University of Thi-Qar Journal of agricultural research

ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24

https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

LD50 and affective dose of Eruca sativa mill (gergeer) ethanolic extract

[1]Zuhair Abdulkareem. Dawah ,[2]Rana Abdulla Salih

Department of physiology, biochemistry and pharmacology, college of veterinary medicine, university of Baghdad

[1]*Email: Zouher.kareem1206h@covm.uobaghdad.edu.iq*

[2]*Email: Rana.a@covm.uobghdad.edu.iq*

Key word: LD50 , affective dose, Rabbit.

Abstract

Cardiotoxicity is defined as drug-induced damage to the heart muscle, which can result in cardiac failure or cardiac arrest. Cardiotoxicity is a common side effect of traditional anticancer drugs such as doxorubicin, taxanes, 5-fluorouracil, and cyclophosphamide, as well as newer agents such as biological antibodies such as Nivo. Cardiotoxicity is also caused by the long-term use of neurologic/psychiatric medications. This study was designed to determine the effect of *eruca sativa* mill (Gergeer) leaves extract on cardiotoxicity induced by doxorubicin in rabbits. The fifty-four rabbits used in this study were obtained from a local rabbit breeder. Animals ranged between 1-1.3 years old, and their weight was approximately 1-2kg without specifying gender experiment Acute toxicity study, The pilot study to determine the LD50 dose of *Eruca sativa* mill (gergeer) by "up and down" method after acute exposure in rabbits, The effective dose of *Eruca sativa* mill (gergeer) was determined by using a simple line equation. The effective dose of Eruca sativa. troponin I determine the level of proteins in the blood increase in heart attack,.The Troponin-I level of the control positive group revealed a significant increase as compared with G3, G4, G5, and the control negative group.The cTn-I level of the G3 significant increase is compared with treated G4,G5 group and control negative group with a significant decrease is compared with G1, G2 group and control positive group ; while treated G4 ,G5 group significant decrease is compared with treated G1,G2,G3 group and control positive, Log number dose-response curve and maximum effective dose the simple line equation revealed that the effective Eruca sativa dose of Troponin I level was (3.01 , 2.99 ,1.97 ,1.015, 1.030) (ng/ml) respectively, whereas the effective *Eruca sativa* dose of Troponin I was 316 mg/kg. conclusion , The effective dose-response results asymptotic to 10 % of LD50, induced cardiac toxicity with DOX appear biochemically, cardiographycally and histopathologically, The Eruca sativa mill (gergeer) extract was effective protective for induced cardiac toxicity.

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main https://doi.org/10.54174/UTJagr.Vo11.N2/02

I. INTRODUCTION

Cardio toxicity refers to the occurrence of myocardial injury and/or cardiac electrophysiological malfunction. Increased production of free radicals causes oxidative stress, which has been implicated in the pathophysiology of cardiovascular disorders such as ischemic heart disease, myocardial infarction, arrhythmias, atherosclerosis, and congestive heart failure (CHF) **(Taleb *et al.*, 2018).**

Cardio toxicity is a side effect of doxorubicin (DOX) that causes myocardial or valve damage. It can happen during or soon after treatment, days or weeks later, It may not become obvious for months, if not years, after cancer therapy has ended. Fatigue, anemia, and shortness of breath were signs of this consequence, indicating that the heart was having trouble performing its important activities **(Kancharlapalli *et al.*, 2014).**

Doxorubicin (DOX) is an antineoplastic drug that belongs to the anthracycline family. It has been the mainstay of cancer chemotherapy for decades, with well-established, highly effective, and notable successes in the treatment of both solid and hematological malignancies in both adult and pediatric patients **(Shaker *et al.*, 2018).**

Doxorubicin's mechanism of action Doxorubicin's principal method of action was the drug's ability to intercalate between DNA base pairs, inducing DNA strand breaking and inhibiting DNA and RNA synthesis. Doxorubicin inhibits topoisomerase II, resulting in DNA damage and death in cells. Doxorubicin produced free radical-mediated oxidative damage to DNA when coupled with iron, significantly reducing DNA synthesis **(Johnson-Arbor and Dubey, 2017)**.

