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# Research Article Chemical and Nutraceutical Studies on Infertility of Albino Rats Induced by Cadmium Chloride

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# Abstract

**Background and Objective:** Infertility in couples is rated one in every eight couple worldwide which affects 15% of couples and a male factor is found to be solely responsible or in conjunction with a female factor in 50% of cases. The natural chemicals found in rocca and red cabbage leaves breakdown into compounds like indole-3-carbinol, which has anti-cancer property. Flavonoids of the crop have good therapeutic potential in inflammation and pain. Meanwhile, this investigation aimed to evaluate the effect of rocca leaves and red cabbage leaves on male infertility rats. **Materials and Methods:** Thirty-six adult male Sprague Dawley rats were divided into six groups. Group 1: Normal rats fed on basal diet as control negative (C<sup>-</sup>), Group 2: Control positive C<sup>+</sup>, in which infertility rats were fed on basal diet. Group 3: Infertility rats fed on basal diet and 5% rocca leaves. Group 4: Infertility rats fed on basal diet and 10% rocca leaves. Group 5: Infertility rats fed on basal diet and 5% red cabbage leaves. Group 6: Infertility rats fed on basal diet and 10% rocca leaves. At the end of experiment, after 28 days of feeding, all serum samples were analyzed for biochemical parameters. **Results:** Injection with cadmium chloride caused a significant increase in the level of glucose, urea, creatinine, uric acid, AST, ALT, ALP, total cholesterol, triglycerides, LDLc, VLDLc, Al, Glob, TB, IB, DB and LH hormone while a significant decrease was recorded in HDLc, testosterone, FSH hormones, TP and Alb. Meanwhile, in infertility rats then treated with rocca leaves 5 and 10% and red cabbage leaves at the same doses 5 and 10% caused significant improvement in all tested parameters. **Conclusion:** The obtained results demonstrated that rocca leaves and red cabbage leaves had significant improvement in testosterone, Follicle-stimulating hormone, luteinizing hormone, total protein, albumin and lipids profile in cadmium chloride induced infertility in rats.

Key words: Infertility, rocca leaves, red cabbage leaves, Luteinizing hormone, total protein, albumin, lipids profile, cadmium chloride

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Data Availability: All relevant data are within the paper and its supporting information files.

# **INTRODUCTION**

Infertility is defined as one year of regular and unprotected intercourse without conception<sup>1</sup>. Infertility affects an estimated 15% of couples globally, amounting to 48.5 million couples. Males are found to be solely responsible for 20-30% of infertility cases and contribute to 50% of cases overall<sup>2</sup>. Infertility affects 15% of couples and a male factor is found to be solely responsible or in conjunction with a female factor in 50% of cases<sup>3</sup>. Over the years, infertility has been on the increase in both males and females. The increase in male infertility however has become a source of global concern. These have been attributed to factors such as smoking, drinking of alcoholic beverages and use of restricted drugs, stress, poor nutrition and lack of exercise. Occupational risk factors, including exposure to heat, chemicals and heavy metals needs to be examined. Lifestyle and dietary choices, pesticide residues and xenoestrogens all may adversely affect spermatogenesis. Also, injury to the testes, blockage in the vas deferens, excessive heat to the testes, vitamin deficiencies and varicoele have also been associated with infertility in men<sup>4,5</sup>. Eruca sativa originated in Mediterranean countries which called as: Rocket, rocket salad, arugula, roquette and white pepper. Eruca sativa consider as a traditional medicine according to therapeutic characteristics including digestive, laxative, diuretic and stimulant<sup>6</sup>. The *E. sativa* has been recognized as a rich source of health-promoting phytochemicals, vitamins, carotenoids, fibers, minerals, glucosinolates, isothiocyanates and flavonoids such as; kaempferol, quercetin and isorhamnetin, flavanols and phenolic compounds<sup>7</sup>.

