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EFFECT OF USE OF (*LACTUCA SATIVA*) ON THE FUNCTION AND HISTOLOGICAL STRUCTURE OF KIDNEY IN LOCALE MALE RABBITS

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Abstract

The study was conducted to investigate the effect of use (*Lactuca sativa)* oil on the body weight, and the function of kidney through measurement of level of Blood urea nitrogen (BUN), Creatinine (Cr.) and Malondialdehyde enzyme (MAD) and histopathological changes in kidney of local male rabbits. 21 rabbits average 1026 - 1322 gram. were divided to 3 groups, every group have 7 local male rabbits, group 1 the control gives only 0.2ml normal saline orally, and group 2, 3 gives only Lettuce plant oil at 0.1 ml/kg, 0.2ml/kg respectively orally for 17day. We conducted that the administration of Lettuce oil produce a significance decrease ($P< 0.05$) at 0.2 ml/Kg. B.W. in the body weight and there is a significant increase ($P< 0.01$) in level of Creatinine and there is a significant increase ($P< 0.05$) in level of Blood urea nitrogen and Malondialdehyde enzyme. Furthermore, there is a significant decrease ($P< 0.01$) at 0.1 ml/Kg. B.W. in the level of Creatinine and there is a significant decrease ($P< 0.05$) in the level of Blood urea nitrogen and there is non significant differences ($P< 0.05$) in the Malondialdehyde enzyme. We concluded that the administration of Lettuce oil in low dose lead to produce good effects whereas administration of Lettuce oil with high dose that will produces bad effects in the function of kidneys and the body weight of local male rabbits.

Key words : Kidney, Lettuce oil, Creatinine, Rabbits.

Introduction

Lettuce plant (*Lactuca sativa*) called romaine Lettuce or cos Lettuce (Msilini *et al.,* 2013). is a vegetable plant belonging to Asteraceae family. It's often grown everywhere as a leaf vegetable. This leaf vegetable was first cultivated by the Egyptians. After first cultivation during 16th to 18th century to Europeans first saw and found many varieties and species of lettuce (Mishra *et al.,* 2010 and Gulten *et al.,* 2012). By the mide-18th century varieties were being described. Many varieties and species can still be found in gardens in the 21st century. The market demand of lettuce was first fulfilled by the Europe and North America. But nowadays the consumption of lettuce has spread tremendously throughout the world due to their medicinal importance. Lettuce is most often consumed fresh or used for salads, but also in other kinds of food like soups, sandwiches and wraps (Shah *et al.,* 2010 and ALNomaani, *et al.,* 2013). Lettuce contains different kinds of essential elements for human healthy such as potassium, sodium and calcium as well as minor source for several other vitamins and nutrients (Khater *et al.,* 2011). Many of scientific and academic data dealing with this subject refers to the effects from the content of potassium, inositol and lipophilic flavones in leaves of lettuce.In addition, these above mentioned chemical components, saponins, sterols, polyphenols, rosmarinic acid and ursolic acid have also been detected and reported in these plant species (Gutierrez *et al.,* 2009; Berrie, *et al.,* 2011 and Claudia *et al.,* 2012). The seeds of Lettuce have 96.1% from essential oils and it's leaves have 91.8% from essential oils, major volatile constituents of the seeds oil were linoleic

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acid 38.8%, oleic acid 17.9% and palmitic acid 4.9%, also main volatile constituents of fresh leaves oil were linoleic acid 37.7%, oleic acid 19.6% and palmitic acid 16.7% (Afsharypuor *et al.*, 2018). Lettuce exhibits health properties mainly due to the presence of anti-oxidant compounds (vitamins E and C, carotenoids, polyphenols) also has alongside significant fiber content and useful amounts of certain minerals (Llorach *et al.,* 2008 and Baslam *et al.,* 2013). Therefore it used for treatment of a variety of disorders such as insomnia, neurosis, dry coughs, rheumatic pain and anxiety (Araruna and Carlos, 2010; Harsha and Anilakumar, 2013). Studies have shown the health impacts of lettuce in preventing cardiovascular diseases rats and humans (Serafini *et al.,* 2002 and Nicolle *et al.,* 2004). Anti-convulsant and sedative hypnotic effects have been mentioned for the leaves of this plant (Chu *et al.,* 2002). The extraction of lettuce seeds have analgesic and anti-inflammatory activities in rats (Sayyah *et al.,* 2004). The extraction of lettuce methanol has high phenolic contents and shows strong radical scavenging activities. It was effective against to Gram positive bacteria (*Staphylococcus aureus*, *Bacillus subtilis*, *Corynebacterium* Spp., *Enterococcus faecium* and *E. faecalis*) and some Gram negative bacteria (*Klebsiella pneumoniae, Escherichia coli, Serratia marcescens, Enterobacter cloacae*, and *Acinetobacter baumannii*).Also the extraction of aqueous and methanol of lettuce exhibited anti-viral activity against HCMV and Cox-B3 viruses (Edziri *et al.,* 2011).