Mechanisms of doxorubicin cardiotoxicity Oxidative stress causes the mitochondria to produce harmful reactive oxygen species (ROS), which are harmful to cells. Increased intracellular concentration of DOX due to alterations in MDR efflux proteins, decreased uptake of Ca2+ into the sarcoplasmic reticulum (SR) causes myocardial contractility disturbances, and decreased mesenchymal and circulating progenitor cells reduces the heart's regenerative capacity following damage **(Rockley, 2018)**.

Eruca sativa Mill, often known as arugula, gergeer, or rocket leaves, is a prominent plant of the Brassicaceae family. It's also commonly used in salad dressings. The presence of various key elements, such as dietary fiber, oligosaccharides, aminoacids, peptides, proteins, polyunsaturated fatty acids, vitamins, carbohydrates, L-ascorbic acid, and mineral content, makes *E. sativa* leaves a good source of nutrition. Additionally, dieticians have noted that *E. sativa* is recognized for its low calorific content as well as strong nutritional benefits. *E. sativa* has risen to prominence in recent years as a result of its high phytochemical content and importance in a variety of biological activities. Flavonoids, glucosinolates, phenolics, saponins, tannins, and essential oils are all found in different regions of the E. sativa plant. Isothiocyanates, derivatives of butane, octane, nonane, 4-methylthiobutyl,isothiocyanate,cis-3-hexen-1-ol,5methylthiopentylisothiocy -anate,cis-3-hexenyl 2-methylbutanoate, 5-methylthiopentane nitrile, quercetin, kampferol, rutin, The essential oil composition of the *E. sativa* seed was studied, and it was shown to have a considerable amount of sulfur and nitrogen compounds. Biological effects, such as antibacterial and

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main https://doi.org/10.54174/UTJagr.Vo11.N2/02

antioxidant, are mainly attributed to active molecules created by essential oils and other phytoconstituents during secondary vegetal metabolism. Antimicrobial, antigenotoxic, antidiuretic, stimulant, stomach disorders analgesic, antioxidant, antiulcer, hepatoprotective activities, antidiabetic, antiacne, antihyperlipidemic, antihyperglycemic, and anti-inflammatory properties have been reported for a variety of phytochemicals found in *E. sativa*. In Central Asia, *E. sativa* seed oil, also known as taramira oil or jamba oil, is used for massages, hair treatments, and as an anti-influenza drug **(Awadelkareem *et al*., 2022).** investigation found that *E. sativa* had no discernible impact on VLDL levels **(Weli *et al*., 2021)**.

II. MATERIALS AND METHODS

Plant Materials:

The *Eruca sativa* Mill leaves were collected in December 2021 from the Baghdad region (Dora). The classification of the plant was done in the Ministry of Agriculture/ State Board for Seed Testing and Certification (S.B.S.T.C) in Abu Graib /Baghdad at the certification number 87 in 16 /12/2021. The shape of Eruca sativa Mill (Gergeer) as in figure (1-1).

Figure (1-1): appearance of Eruca sativa mill (Gergeer) leaves

Extraction:

The fresh leaves of *Eruca sativa* were dried by air in the shade, ground into a fine powder using electrical grinder and weighing 200 gm then put it in a volumetric conical flask, 2000 ml of 70 % ethyl alcohol was added to the powder which makes the ratio (1/10) (W/V). After that the solution was mixed by using magnetic stirrer apparatus for 24hr, the mixture was filtered by using 4 layers of medical gauze then was filtered again using Whatman (No.1) filter paper. The filtered mixture was

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main https://doi.org/10.54174/UTJagr.Vo11.N2/02

concentrated by using incubator on (40°C) for 72hr, to obtain crude extract. This yield equal of 24 gm, which equal 12 %, the extract was stored in a dark sterile screw bottle at (4°C) until used **(Jin *et al*., 2009).**

Experimental design:

The pilot study:

The experiment began with a series of oral doses of *Eruca sativa* mill at various doses. In this investigation, twelve adult's rabbits' local breed were used. The doses of Eruca sativa mill were initiated at (1000mg/kg) and were orally given to two rabbits with stomach tubes (5ml). The doses (2000mg/kg), (3000 mg/kg), (4000mg/kg), (5000 mg/kg), (6000 mg/kg), were estimated based on the weight of the rabbits. The dose that killed the two animal was (4000 mg/kg).