Rocket leaves being a valuable source of protein, carbohydrates, L-ascorbic acid and mineral nutrients have a high nutritional value<sup>8</sup>. Red cabbage (*Brassica oleracea* L. var. capitata L. f. rubra (L.) Thell] is a cool season leafy vegetable belonging to the group of cole crops (Brassicaceae family). The dry matter, vitamin C, total sugar and anthocyanin contents in red cabbage had been improved when integrated nutrient management was followed which increased nutritive value<sup>9</sup>. The popularity of red cabbage is for its rich content of phytochemicals, antioxidants, vitamins (C, E, A, K) and minerals (calcium, manganese, magnesium, iron and potassium) and low content of saturated fats and cholesterol. The B vitamins e.g., thiamine (B1), riboflavin (B2) and folate (B2) are also found in this crop. Besides vitamins and minerals, cabbage also contains a small amount of protein <sup>10</sup>. It protects us from cancer, premature aging, diabetes, ulcer and Alzheimer's diseases. It helps in weight loss, boosting the immune system, improving the skin and eye and detoxification of body.

Secondary plant metabolites like glucosinolates (GSs) present in red cabbage are known for the health-promoting properties<sup>11</sup>. These natural chemicals breakdown into compounds like indole-3-carbinol, which has anti-cancer property. Flavonoids of the crop have good therapeutic potential in inflammation and pain<sup>12</sup>. This study was aimed to evaluate the effect of rocca leaves and red cabbage leaves on male rat infertility.

## **MATERIALS AND METHODS**

# Materials

**Study area:** Rats were purchased from Research Institute of Ophthalmology, Department of Medical Analysis, Giza, Egypt. Meanwhile, the experiment was done in Animal Lab., at Faculty of Home Economics. Menofia University, Shebein El-Kom, Egypt. The research was conducted from April, 2019 to January, 2020. Feeding trial lasted, however, for 45 days.

**Plants leaves:** Rocca leaves and red cabbage were obtained dry from herb shop in Cairo, Egypt.

**Materials:** Rocca leaves and red cabbage were obtained dry from herb shop in Cairo, Egypt.

**Chemicals:** Cadmium Chloride Hydrate (CdCl<sub>2</sub>, 2.5 H<sub>2</sub>O) was purchased from Merck Chemical Company, product of Germany.

**Rats:** Thirty six adult male Sprague Dawley rats, average body weight ( $150\pm10$  g) were used in this study. Rats were obtained from Research Institute of Ophthalmology, Department of Medical Analysis, Giza, Egypt.

**Animals:** Thirty six adult male Sprague Dawley rats, average body weight  $(150\pm10 \text{ g})$  were used in this study. Rats were obtained from Research Institute of Ophthalmology, Department of Medical Analysis, Giza, Egypt.

# Methods

**Basal diet composition of tested rats:** The basal diet in the experiment consisted of casein (12%), corn oil (10%), mineral mixture (4%), vitamin mixture (1%), cellulose (5%), chorine chloride (0.2%), methionine (0.3%) and the remained is corn starch (67.5%), according to American Institute of Nutrition<sup>13</sup>.

**Preparation of plants leaves:** All materials were milled to soft powder by using electric grinder and kept in dusky stoppered glass bottles in a cool and dry location till use according to Russo<sup>14</sup>.

**Induced infertility for rats:** Rats were injected by Cadmium Chloride (CdCl<sub>2</sub>, 0.1%) at 0.1 mL/100 g b.wt., to induce male infertility for rats.

**Experimental design and animal groups:** Rats were housed in wire cages under the normal laboratory condition and were fed on basal diet for a week as an adaptation period. The rats were divided into 6 groups each of 6 rats. All groups of rats were housed in wire cages at room temperature 25°C and kept under normal healthy condition. Rats were divided into the following groups:

- **Group 1:** Control negative (-) group in which normal rats were fed on basal diet
- **Group 2:** Control positive (+) group in which infertility rats were fed on basal diet
- Group 3: Infertile rats fed on rocca leaves 5%
- **Group 4:** Infertile rats fed on rocca leaves 10%
- **Group 5:** Infertile rats fed on red cabbage leaves 5%
- Group 6: Infertile rats fed on red cabbage leaves 10%

**Determination of biochemical blood parameters:** Blood samples were collected after 12 h fasting at the end of experiment using the abdominal aorta. The rats were scarified under ether anaesthesia. Blood samples were received into in clean dry centrifuge tubes, in which blood was left to clot at room temperature and then centrifuged for 10 min at 3000 rpm to separate the serum. Serum was carefully aspirated and transferred into clean cuvette tubes and stored frozen at -20°C for biochemical analysis as described by Schermer<sup>15</sup>. All serum samples were analyzed for determination the following parameters:

