

Effects of different doses of aspirin on the liver and kidney functions of the female albino rats

Nuha SH. Ali

Coll. of Dentistry/ Univ. of Al- Qadisiyah

E-mail: NuhaAli@qu.edu.iq

(Received 18/1/2017, Accepted 2/5/2017)

Abstract

The confer study was carried out to examines in detail the effect of Aspirin on the liver and kidney function of the female albino rats. (15) mature female albino rats (150-170 gm) were divided in to three groups :1st group which treated as negative control group ,it was drenched only 0.2 ml of PBS.2nd group was drenched with Aspirin (10mg/kg of B.W once daily for 30 days).3rd group was drenched with Aspirin (20mg/kg of B.W once daily for 30 days).Our results showed that 2nd and 3rd groups demonstrated significantly increased ($P \leq 0.05$) in the values of plasma GOT and GPT enzymes compared with control group. In the values of plasma total protein, the 2nd and 3rd groups showed meaningfully decreased ($P \leq 0.05$) in the relation with the control group. The values of plasma creatinine and uric acid, the 2nd and 3rd groups showed significantly increased ($P \leq 0.05$) as compared with control group. On the other hand, the microscopic examination of the histopathological sections of livers of all the treated and control groups showed that the 2nd group demonstrated congestion of central vein and presence of radially arrangement of hepatocyte around the central vein ,mild dilation of sinusoids ,mild hemorrhage and mild infiltration of inflammatory cells. In 3rd group, the histopathological examination of livers showed infiltration of inflammatory cells mainly macrophage and lymphocytes, congestion and hemorrhage in the hepatic tissue and vaculation of hepatocyte. In kidneys of 2nd group showed high cellularity of the glomeruli, mild infiltration of inflammatory cells, mild hemorrhage in the renal tissue and few dilation of renal convoluted tubule. The kidneys section of 3rd group showed atrophy and small in size of the glomeruli with marked dilation in the renal convoluted tubules. In addition, there is degeneration in the epithelial cells which lining the renal tubules and hemorrhage along the renal tissue.

Keywords: Aspirin, liver, kidney.

Introduction:

Aspirin was the first exposed associate of the period of the non-steroidal anti-inflammatory drugs, not all of which are salicylates; although they all have similar effects and a alike instrument of act Aspirin a drug in the intimate of salicylates is often used as analgesic, antipyretic and as an anti-inflammatory. It also has an anti-platelets effect which in long-standing beside low-dose can avoid heart bouts and thrombus creation in hyper disputable states (e.g Cancer). Stumpy dose and long term Aspirin use irreversibly blocks the creation of thromboxane A2 in platelets, producing an inhibitory effect on platelets aggregation, this anticoagulant property makes it convenient for reducing the incidence of heart attacks (1). Liver plays a vital role in cleaning and transforming chemicals but some medical

agents can damage the organ if given therapeutic or high dosage. Non-medicinal agents inducing industrial and environmental chemicals also lead to hepatotoxicity and are called hepatotoxins. Liver metabolizes the xenobiotics by the reduction in fat solubility and alters the biological activity after chemical transformation; some biochemical markers such as bilirubin, alanine transferase and alkaline phosphate indicate the normal function of hepatocytes or liver damage (2). The normal functions of liver can be changed by the actions of toxins or infections (3). Aspirin is a anodyne drug at stumpy doses but also it has severe side effects when managed at high doses. Long-term calming administration of Aspirin is accompanying with nephrotoxicity. Hepatotoxicity, gastrointestinal ulcerations, and uniform

renal cell cancer due to its adverse effects on multiple organ systems (4,5). Aspirin can cause adverse effects in pregnancy (6). In vitro and in-vivo studies showed that Aspirin at high doses instigated decrease of the blood vessel tissues (7). The inhibitory action of aspirin also found on the endocrine hormones. ACTH, cortisol, prolactin and

growth hormone via probable stimulatory role of prostaglandin. Also, overdose of Aspirin stimulate corticosteroid discharge by the adrenal cortex (5). The purpose of the current study is to observe the crosswise special effects of Aspirin (10 and 20mg/kg of B.W) on the liver and kidney functions of the female albino rats.

Materials and Methods:

Experimental animals:

Fifteen of female albino rats (150-170) gm were attained from animal house of Vet. Med. College of AL-Qadisiyah University and prior to use, were adapted for 7 days 12 hr. light/ dark cycle. The animals were housed in plastic cages in an air-conditioned room with temperature maintained at $25 \pm 2^\circ\text{C}$. Rats were given sterile food pellets and water ad libitum. All rats were randomized in to three groups of 5 rats each one and were treated as below for (30 days):

1-First group (1st): which treated as negative control group, it was wet only 0.2 ml of PBS.