Median lethal dose (LD50):

LD50s of extract was conducted to estimate the toxic potency using up and down method. The ranges of toxic doses were estimated by primary study (Pilot) for extract. Seven rabbits that were given increasing doses of the extract orally (by stomach tube), the dead animals was recorded during 24 hrs.

Up and Down method:

The improved estimates are available directly in table (1-1). The (O) for the survive and (X) for dead, according to the rule to calculate the LD50 **(Dixon, 1965)**.

LD50 = (Final test level) + (Value from table) (Difference between dose levels).

LD50 = Xf +Kd

Xf = final test level.

K = value from table.

d = difference within dose levels.

University of Thi-Qar Journal of agricultural research

ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24

https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

N	Second Part of Series	k for Test Series Whose First Part is					Standard Error of LD_{50}
		O	*OO*	*OOO*	*OOOO*		
2	*X*	−.500	−.388	−.378	−.377	*O*	.88σ
3	*XO* *XX*	.842 −.178	.890 .000	.894 .026	.894 .028	*OX* *OO*	.76σ
4	*XOO* *XOX* *XXO* *XXX*	.299 −.500 1.000 .194	.314 −.439 1.122 .449	.315 −.432 1.139 .500	.315 −.432 1.140 .506	*OXX* *OXO* *OOX* *OOO*	.67σ
5	*XOOO* *XOOX* *XOXO* *XOXX* *XXOO* *XXOX* *XXXO* *XXXX*	−.157 −.878 .701 .084 .305 −.305 1.288 .555	−.154 −.861 .737 .169 .372 −.169 1.500 .897	−.154 −.860 .741 .181 .380 −.144 1.544 .985	−.154 −.860 .741 .182 .381 −.142 1.549 1.000^{+1}	*OXXX* *OXXO* *OXOX* *OXOO* *OOXX* *OOXO* *OOOX* *OOOO*	.61σ
6	*XOOOO* *XOOOX* *XOOXO* *XOOXX* *XOXOO* *XOXOX* *XOXXO* *XOXXX* *XXOOO* *XXOOX* *XXOXO* *XXOXX* *XXXOO* *XXXOX* *XXXXO* *XXXXX*	−.547 −1.250 .372 −.169 .022 −.500 1.169 .611 −.296 −.831 .831 .296 .500 −.043 1.603 .893	−.547 −1.247 .380 −.144 .039 −.458 1.237 .732 −.266 −.763 .935 .463 .648 .187 1.917 1.329	−.547 −1.246 .381 −.142 .040 −.453 1.247 .756 −.263 −.753 .952 .500 .678 .244 2.000 1.465	−.547 −1.246 .381 −.142 .040 −.453 1.248 .758 −.263 −.752 .954 .504^{+1} .681 .252^{+1} 2.014^{+1} 1.496^{+1}	*OXXXX* *OXXXO* *OXXOX* *OXXOO* *OXOXX* *OXOXO* *OXOOX* *OXOOO* *OOXXX* *OOXXO* *OOXOX* *OOXOO* *OOOXX* *OOOXO* *OOOOX* *OOOOO*	.56σ
		X	*XX*	*XXX*	*XXXX*	Second Part of Series	
		−k for Series Whose First Part is					

Table (1-1): Values of K for estimating LD50 from Up and Down .If the table is entered from the foot, the sign of K is to be reversed (Dixon, 1965)

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main https://doi.org/10.54174/UTJagr.Vo11.N2/02

Determination effective dose of Eruca sativa mill ethanolic extract:

Thirty-five rabbit were divided into three groups, the first group included five adult rabbit were given D.W orally as control negative group, the second group included five adult rabbit were induced cardio toxicity by DOX at dose (4 mg/kg. B.W) intraperitoneally I.P between day to day for 14 days **(Al-okaily *et al*., 2013)** as control positive group, the third group included twenty-five rabbits divided into five equal subgroups and treated with Eruca sativa mill ethanolic extract as protective The first subgroup (G1), second subgroup (G2), third subgroup (G3), fourth subgroup (G4), and fifth subgroup (G5) were given doses(25 ,50 ,100 ,200, 400) mg/kg respectively, orally daily by gavage needle for 14 days then rabbit induced cardio toxicity by DOX at dose (4 mg/kg) I.P between day to day for 14 days **(Al-okaily *et al*., 2013)**, with continuous give *eruca sativa* orally for 14 days.