Urea was determined according to the enzymatic method of Patton and Crouch<sup>16</sup>, creatinine was determined according to kinetic method of Henry et al.<sup>17</sup> and uric acid was according to the enzymatic colorimetric test of Kageyama<sup>18</sup>. Aspartate amino transaminase (AST) and alanine amino transferase (ALT) were carried out according to the method of Suzuki<sup>19</sup> and Tietz<sup>20</sup>. Alkaline phosphatase (ALP) was determined according to Belfield and Goldberg<sup>21</sup>. Total Cholesterol (TC) was determined according to Allain et al.22 and High Density Lipoprotein-cholesterol (HDL-c) according to Lopes-Virella et al.23. The calculation of low density lipoprotein cholesterol (LDL-c) was carried out according to the method of Lee and Niemann<sup>24</sup>, Atherogenic Index (AI) was calculated according to Kikuchi-Hayakawa et al.25 and triglyceride according to Fossati and Prencipe<sup>26</sup>. Serum glucose determined according to Kaplan and Pesce<sup>27</sup>. Testosterone hormone was determined colorimetrically according to the method of Pradelles *et al.*<sup>28</sup>. Luteinising Hormone (LH) and Follicle Stimulating Hormone (FSH) were determined colorimetrically according to the method of Akram *et al.*<sup>29</sup>. Serum Total Protein (TP) determined according to Buzanovskii<sup>30</sup>, serum albumin was carried out to the method of Doumas *et al.*<sup>31</sup> and globulin was calculated as the method of Charry and Sharma<sup>32</sup>. Bilirubin was determined according to Doumas *et al.*<sup>33</sup>. Finally, direct bilirubin and indirect bilirubin were measured according to Sepulveda and Osterberg<sup>34</sup>.

**Statistical analysis:** The data were statistically analyzed using a computerized Costat Program by one way ANOVA using a Completely Randomized Factorial Design<sup>35</sup> when a significant mean effect was detected, the means were separated with the Duncan's Multiple Range Test. Differences between treatments at p<0.05 were considered significant. The results are presented as Mean±SD.

# RESULTS

Data presented in Table 1 illustrated the effect of rocca leaves and red cabbage leaves, on total cholesterol and triglycerides of infertility rats. It could be observed that the mean value of Total Cholesterol (TC) of control (+) group was higher than control (-) group, being  $176 \pm 4.81$  and  $110 \pm 2.09$  mg dL<sup>-1</sup>, respectively. The best serum (TC) level was showed for groups 4, 6 (rats fed on basal diet containing 5% rocca leaves and 10% red cabbage leaves) when compared to control (+) group.

It could be noticed that the mean value of triglycerides TG of control (+) group was higher than control (-) group, being  $130\pm3.71$  and  $110\pm1.36$  mg dL<sup>-1</sup>, respectively. The best serum (TG) level was showed for group 4 (rats fed on basal diet+10% rocca leaves) when compared to control (+) group.

Data presented in Table 2 show the effect of rocca leaves and red cabbage leaves on HDLc, LDLc, VLDLc and AI of infertility rats.

Table 1:	Effect of rocca leaves and red cabbage leaves on Total Cholesterol (TC)
	(mg dL <sup><math>-1</math></sup> ) and triglycerides (TG) (mg dL <sup><math>-1</math></sup> ) of infertility rats

	Parameters (Mean :	± SD)
Groups	TC (mg dL <sup>-1</sup> )	TG (mg dL <sup>-1</sup> )
G1: Control (-ve)	110±2.09 <sup>d</sup>	110±1.36 <sup>e</sup>
G2: Control (+ve)	176±4.81ª	130±3.71ª
G3: Rocca leaves (5%)	125±2.11°	127±2.38°
G4: Rocca leaves (10%)	110±3.33 <sup>d</sup>	120±2.42 <sup>d</sup>
G5: Red cabbage leaves (5%)	134±2.68 <sup>b</sup>	129±3.31 <sup>b</sup>
G6: Red cabbage leaves (10%)	110±2.03 <sup>d</sup>	126±2.98°