2-Second group (2nd): was wet with Aspirin (10mg/Kg B.W once daily for 30 days).

3-Third group (3rd): was wet with Aspirin (20 mg/Kg B.W once daily for 30 days).

Chemicals:

Aspirin: this drug was obtained from the market. (400mg) of Aspirin powder (molecular weight 180.157g/mol, Sigma-Aldrich, USA) prepare by dissolving with (100) ml of 4% carboxymethyl cellulose to prepare the stock solution of Aspirin (8).

Blood collection:

Blood collection was done at 30 days of experiment via the abdominal vein. The collected bloods were collected in test tube with anticoagulants that allowed coagulating for 15 min in refrigerator. Serum was

disconnected from viscous blood testers by centrifugation at 3000 rpm for 15 min, and then saved in the frozen at -20°C until using to assessment GOT, GPT, Total protein, creatinine and uric acid by using an automatic analyser (Reflotron® plus system, Roche, Germany).

Histopathology:

After (30) days, the animals were sacrificed by the inspiration of chloroform in the Colton swab put in closed container. Specimens were taken from the uteri and ovaries and the tissues were kept in 10% formaldehyde solution for fixation, and then processed routinely by using the histokinette (SLEE medical/Germany). Tissue sections were embedded in paraffin wax, and sectioned by rotary microtome and all sections were stained with hematoxylin and eosin stain (9).

Statistical analysis

All the grouped data were statistically analyzed by SPSS program, Version 17 software (2010). Testing methods including one way ANOVA for comparisons among groups followed by least significant differences (LSD) test for comparison between two groups. P values of $p \leq 0.05$ were considered to record statistical significance, all data were expressed as mean $\pm$ standard error (SE) (10).

Results:

A-Biochemical parameters:

Our results showed that 2nd group and 3rd group demonstrated significantly increase ($P \leq 0.05$) in plasma GOT and GPT enzymes (174.16 ± 4.52) and (156.16 ± 3.98) for GOT and (116.66 ± 5.41) and (163.16 ± 0.79) for GPT compared with control group as in table (1). In the values of plasma total protein showed significantly decreased ($P \leq 0.05$) in

the 2nd and 3rd groups (64.24 ± 0.67), (65.33 ± 0.91) compared with control group. Finally, the present study showed that 2nd and 3rd group demonstrated significantly increase ($P \leq 0.05$) in plasma uric acid and creatinine (2.93 ± 0.33) and (3.93 ± 0.39) for uric acid and (0.46 ± 0.02), (0.55 ± 0.02) for creatinine concentration compared with control group.

Table (1): Effects of Aspirin on plasma GOT, GPT, Total protein, uric acid and creatinine concentration in studied groups.

Test Groups	GOT(mg/dl)	GPT(mg/dl)	Total protein mg/dl	Uric acid mg/dl	Creatinine mg/dl
1 st group	138.66±4.09 a	71.16±2.92 a	73.66±1.2 a	1.36±0.04 a	0.36±0.02 a
2 nd group	174.16±4.52 b	116.66±5.41 b	64.24±0.67 b	2.93±0.23 b	0.46±0.02 b
3 rd group	156.16±3.98 c	163.16±0.79 c	65.33±0.91 b	3.93±0.39 c	0.55±0.02 c

-Results expressed as mean± S.E.

-Different letters refer to significant differences between groups at ($P \leq 0.05$).

-Similar letters refer to no significant differences between groups at ($P \leq 0.05$).

-1st : control group.

-2nd : first treatment group gavages with Aspirin at a dose 10 mg/Kg of B.W.

-3rd : second treatment group gavages with Aspirin at a dose 20 mg/Kg of B.W.

B-Histopathological changes

1-Livers:

In the 2nd group, there is congestion of central vein and presence of radially arrangement of hepatocyte around the central vein. The hepatocyte showed normal hexagonal shaped, mild dilation of sinusoids, mild haemorrhage, proliferation of kuffer cells and mild infiltration of inflammatory cells(as in figure 2,3). On the other hands, the liver histological sections of 3rd group animals showed infiltration of inflammatory cells mainly macrophage and lymphocyte, congestion and haemorrhage in the hepatic tissue, vaculation of hepatocyte. Also, other sections showed congestion of central vein and degeneration of hepatocyte(s in fig 4,5). The liver histological sections of control group animals showed normal radially arrangement of hepatocyte around the central veins and the hepatocyte showed normal

prominent nuclei and esinophilic cytoplasm (as in fig1).