Materials and Methods

Study was conducted at the period first to seventh from October 2019 in the public health department of Veterinary medicine of AL-Qassim green university. Number of local male rabbits used in experiment are 21 healthy animals at 1026 – 1322gr. weight, all local male rabbits were obtained from local markets. And the rabbits were divided as a randomly into three groups, each one of them consisting from 7 animals. Were kept to one week for acclimatizing period, before start of study. The aim of study involved of use 2 concentration over 4 replications were used materials as the following:-

1. Lettuce plant oil (*Lactuca sativa*) with natural concentration 100%. Manufactured by Fabrique par. Hemani International KEPZ. Karachi-Pakistan.
2. Dosage of the local male rabbits by stomach tube for this purpose.

Experimental design

After acclimatization period, all of the local male rabbits were divided into three equal groups, each one of them have seven rabbits.

- The first group (G1), was control administered normal saline daily.
- The second group (G2), was administered Lettuce oil at 0.1 ml /Kg B.W.
- The third group (G3), was administered Lettuce oil at 0.2 ml /Kg B.W.

All local male rabbits were administrated orally for period 17 days. By using stomach tube, the experimental animals have feed with concentrated feed (Pellets), and all of them given plain water. The animals have been isolated in room temperature about 19-23Cº, with a humidity about 45-55 %, the room was washed an sterilized once in a week. The body weight of rabbits were taken in started and in the end of experiment by digital electronic balance. At end of study period, all animals were fasted to 10 hrs. And used (Diethyl ether) for anesthesia to all rabbits, and the blood samples were collected from heart puncture in non-heparinized tubes centrifuged at 4000 rpm for 10 min. (Laessig *et al.,* 1976). After separation serum from clot, using a sampler, samples were used to measurement of level of Blood urea nitrogen, Creatinine and Malondialdehyde enzyme concentration. The rabbits were sacrificed by cervical dislocation and the abdominal cavity was immediately opened, kidneys were removed and processed for histopathological studies.

Histopathological techniques

The sections were taken from kidneys tissues from different rabbits in each group immediately after sacrificed. These tissues were washed with the normal saline solution to remove blood, then fixed in 10 % neutral formalin for 24 hrs. Dehydrated in different concentration of alcohol, and processed for paraffin embedding. Sections of 5 µm thickness were cut using a rotary microtome. The sections were processed and passed through graded alcohol series stained with Haematoxylin and Eosin, cleared in xylene and examined microscopically according to (Bancroft *et al.,* 1996).

Statistical analysis

The statistical analysis was carried out for data on this experience according to Complete Randomized Design (AI-Rawi and Abdui-Aziz, 2000). In order to determine the impact of different levels of Myrtle oil in the oral dosage on the study traits of local male rabbits. Random experimental error of the unit test, which is distributed normal and independent with an average of zone and the variance is equal to Õ2e. It was the comparison between the averages by using the Duncan

test (Duncan, 195). At tow way of level of probability 1% and 5% to test the significant differences between the averages of traits and applying the statistical program (SAS, 2010).

Results

Body weight

The result of body weight (day/gram) is shown in the table 1. Results showed there was significant decrease ($p<0.05$) between G3 which body weight was 1270.59±28.96 whine compared with G1, which body weight was 1361.33±45.46.

Biochemical parameters

The result of Blood urea nitrogen level (mg/dl) is shown in table 2. The result showed there was a significant increase ($p<0.05$) of G3 which Blood urea nitrogen level was 43.24±0.24 whine compared with G1and G2 respectively, which were 30.87±0.28 and 25.84±0.25. And there was a significant decrease ($p<0.05$) of G2, which Blood urea nitrogen level was 25.84±0.25 whine compered with G1, which was 30.87±0.28. The data of Creatinine level (mg/dl) are listed in table 2. Showed a significant in crease ($p<0.01$) of G3 which Creatinine level was 0.94±0.07 whine compared with G1 and G2, which were 0.79±0.06 and 0.64±0.06 respectively. And there was a significant decrease ($p<0.01$) of G2, which Creatinine level was 0.64±0.06 whine compared with G1, which was 0.79±0.06. The result of Malondialdehyde level (µmol/dl) is shown in table 2. The results showed a significant increase ($p<0.05$) of G3 which Malondialdehyde level was 3.89±0.12 whine compared with G1 and G2 respectively, which were 2.78±0.11 and 2.48±0.34.