TC: Total cholesterol, TG: Triglycerides, Values denote arithmetic Means  $\pm$  standard error of the mean, <sup>ab.c.d.e</sup>Different letters in the same column indicate significant differences among groups (p<0.05)

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Table 2: Effect of rocca leaves and red cabbac	ge leaves on HDLc, LDLc, '	VLDLc (mg dL $^{-1}$ ) and A	Atherogenic Index (AI) of infertility rats

	Parameters (mg dL	)		
Groups	HDL <sub>c</sub>	LDL <sub>c</sub>	VLDL <sub>c</sub>	AI
G1: Control (-ve)	58±0.88ª	30±0.21 <sup>d</sup>	22±0.33 <sup>d</sup>	0.90±0.02 <sup>d</sup>
G2: Control (+ve)	50±1.91 <sup>d</sup>	100±1.92ª	26±1.81ª	2.52±0.05ª
G3: Rocca leaves (5%)	55±0.97 <sup>b</sup>	45±0.51°	25±0.23 <sup>b</sup>	1.27±0.03°
G4: Rocca leaves(10%)	58±0.62ª	28±0.91 <sup>d</sup>	24±0.46°	$0.90 \pm 0.02^{d}$
G5: Red cabbage leaves (5%)	53±1.12°	55±0.28 <sup>b</sup>	26±0.55ª	1.53±0.03 <sup>b</sup>
G6: Red cabbage leaves (10%)	56±1.10 <sup>b</sup>	29±0.11 <sup>d</sup>	25±0.08 <sup>b</sup>	$0.96 \pm 0.01^{d}$

HDLc: High density lipoproteins cholesterol, LDLc: Low density lipoproteins cholesterol, VLDLCc Very low-density lipoproteins cholesterol, AI: Atherosclerosis index, Values denote arithmetic mean±standard error of the mean, <sup>abc,de</sup>Different letters in the same column indicate significant differences among groups (p≤0.05)

Table 3: Effect of Rocca leaves and re	d cabbage leaves on	total protein, albumin	, Globulin and albumin	/globulin (A/G) of infertil	ity rate
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	Parameters (g dL <sup>-1</sup> )			
Groups	ТР	Alb.	Glob.	A/G
G1: Control (-ve)	8.30±0.07ª	4.80±0.01 <sup>b</sup>	3.50±0.02 <sup>b</sup>	1.37±0.003 <sup>e</sup>
G2: Control (+ve)	6.00±0.09°	1.10±0.04 <sup>e</sup>	4.90±0.03ª	0.23±0.005 <sup>e</sup>
G3: Rocca leaves (5%)	6.50±0.04 <sup>c</sup>	$4.80 \pm 0.03^{b}$	1.70±0.02 <sup>e</sup>	$2.82 \pm 0.003^{b}$
G4: Rocca leaves (10%)	6.60±0.03 <sup>b</sup>	4.90±0.02ª	1.70±0.03 <sup>e</sup>	2.88±0.002ª
G5: Red cabbage leaves (5%)	6.30±0.02 <sup>d</sup>	$4.50 \pm 0.02^{d}$	1.80±0.04 <sup>d</sup>	2.50±0.004°
G6: Red cabbage leaves (10%)	6.50±0.01°	4.60±0.01°	1.90±0.01°	$2.42 \pm 0.001^{d}$

TP: Total protein, Alb: Albumin, Glob Globulin, A/G: Albumin/globulin, Values denote arithmetic mean  $\pm$  standard error of the mean, <sup>a,b,c,d,e</sup>Different letters in the same column indicate significant differences among groups (p<0.05)

It could be observed that the mean value of HDLc of control (+) group was lower than control (-) group being  $50\pm1.91$  and  $58\pm0.88$  mg dL<sup>-1</sup>, respectively. The best serum HDLc was shown for group 4 (rats fed on basal diet+10% rocca leaves) when compared to control (+) group.

It could be showed that the mean value of LDLc of control (-) group was lower than control (+) group being  $30\pm0.21$  and  $100\pm1.92$  mg dL<sup>-1</sup>, respectively. The best serum LDLc was shown for group 4 (rats fed on basal diet containing 10% rocca leaves) when compared to control (+) group.

The data indicated that the mean value of VLDLc of control (+) group was higher than control (-) group, being  $26\pm1.81$  and  $22\pm0.33$  mg dL<sup>-1</sup>, respectively. The best serum VLDLc was shown for group 4 (rats fed on basal diet+10% rocca leaves) when compared to control (+) group.