2-Kidneys:

In the 2nd group, the kidneys sections showed high cellularity of the glomeruli, circled and rounded. There is insignificant access of inflammatory cells, mild bleeding in the renal tissue and few dilation of renal convoluted tubule (in fig 7,8). In the other hands, the 3rd group showed atrophy and small in the size of the glomeruli with marked dilation in the renal convoluted tubules. There is degeneration in the epithelial cells which lining the renal tubules, also there is haemorrhage along the tissue (in fig9,10). The kidneys of control group animals showed normal and rounded with high cellularity glomeruli and normal renal convoluted tubule which lining with normal low columnar epithelium (in fig 6).

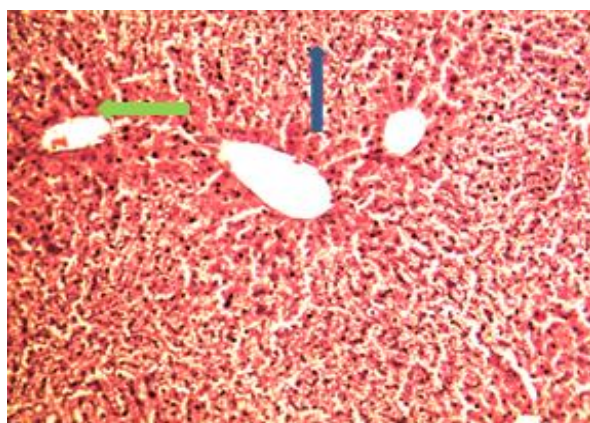


Fig. (1): Control liver: There is normal radially arrangement of hepatocyte around the central veins (blue arrows) and the hepatocyte showed normal prominent nuclei and eosinophilic cytoplasm.10XH&E

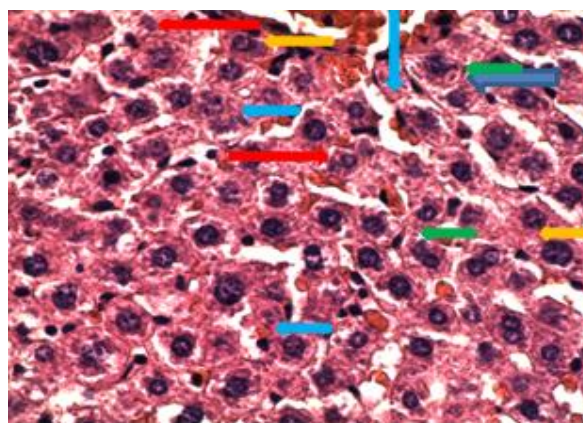


Fig.(2):T1 liver: The hepatocyte showed normal hexagonal shaped, mild dilation of sinusoids, mild hemorrhage and proliferation of kuffer cells (red arrows) .Mild infiltration of inflammatory cells (blue arrows). 40XH&E.

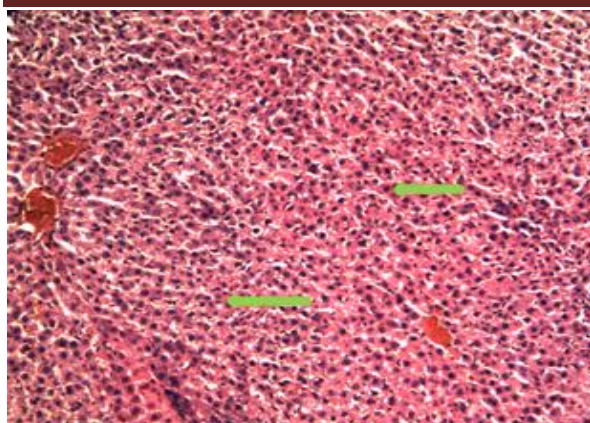


Fig. (3):T1 liver: There is congestion of central vein (green arrows) and presence of radially arrangement of hepatocyte around the central vein.10XH&E.