Blood Serum Sample:

Blood samples were collected gently from heart by sterile syringe provided with needle gage 23, and all blood samples were transferred in to clean sterile test tubes without anticoagulant and left for 20 minutes. Serum was obtained by centrifugation blood samples at 3000 rpm for 15 minutes and finally preserved in Eppendorf tube under -2 C◦ till used.

Troponin I parameter procedure :

1- Using a transfer pipette, transfer 75µl. (serum) to a blank sample mixing tube and add 75µl detection buffer.

2- Close the lid of the sample mixing tube and shake it 10 times to fully mix the sample. The sample mixture must be used as soon as possible.

3-Pipette out 75µl of a sample mixture and place it in the cartridge's sample well.

4- Let 12 minutes for the sample-loaded cartridge at room temperature.

5- Insert the sample-loaded cartridge into the ichromaTM testing instrument's cartridge holder to scan it.

6- Before placing the cartridge all the way into the cartridge holder, be sure it is properly oriented. On the cartridge, a specific indicator has been marked for this reason.

7- To begin the scanning process, press the ichromaTM testing 'Select' button on the device.

8- The device for ichromaTM testing will directly begin scanning the sample-loaded cartridge.

9- For ichromaTM tests, read the test result on the device's digital screen.

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

III. RESULTS AND DISCUSSION

Eruca sativa Mill extract:

The extract of *Eruca sativa* was extracted by using 70% ethanol and the percentage of extract yield was 12% ; this was calculated by the following equation:

yield of the extract (%) = weight of extract (gm) / weight of *Eruca sativa* (gm) × 100 (Banso and Adeyemo, 2006).

yield of the extract (%) = 12(gm) /100 (gm) x 100 = 12%

Figure (1-2): Eruca sativa mill extract

This result is similar to the result of **(Nowfel, 2015)** who found that the percentage recovery of ethanolic extract was 10% from 100 gm of fine Eruca sativa powder.

Pilot study:

The pilot study of *Eruca Sativa* Mill in rabbits as in (Table 4-1)

Table (4-1): Pilot study of *Eruca sativa* mill in rabbit during 24 hrs.

referred to No. of rabbits that died or lived.

Different doses were used to perform this experiment, and the following results were shown, due to the lack of sources that stated the dose accurately and in detail in such experiments.

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

Dose mg/kg	No. of rabbit	No. of rabbit lived	No. of rabbit died
1000	2	2	0
2000	2	2	0
3000	2	2	0
4000	2	1	1
5000	2	0	2
6000	2	0	2

Median lethal dose (LD50) :

The calculated value of LD50 of Eruca sativa in the rabbit was estimated by "up and down" method, the result of rabbit were shown in table (1-3) for each group.

Table (1-3): LD50 of Eruca sativa in the rabbit in acute toxic dose for 7 rabbits.

Dose Mg/kg	Conc. Mg/ml	Dead and survived Rabbits
4500	900	X
4000	800	X
3500	700	X
3000	600	O
3500	700	X

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

3000	**500**	**O**
3500	**700**	**X**

(XXXOXOX)

O for survived rabbits and X for dead rabbits.

LD50= Xf +Kd

= 3500+(-0.741) x 500

= 3500 -370.5

= 3129.5 mg/kg

This result similar to **(Hammami *et al.*, 2019)** who found the LD50 of Eruca sativa was 3.2 gm for the extract.

Determination Effective Dose of Eruca sativa Mill ethanolic extract:

After the end of period of *Eruca sativa* mill ethanolic extract (gergeer) treatment (4 weeks). The rabbit Troponin I (cTn-I) (ng/ml) in serum was measured. The oral administration of Eruca sativa at dose 25,50,100,200 and 400 mg/kg B.W to rabbit showed a gradual decrease in cTn-I level in serum when compared with the control until reached plateau at dose 200 and 400 mg/kg .B.W as in figure(1-3).