Also, data of Table 2 observed that the mean value of Al of control (+) group was higher than control (-) group being  $2.52\pm0.05$  and  $0.90\pm0.02$  mg dL<sup>-1</sup>, respectively. The best Al was shown for group 4 (rats fed on basal diet+10% rocca leaves) when compared to control (+) group.

Data presented in Table 3 show the effect of rocca leaves and red cabbage leaves on Total Protein (TP), albumin (Alb), globulin (Glob) and albumin/globulin (A/G) of infertility rats.

It could be indicated that the mean value of TP of control (+) group was lower than control (-) group being  $6.00\pm0.09$  and  $8.30\pm0.07$  g dL<sup>-1</sup>, respectively. The best serum TP was showed for group 4 (rats fed on basal diet+10% rocca leaves) when compared to control (+) group.

It could be shown that the mean value of Alb. of control (+) group was lower than control (-) groups, being  $1.10\pm0.04$  and  $4.80\pm0.01$  g dL<sup>-1</sup>, respectively. The best serum Alb. showed for group 4 (rats fed on basal diet+10% rocca leaves) when compared to control (+) group.

Data indicated that the mean value of Glob of control (+) group was higher than control (-) group, being  $4.9\pm0.03$  and  $3.50\pm0.02$  g dL<sup>-1</sup>, respectively. The best serum Glob showed for group 6 (rats fed on basal diet containing 10% red cabbage leaves) when compared to control (+) group.

Also, it was observed that the mean value of A/G of control (+) group was less than control (-) group being  $0.23\pm0.005$  and  $1.37\pm0.003$  g dL<sup>-1</sup>, respectively. The best A/G was showed for group 4 (rats fed on basal diet containing 10% rocca leaves) when compared to control (+) group.

Data presented in Table 4 illustrate the mean value of Total Bilirubin (TB), Direct Bilirubin (DB) and Indirect Bilirubin (IB) of infertility rats fed on rocca leaves and red cabbage leaves.

It could be noticed that the mean value of TB of control (-) group was lower than control (+) group, being  $0.19\pm0.003$  and  $0.49\pm0.009$  mg dL<sup>-1</sup>, respectively. Infertility rats fed on basal diet containing 10% rocca leaves (group 4) showed the best treatment of TB when compared to control (+) group.

According to data presented in the Table 4, it could be revealed that the mean value of DB of control (-) group was lower than control (+) group being  $0.07\pm0.002$  and  $0.10\pm0.004$  mg dL<sup>-1</sup>, respectively. The best DB was recorded

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Table 4: Effect of rocca leaves and red cabbag	e leaves on total bilirubin, d	lirect bilirubin and indirect bilirubin o	of infertility rats

	Parameters (mg dL <sup>-1</sup> )			
Groups	ТВ	DB	IB	
G1: Control (-ve)	0.19±0.003 <sup>e</sup>	0.07±0.002 <sup>d</sup>	0.12±0.004 <sup>d</sup>	
G2: Control (+ve)	0.49±0.009ª	0.10±0.004ª	$0.39 \pm 0.009^{a}$	
G3: Rocca leaves (5%)	0.26±0.002°	0.08±0.002°	0.18±0.003 <sup>bc</sup>	
G4: Rocca leaves (10%)	0.24±0.003 <sup>d</sup>	$0.07 \pm 0.003^{d}$	0.17±0.002 <sup>c</sup>	
G5: Red cabbage leaves (5%)	0.28±0.004 <sup>b</sup>	0.09±0.002 <sup>b</sup>	0.19±0.003 <sup>b</sup>	
G6: Red cabbage leaves (10%)	0.25±0.002 <sup>cd</sup>	0.08±0.001°	0.17±0.001°	

TB: Total bilirubin, DB: Direct bilirubin, IB: Indirect bilirubin, Values denote arithmetic mean ± standard error of the mean, <sup>abc.de</sup>Different letters in the same column indicate significant differences among groups (p<0.05)