Histopathological examination

Light microscopic of kidney examination using H & E 400X stain in control rabbits, showed normal histological structure to renal cortex and medulla of kidney tissue respectively. Histopathological effects of Lettuce oil on kidney of treated rabbits are presented in rabbits treated with Lettuce oil at 0.1 ml/kg.B.W. for 17 days, showed in renal cortex there is mild vascular regeneration of renal tubules**.** And the section of renal medulla showed normal proliferation to epithelial cells linen tubules and there is regeneration of renal tubules**.** Histopathological effects of Lettuce oil on kidney of treated rabbits are presented in rabbits treated with Lettuce oil at 0.2 ml/kg. B.W. for 17 days, showed in renal cortex there is sever cloudy swelling of renal tubules and focal infiltration of mono nuclear leukocytes**.** And the section of renal medulla showed marked deterioration of collecting tubules and cast formation**.**

Discussion

The observed of body weight, which recorded in table 1 show there was a significant decrease in body weight between G3 compared with G1 and G2 at 0.2 ml/ Kg.B.W., may be due to present the Poly phenol compounds and lipophilic flavones in the lettuce oil and this compounds lead to increase the metabolism in the body, specially increase of catabolism of fat in the body and this lead to loss of body weight (Tia. *et al.,* 2011). The observed of decrease of Creatinine and Blood urea nitrogen which recorded in table 2. Show there was a significant decrease between G2 compared with G1and G3 at 0.1 ml/Kg.B.W. may be due to administration of lettuce oil which have poly phenol and flavonoids compounds (Rastogi and Mehrotra, 1991). Which have activity effects to protects of epithelial cells to cortex of kidney from damages such as free-radicals (Yousef *et al.,* 2009). And this compounds have nephron protective properties from free-radicals (Kudeir, 2016). And this leads to good normal function of kidney (Neooein *et al.*,2011). And this results agreement with (Caruso *et*

Table 1: Effect of use (Lettuce oil) on initial and final body weight (mean ± SE).

Parameter \ Groups	G1 Control	G2 (0.1 mg/ k.g B.W) orally	G3 (0.2mg/k.g B.W) orally
Initial body weight at 1 day/(gm).	109.64±47.64 A	1270.59±39.92 A	1308.56±15.65 A
Final body weight at 17 days/(gm).	1361.33±45.46 A	1308.56±15.60 A	1270.59±28.96 B

Different letter means significant ($p<0.05$) between groups in the same row.

Table 2: Effect of use (Lettuce oil) on Creatinine mg/dl, Blood urea nitrogen mg/dl and Malondialdehyde µmol/dl (mean ± SE).

Parameter \ Groups	G1 Control	G2 (0.1 mg/ k.g B.W) orally	G3 (0.2mg/k.g B.W) orally
Blood ureanitrogen	30.87±0.28 B	25.84±0.25 C	43.24±0.24 A
Creatinine	0.79±0.06 AB	0.64±0.06 B	0.94±0.07 A
Malondialdehyde	2.78±0.11 B	2.48±0.34 B	3.89±0.12 A

Different letter means significant ($p<0.01$) between groups in the same row.

al., 2001). When given olive leaves extracted to stressful thermally rabbits. The observed of increase of Malonaldehyed enzyme which recorded in table 2. Show there was significant increase between G3 compared with G1 and G2 at 0.2 ml/Kg.B.W. my be due to administration of lettuce oil caused increased of Malondialdehyde levels, these results are in acceptance with those obtained by other investigators (Weyers *et al.*, 2002). Malondialdehyde is a stable metabolite of free radical-mediated lipid oxidation cascade and is used widely as a marker of stress which produce by oxidation and destruction of lipids (Sahna *et al.*, 2006). Lipid oxidation in an important cause of destruction of cell membranes and is thought to participate to the development of tissue injury (Parks and Granger, 1988). Histopathological changes induce by lettuce oil at 0.2ml/K.g. B.W. which add with clearly elevated levels of kidney biochemical markers Blood urea nitrogen, Creatinine and Malonaldehyed activities, the Histopathological changes induced by lettuce oil at 0.2ml/K.g. B.W. for 17 days induce in renal cortex there is sever cloudy swelling of renal tubules and focal infiltration of mono nuclear leukocytes. And the section of renal medulla showed marked deterioration of collecting tubules and cast formation. And Histopathological effects of lettuce oil on kidney of treated rabbits are presented in rabbits treated with lettuce oil at 0.1 ml/kg.B.W. for 17 days, in renal cortex there is mild vascular regeneration of renal tubules, and the section of renal medulla showed normal proliferation to epithelial cells linen tubules, and there is regeneration of renal tubules.

Conclusion

The administration of Lettuce oil in low dose lead to produce good effects whereas administration of Lettuce oil with high dose that will produces bad effects in the function of kidneys and in the body weight of local male rabbits.

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