The Troponin-I level of the control positive group revealed a significant increase ($P<0.05$) as compare with G3,G4,G5 and control negative group (Dox) with no significant variance ($P<0.05$) in comparison with G1 at dose (25 mg/kg B.W) and G2 at dose (50 mg/kg). The cTn-I level of the G3 at dose (100 mg/kg B.W) exhibited a significant elevation ($P>0.05$) is compared with treated G4,G5 group at a dose (200,400 mg/kg B.W) and control negative group with a significant reduction ($P<0.05$) is compared with G1, G2 group and control positive group ; while treated G4 ,G5 group at dose(200 ,400 mg/kg B.W) exhibited a significant decline ($P>0.05$) is compared with treated G1,G2,G3 group and control positive at a dose (25,50,100 mg/kg B.W) and there was no statistical different ($P>0.05$) when compared with control negative group. As in table (1-4).

Table (1-4) : The effect of Eruca sativa mill (gergeer) ethanolic extract (25,50,100,200,400) mg/kg B.W on Troponin I (ng/ml) in reduced cardio toxicity rabbit by doxorubicin.

University of Thi-Qar Journal of agricultural research

ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24

https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

Groups	Dose (mg/kg) B.W.	Response Troponin I (ng/ml)
Control	D.W	1.01 ±0.04 C
DOX	4	3.23 ±0.07 A
G1	25	3.01±0.06 A
G2	50	2.99 ±0.06 A
G3	100	1.97 ±0.04 B
G4	200	1.015 ±0.03 C
G5	400	1.030 ±0.03 C
LSD value		0.671 *
Means having with the different letters in same column differed significantly. * (P≤0.05).		

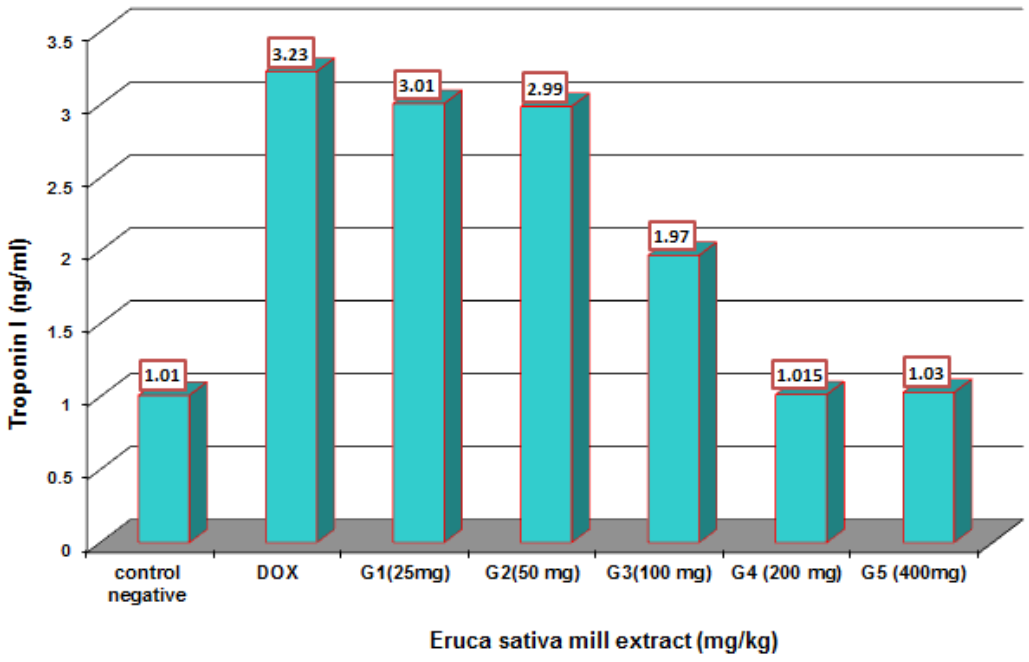

Figure (1-3) : Effect of Eruca sativa mill extract at doses (25,50,100,200,400) mg/kg of B.W on Troponin I(ng/ml) in serum of reduced cardio toxicity by doxorubicin rabbits.