Table 5: Effect of rocca leaves	and red cabbage leaves on	AST, ALT, AST/ALT ar	d ALP of infertility rats
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	Parameters (U $L^{-1}$ )			
Groups	AST	ALT	ALP	AST/ALT
G1: Control (-ve)	27±1.11 <sup>d</sup>	20±0.56°	70±1.05 <sup>e</sup>	1.35±0.03 <sup>e</sup>
G2: Control (+ve)	96±3.02ª	52±1.45ª	204±3.88ª	1.85±0.06ª
G3: Rocca leaves (5%)	30±2.01°	19±1.03 <sup>cd</sup>	92±1.33°	1.58±0.02°
G4: Rocca leaves (10%)	27±1.16 <sup>d</sup>	18±1.23 <sup>d</sup>	71±2.11 <sup>e</sup>	$1.50 \pm 0.03^{d}$
G5: Red cabbage leaves (5%)	37±1.23 <sup>b</sup>	22±0.72 <sup>b</sup>	98±1.87 <sup>b</sup>	1.68±0.02 <sup>b</sup>
G6: Red cabbage leaves (10%)	30±0.88°	20±0.22°	$80 \pm 1.15^{d}$	$1.50 \pm 0.01^{d}$

AST: Aspartate amino transaminase, ALT: Alanine amino transferase, ALP: Alkaline phosphatase, AST/ALT: Aspartate amino transaminase/alanine amino transferase, Values denote arithmetic Mean±standard error of the mean, <sup>a,b,c,de</sup>Different letters in the same column indicate significant differences among groups (p<0.05)

Table 6: Effect of rocca leaves and red cabbage leaves on serum glucose of

infertility rats	
Groups	Glucose (mg dL <sup>-1</sup> )
G1: Control (-ve)	112±1.82 <sup>b</sup>
G2: Control (+ve)	192±3.61ª
G3: Rocca leaves (5%)	95±1.59 <sup>d</sup>
G4: Rocca leaves (10%)	77±2.62 <sup>f</sup>
G5: Red cabbage leaves (5%)	98±1.45°
G6: Red cabbage leaves (10%)	79±1.01°

Values denote arithmetic means $\pm$ standard error of the mean, <sup>a,b,c,d,e</sup>Different letters in the same column indicate significant differences among groups (p $\leq$ 0.05)

for group 4 (basal diet containing 10% rocca leaves) when compared to control (+) group.

Findings observed that the IB of control (-) group was lower than control (+) group, being  $0.12\pm0.004$  and  $0.39\pm0.009$  mg dL<sup>-1</sup>, respectively. Infertility rats fed on basal diet containing 10% rocca leaves and 10% red cabbage leaves (group 4, 6) showed the best IB as compared to control (+) group.

Data of Table 5 show the effect rocca leaves and red cabbage leaves on serum levels of AST, ALT, ALP enzymes and AST/ALT ratio of infertility rats.

It could be noticed that the mean value of AST enzyme of control (+) group was higher than control (-) group, being  $96\pm3.02$  and  $27.0\pm1.11$  U L<sup>-1</sup>, respectively. The best treatment was observed for group 4 (basal diet containing 10% rocca leaves) when compared to control (+) group.

It could be noticed that the mean value of ALT enzyme of control (+) group was higher than control (-) group, being  $52\pm1.45$  and  $20\pm0.56$  UL<sup>-1</sup>, respectively. The best treatment was observed for group 4 (basal diet containing 10% rocca leaves) when compared to control (+) group.

Data of the Table 5 illustrate the mean value of ALP enzyme of control (+) group was higher than control (-) group, being  $204\pm3.88$  and  $70\pm1.05$  U L<sup>-1</sup>, respectively. Group 4 showed the lowest mean value of ALP enzyme level as compared to control (+) group which and recorded the best result.

It could be noticed that the mean value of AST/ALT of control (+) group was higher than control (-) group, being  $1.85\pm0.06$  and  $1.35\pm0.03$  U L<sup>-1</sup>, respectively. The best treatment was observed for group 4 and 6 when compared to control (+) group.

Data presented in Table 6 show the effect of rocca leaves and red cabbage leaves on serum glucose of infertility rats. It could be noticed that the mean value of glucose of control (+) group was higher than control (-) group, being  $192\pm3.61$  and  $112\pm1.82$  mg dL<sup>-1</sup>, respectively. The best serum glucose was observed for group 4 (basal diet containing 10% rocca leaves) when compared to control (+) group.

