bioactivation of VCD in the ovary results in accelerated atresia of these early-stage follicles. This focused follicular destruction disrupts the ovarian reserve, which leads to ovarian failure. Research employing mouse models has elucidated this process and provided insight into the temporal progression of follicular depletion subsequent to VCD exposure **(2).**

Primordial and primary follicles are the ovary's principal sites for oestrogen production, hence their depletion directly affects the synthesis of oestrogen. The quantity of oestrogen in the blood drastically decreases when these follicles are gone. **(11).**

By altering the sodium (Na), potassium (K), and calcium currents, oestrogen plays a crucial role in controlling cardiac ion channels and affecting depolarisation and repolarisation. VCD-treated patients' shorter P, QRS, and T wave lengths might be a result of ion channel remodelling brought on by oestrogen loss**. (12)**

Additionally, a lack of oestrogen may decrease the length of P waves and accelerate atrial depolarisation by decreasing late sodium currents, which prolong atrial action potentials. **(13)**

Although it is uncommon in conduction problems, faster ventricular conduction (narrower QRS) may result from myocardial fibrosis or increased sodium channel efficiency, both of which are impacted by estrogen's anti-fibrotic qualities **(14).**

Furthermore, During repolarisation, potassium currents are suppressed by oestrogen. According to **(15),** hypoestrogenism

may reduce T wave time and shorten action potential duration (APD) via increasing outward potassium flow.

Moreover, oestrogen regulates the balance between the sympathetic and parasympathetic nervous systems, and its deficiency may cause the autonomic tone to change in favour of the sympathetic. Reduced waveform lengths may be explained by increased sympathetic activity, which speeds up depolarisation and repolarisation **(16).**

On the other hand, ovarian tissue experiences oxidative damage because to VCD. Reactive oxygen species (ROS) may change conduction dynamics by damaging ion channels or mitochondrial function if comparable effects are seen in the heart **(17).**

In contrast to previous studies of low-voltage ECGs in hypoestrogenic or conduction deficiency animals, the 4-vinylcyclohexene diepoxide (VCD)-exposed group showed a considerable increase in P and QRS wave voltages when compared to controls. This surprising discovery casts doubt on accepted theories on the effects of oestrogen depletion on the heart and points to a special electrophysiological response to endocrine disturbance **(18).**

Oetrogen deprivation is associated ventricular hypertrophy .Because muscle development increases the amount of depolarising tissue, electrical impulses may be amplified **(19).**

Additionally, lowering oestrogen may increase sympathetic tone, which would increase myocardial force production and calcium influx. Similar to the "hyperdynamic phase" in early heart failure, this hypercontractile condition may indirectly increase depolarisation voltages **(20).**

Additionally, oestrogen has anti-adrenergic and parasympathetic actions. speeds up depolarisation by activating β-adrenergic receptors, which raises sodium and calcium currents and may raise myocardial voltage**. (21)**

Additionally, it increases myocardial contractility, which may amplify electrical impulses. This is consistent with findings of hyperadrenergic conditions exhibiting catecholamine-induced increases in QRS amplitude **(22).**

CONCLUSION

There is a decrease in the level of estrogen in the group that was injected with the VCD, and this depletion causes an increase in the heart voltage this increase due to decrease in QRS interval as well as the, which caused a decrease in the time of the beat when compared to the control group.

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