The elevation in the activity of Cardiac Troponin-I (cTn-I) in (the DOX group) indicated injury or damage to cardiac cells. Serum concentrations of cTn-I are important myocardial indicators for the evaluation of cardiotoxicity and congestive heart

University of Thi-Qar Journal of agricultural research
ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24
https://jam.utq.edu.iq/index.php/main https://doi.org/10.54174/UTJagr.Vo11.N2/02

failure, this result is agreed with **(Martín-Peláez *et al*., 2017; AlMalki and Shahid, 2020).** As well, the increased levels of these enzymes may reflect the toxic effect of doxorubicin that causes suppression of protein and nucleic acid synthesis **(Abbas and Kabil, 2017)**. Additionally, doxorubicin has been led to anemic hypoxia which caused by modification of erythropoiesis leading to a deficiency of oxygen supply or glucose may damage the myocytes and the cellular membrane become more permeable and ruptured, resulting in leakage of these enzymes to blood circulation **(Wallace *et al*., 2004; Thippeswamy *et al*., 2011)**. doxorubicin also led to cardiac remodeling by excessive generation of oxidative free radicals and lipid peroxides that might harm cell membranes and lead to the release of cytosolic enzymes **(Oliveira *et al*., 2004)**. Also, The peroxidation of membrane-bound polyunsaturated fatty acids and protein oxidation were brought on by the impairment of oxidant-antioxidant systems., which led to harm to cardiac tissues due to a change in the permeability of myocytes. **(Fadillioglu *et al*., 2004; Abbas and Kabil, 2017)**. Whereas in the (*Eruca sativa* group) the concentrations of cTn-I were reduced when matched to (DOX group), the possible action of *Eruca sativa* mill extract is mediated through scavenging physiologically relevant via preventing the oxidation of LDL, reactive oxygen species **(Hadi *et al*., 2017)**. *Eruca sativa* contained a quantity of antioxidant-rich chemical components activity like various polyphenolic compounds. As well as it contained anthocyanin which could reduce the risk of myocardial infarction (MI) **(Bennett et al., 2006)**. Polyphenols of *Eruca sativa* further to its scavenging capacity. have been shown to have strong in vitro antioxidant activity. They can stop LDL oxidation and lipid peroxidation by acting as chain-breaking peroxyl radical scavengers. **(Heimler *et al*., 2007)**. *Eruca sativa* is a very good source of selenium and vitamin E, it is involved in DNA repair **(Alotaibi *et al*., 2020)**. Antioxidants worked synergistically with vitamin E, keeping the heart healthy and producing antibodies. By halting the production of free radicals, it defended the immune system. It assisted in controlling how thyroid hormones affected how quickly fat is burned. **(Singh *et al*., 2013; Gupta et al., 2019).** the results of control negative agreed with (Salim, 2021) that was recorded.

Log dose-response curve and effective dose :

The log dose-response curve of Troponin I level in groups of rabbit with different doses of Eruca sativa mill ethanolic extract (25, 50, 100, 200,400) mg/kg of B.W. shown in figure (1-4) respectively. The simple line equation revealed that the maximum effective Eruca sativa dose of Troponin I level was (3.01 , 2.99 ,1.97 ,1.015, 1.030) (ng/ml) respectively, whereas the maximum effective Eruca sativa dose of Troponin I was 316 mg/kg, which dependent on the current experiment.

University of Thi-Qar Journal of agricultural research

ISSN Onlin:2708-9347, ISSN Print: 2708-9339 Volume 11, Issue 2 (2022) PP 11-24

https://jam.utq.edu.iq/index.php/main *https://doi.org/10.54174/UTJagr.Vo11.N2/02*