Results of Table 7 show the mean value of serum creatinine, urea and uric acid mg  $dL^{-1}$  on infertility rats fed on various diets.

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Table 7: Effect of rocca	leaves and red cabba	ge leaves on creatinine	, urea and uric acid	of infertility rats
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Groups	Parameters (mg dL <sup>-1</sup> )			
	Creatinine	Urea	Uric acid	
G1: Control (-ve)	0.74±0.02 <sup>d</sup>	31±0.73 <sup>e</sup>	2.80±0.03 <sup>b</sup>	
G2: Control (+ve)	2.50±0.08ª	80±1.69ª	$5.00 \pm 0.05^{a}$	
G3: Rocca leaves (5%)	1.00±0.04°	43±1.11 <sup>b</sup>	2.50±0.09°	
G4: Rocca leaves (10%)	0.60±0.03 <sup>e</sup>	34±1.45 <sup>d</sup>	$2.00\pm0.04^{d}$	
G5: Red cabbage leaves (5%)	1.30±0.05 <sup>b</sup>	45±1.02 <sup>b</sup>	2.80±0.03 <sup>b</sup>	
G6: Red cabbage leaves (10%)	1.10±0.01°	38±0.29°	2.60±0.02°	

Values denote arithmetic means ± standard error of the mean, ab.c.d.eDifferent letters in the same column indicate significant differences among groups (p<0.05)

Table 8: Effect of rocca	leaves and red cabbac	e leaves on testosterone	e, FSH and LH of infertility	y rats
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Groups	Parameters			
	Testosterone (ng mL <sup>-1</sup> )	FSH (mIU mL <sup>−1</sup> )	LH (mIU mL <sup>-1</sup> )	
G1: Control (-ve)	5.10±0.02 <sup>d</sup>	8.10±0.03ª	3.80±0.02e	
G2: Control (+ve)	2.20±0.08 <sup>e</sup>	2.80±0.09 <sup>f</sup>	9.00±0.04ª	
G3: Rocca leaves (5%)	$5.50 \pm 0.04^{b}$	5.70±0.01°	4.30±0.03 <sup>d</sup>	
G4: Rocca leaves (10%)	5.80±0.03ª	5.90±0.05 <sup>d</sup>	4.10±0.03 <sup>d</sup>	
G5: Red cabbage leaves (5%)	5.30±0.05°	6.40±0.02 <sup>c</sup>	5.00±0.02℃	
G6: Red cabbage leaves (10%)	5.10±0.01 <sup>d</sup>	6.90±0.09 <sup>b</sup>	5.50±0.01 <sup>b</sup>	

FSH: Follicle-stimulating hormone, LH: Luteinizing hormone, Values denote arithmetic means  $\pm$  standard error of the mean, <sup>a,b,c,d,e</sup>Different letters in the same column indicate significant differences among groups (p<0.05)

It could be observed that the mean value of creatinine of control (+) group was higher than control (-) group, being  $2.50\pm0.08$  and  $0.74\pm0.02$  mg dL<sup>-1</sup>, respectively. Group 4 (basal diet containing 10% rocca leaves) recorded the best result as compared to control (+) group.

The same Table 7 illustrate that mean value of urea of control (+) group was higher than control (-) group, being  $80\pm1.69$  and  $31\pm0.73$  mg dL<sup>-1</sup>, respectively. In concern to urea the best treatment was recorded for the group 4 (rats fed on basal diet+0% rocca leaves) when compared to control (+) group.

It could be noticed that the mean value of uric acid of control (+) group was higher than control (-) group being  $5.00\pm0.05$  and  $2.80\pm0.03$  mg dL<sup>-1</sup>, respectively. Group 4 (rats fed on basal diet+0% rocca leaves) recorded the best result as compared to control (+) group.

Data of Table 8 illustrate the effect rocca leaves and red cabbage leaves on Testosterone (ng  $mL^{-1}$ ), FSH and LH (mIU  $mL^{-1}$ ) hormones of infertility rats.

It could be noticed that the mean value of testosterone hormone of control (-) group was higher than control (+) group being  $5.10\pm0.02$  and  $2.20\pm0.08$  ng mL<sup>-1</sup>, respectively. The best treatment was observed for group 4 (basal diet containing 10% rocca leaves) when compared to control (+) group.