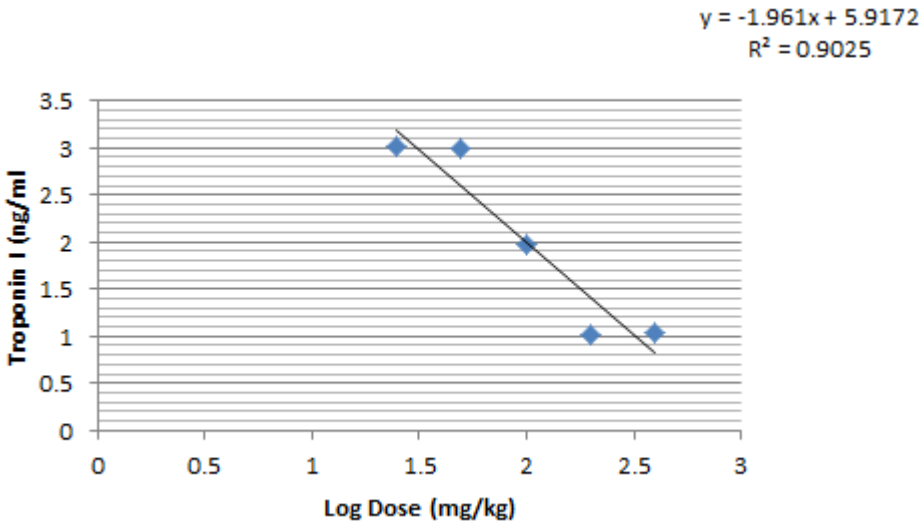

Figure (1-4): Log number dose-response curve of Eruca sativa

Mill ethanolic extract on (cTn-I ng/ml) in serum of rabbit.

IV. REFERENCES

1. Abbas, N. A. T., and Kabil, S. L. (2017). Pentoxifylline and Cilostazol Against Rat Heart Injuries Induced by Doxorubicin. Egyptian Journal of Basic and Clinical Pharmacology, 7(1), 47–56.
2. AlMalki, W. H., and Shahid, I. (2020). Characterization of antihypertensive and cardioprotective effects of extra virgin olive oil against doxorubicin induced cardiomyopathy in rats. Journal of Pharmacy and Pharmacognosy Research, 8(4), 316–326.
3. AL-OKaily,B.N.; AL-Haddad,A.A and Salman,A.D.(2013). Effect of Different Doses of Doxorubicin on pituitary Gland and some Testicular function in adults Male Rabbits. The Iraqi Journal of Veterinary medicine, 37C1):121-128
4. Alotaibi, B., El-Masry, T. A., Tousson, E., Alarfaj, S. J., & Saleh, A. (2020). Therapeutic effect of rocket seeds (Eruca sativa L.) against hydroxyapatite nanoparticles injection induced cardiac toxicity in rats. Pak. J. Pharm. Sci, 33(4), 1839-1845.
5. Awadelkareem, A. M., Al-Shammari, E., Elkhalifa, A. E. O., Adnan, M., Siddiqui, A. J., Snoussi, M., ... & Ashraf, S. A. (2022). Phytochemical and In Silico ADME/Tox Analysis of Eruca sativa Extract with Antioxidant, Antibacterial and Anticancer Potential against Caco-2 and HCT-116 Colorectal Carcinoma Cell Lines. Molecules, 27(4), 1409.