It could be noticed that the mean value of FSH hormone of control (-) group was higher than control (+) group, being

 $8.10\pm0.03$  and  $2.80\pm0.09$  mIU mL<sup>-1</sup>, respectively. The best treatment was observed for group 6 (rats fed on basal diet containing 10% red cabbage leaves) when compared to control (+) group.

Data of the same Table 8 show the mean value of LH hormone of control (+) group was higher than control (-) group, being  $9.00\pm0.04$  and  $3.80\pm0.02$  mIU mL<sup>-1</sup>, respectively. Group 4 showed the best result of LH hormone as compared to control (+) group.

# DISCUSSION

Results of present work indicated that suggested rocca leaves and red cabbage leaves diets improved all evaluated parameters concerning infertility. Improvements of infertility are useful for many male individuals, especially using widespread consuming plants. Our results are in line with previous studies in concern to improvements in lipid profile, function of liver and kidney as well as infertility. Meanwhile, the present work compared between using rocca and cabbage leaves indicating superiority of red cabbage leaves specially at 10% diet.

Rocket oil reduced total lipids, TC and TG in rats fed on high cholesterol diet<sup>36</sup>. Moreover, oral administration of red cabbage extract decreased TG in hyperthyroidimic rats<sup>37</sup>. Meanwhile, rocket oil decreased LDL and increase HDL in rats fed on high cholesterol diet<sup>36</sup>. Also, red cabbage extract decreased LDL and increased HDL in hyperthyroidimic rats<sup>37</sup>. Ethanolic extract of rocket *Eruca sativa* L. reduced TP in mice treated with CCL<sub>4</sub> through its potent antioxidant activity in rats<sup>38</sup>. Furthermore, oral administration of red cabbage extract ameliorated protein profile in hyperthyroidimic rats<sup>37</sup>. As well as, ethanolic extract of Rocket *Eruca sativa* L. reduced bilirubin in mice treated with CCL<sub>4</sub> through its potent antioxidant activity in rats<sup>38</sup>. However, oral administration of broccoli and red cabbage extracts decreased total and direct bilirubin of hepatocellular carcinoma rats due to their antioxidant effect of the phenolic and flavonoids compounds<sup>39</sup>.

*Eurca Saliva* alcoholic extract reduced AST and ALT in albino mice treated<sup>40</sup> with CCL<sub>4</sub>. Meanwhile, red cabbage extract decreased AST, ALT and ALP in diabetic rats<sup>41</sup>. In addition, red cabbage extract improved fasting blood glucose level in diabetic rats<sup>41</sup>. Moreover, rocket leaves reduced blood glucose level in diabetic rats<sup>42</sup>.

Accordingly, *Eurca Saliva* alcoholic extract reduce creatinine and urea in Albino mice treated<sup>40</sup> with CCL<sub>4</sub>. Furthermore, red cabbage extract decrease creatinine and uric acid in hyperthyroidimic rats<sup>37</sup>. Therefore, *Eruca sativa* leaves showed a significant ( $p \le 0.05$ ) increase in testosterone level in mice<sup>43</sup>. As well as, red cabbage extract increased testosterone hormone levels in hyperthyroidimic as compared with normal rats because it contains indole-3-carbinol, which reduces levels of oestrogen, allowing testosterone to do its function <sup>43</sup>. Moreover, it is recommend to apply this in female rats. So, further studies should be conducted to support the relationship of eating rocca leaves and red cabbage leaves and fertility not only for males, but also for females.

# CONCLUSION

Injection with cadmium chloride caused a significant increase in the level of glucose, urea, creatinine, uric acid, AST, ALT, ALP, total cholesterol, triglycerides, LDLc, VLDLc, AI, Glob, TB, IB, DB and LH hormone while a significant decrease was recorded in HDLc, testosterone, FSH hormones, TP and Alb. In infertility rats treated with various diets, the results showed the improvement in all previous parameter.

# SIGNIFICANCE STATEMENT

This study discovered the effect of rocca leaves and red cabbage leaves on male infertility rats. that can be beneficial for infertility. Moreover, this study will help the researcher to uncover the critical areas of using rocca leaves and red cabbage leaves that many researchers were not focus and criticize.

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