6. Bennett, R. N., Rosa, E. A., Mellon, F. A., & Kroon, P. A. (2006). Ontogenic profiling of glucosinolates, flavonoids, and other secondary metabolites in Eruca sativa (salad rocket), Diplotaxis erucoides (wall rocket), Diplotaxis tenuifolia (wild rocket), and Bunias orientalis (Turkish rocket). Journal of agricultural and food chemistry, 54(11), 4005-4015.
7. Dixon, W. J. (1965). The "Up and Down" method for small samples. American Statistical Association Journal, P. 967-978.
8. Fadillioglu, E., Oztas, E., Erdogan, H., Yagmurca, M., Sogut, S., Ucar, M., and Irmak, M. K. (2004). Protective effects of caffeic acid phenethyl ester on doxorubicin-induced cardiotoxicity in rats. Journal of Applied Toxicology: An International Journal, 24(1), 47–52.
9. Gupta, M., Kaur, H., G. Singhal, K., and K. Gupta, V. (2019). Evaluation of avena sativa seeds extract as cardioprotective agent in rats. IP International Journal of Comprehensive and Advanced Pharmacology, 4(2), 39–47.
10. Hadi, M. A., Almamoori, A. M., Al-Hassnawi, A. T., & Hameedi, E. H. (2017). Oxidative response associated with treatment of male Albino rats with Eruca sativa Mill leaves extract and correlations with complete blood picture. Journal of Pharmaceutical Sciences and Research, 9(11), 2278-2285.
11. Hammami, I. et al. (2019) 'Protective Action of Eruca sativa Leaves Aqueous Extracts Against Bisphenol A-Caused In Vivo Testicular Damages', 00(0), pp. 1–11.
12. Heimler, D., Isolani, L., Vignolini, P., Tombelli, S., & Romani, A. (2007). Polyphenol content and antioxidative activity in some species of freshly consumed salads. Journal of Agricultural and Food Chemistry, 55(5), 1724-1729.
13. Jin, J.; Koroleva, O.A.; Gibson, T.; Swanston, J. and Magan, J. et al., (2009). Analysis of phytochemical composition and chemoprotective capacity of rocket (Eruca sativa and Diplotaxis tenuifolia) leafy salad following cultivation in different environments. J. Agric. Food Chem., 57(12): 5227-5234.
14. Johnson-Arbor, K., and Dubey, R. (2017). Doxorubicin.
15. Kancharlapalli, S., Sweeya, P. S., Gandham, A. P. D., and Rao, V. U. M. (2014). CARDIOTOXICITY AND USE OF HERBAL DRUGS- A REVIEW. Asian Journal of Pharmaceutical Research and Development, 2(1), 1–9.
16. Martín-Peláez, S., Castañer, O., Konstantinidou, V., Subirana, I., Muñoz-Aguayo, D., Blanchart, G., Gaixas, S., de la Torre, R., Farré, M., and Sáez, G. T. (2017). Effect of olive oil phenolic compounds on the expression of blood pressure-related genes in healthy individuals. European Journal of Nutrition, 56(2), 663–670.
17. Nowfel, A.J. (2015) 'Role of alcoholic extract of Roket (Eruca sativa) leaves on male reproduction of experimentally induced-oxidative stressed rats', 39(2), pp. 47–54.
18. Oliveira, P. J., Bjork, J. A., Santos, M. S., Leino, R. L., Froberg, M. K., Moreno, A. J., & Wallace, K. B. (2004). Carvedilol-mediated antioxidant protection against doxorubicin-induced cardiac mitochondrial toxicity. Toxicology and applied pharmacology, 200(2), 159-168.

19. Rockley, K. (2018). In Vitro Evaluation of Anthracycline-induced Cardiotoxicity and Mitigation by Perturbation of Angiotensin Signalling (Doctoral dissertation, Durham University).
20. Salim, A.G. (2021) 'Effect of Avena Sativa Seeds Ethanolic Extract and Enalapril Alone and Combined on Induced Cardiotoxicity by Doxorubicin in Adult Female Rats'.
21. Shaker, R. A., Abboud, S. H., Assad, H. C., and Hadi, N. (2018). Enoxaparin attenuates doxorubicin induced cardiotoxicity in rats via interfering with oxidative stress, inflammation and apoptosis. BMC Pharmacology and Toxicology, 19(1), 1-10.
22. Singh, R., De, S., and Belkheir, A. (2013b). Avena sativa (Oat), A Potential Neutraceutical and Therapeutic Agent: An Overview. Critical Reviews in Food Science and Nutrition, 53(2), 126-144.
23. Taleb, A., Ahmad, K. A., Ihsan, A. U., Qu, J., Lin, N., Hezam, K., Koju, N., Hui, L., and Qilong, D. (2018). Antioxidant effects and mechanism of silymarin in oxidative stress induced cardiovascular diseases. Biomedicine and Pharmacotherapy, 102, 689–698.
24. Thippeswamy, A. H. M., Shirodkar, A., Koti, B. C., Sadiq, A. J., Praveen, D. M., Swamy, A. H. M. V., and Patil, M. (2011). Protective role of Phyllantus niruri extract in doxorubicin-induced myocardial toxicity in rats. Indian Journal of Pharmacology, 43(1), 31.
25. Wallace, K. B., Hausner, E., Herman, E., Holt, G. D., MacGregor, J. T., Metz, A. L., Murphy, E., Rosenblum, I. Y., Sistare, F. D., and York, M. J. (2004). Serum Troponins as Biomarkers of Drug-Induced Cardiac Toxicity. Toxicologic Pathology, 32(1), 106–121.
26. Weli, S. M., Kareem, S. I., Shareef, O. H., Rauf, H. G., Raouf, C. G., & Hama, A. A. (2021). The effects of the aqueous extract of the leaves of the local plant eruca sativa on lipid profile and some minerals in the blood of male white mice. Iraqi Journal of Science, 62(4), 1088–1093.