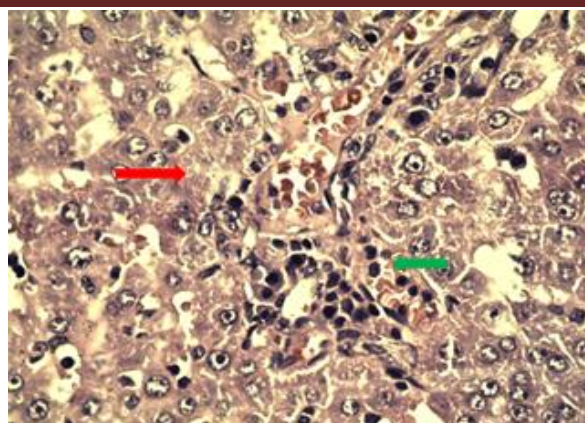


Fig. (4):T2 liver: There is infiltration of inflammatory cells mainly macrophage and lymphocyte (green arrows), congestion and hemorrhage in the hepatic tissue (red arrows), vacuolation of hepatocyte (blue arrows), dilation of sinusoids (orange arrows) with proliferation of kuffer cells.40XH&E

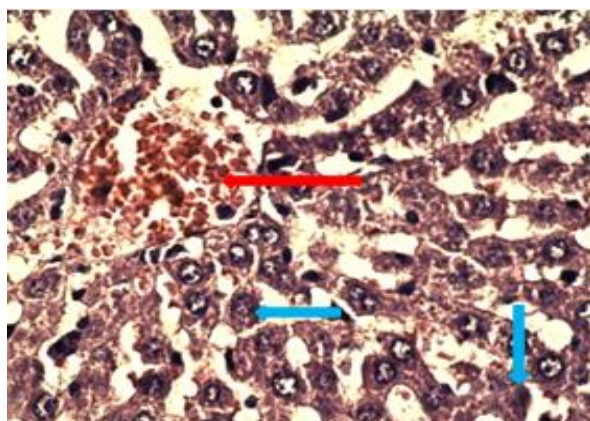


Fig. (5):T2 liver: This section show congestion of central vein (red arrow), degeneration of hepatocyte (blue arrows). 40XH&E.

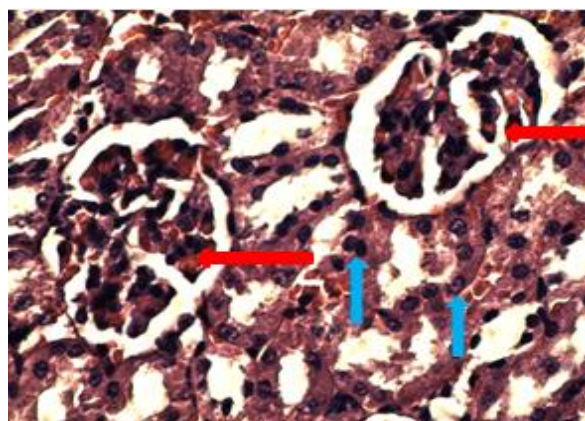


Fig. (6): Control kidney: There is normal circled and rounded with high cellularity glomeruli (red arrows) and normal renal convoluted tubule which lining with normal low columnar epithelium (blue arrows).40XH&E.

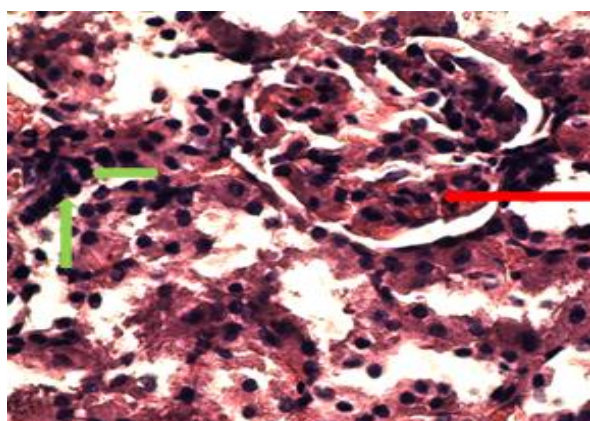


Fig. (7): T1 kidney: There is infiltration of inflammatory cell, mainly macrophages (green arrows) and high cellularity glomerulus (red arrow).40XH&E

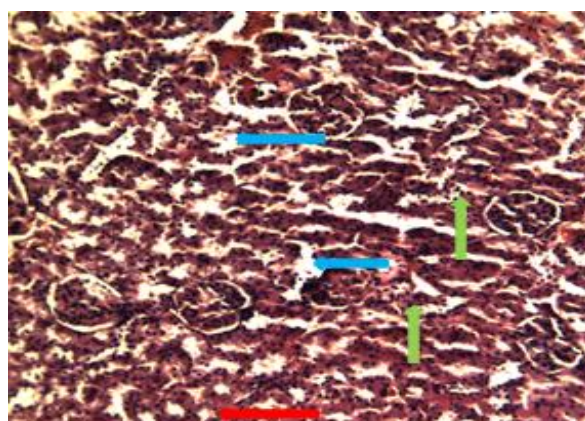


Fig. (8): T1 kidney: The glomeruli showed high cellularity, circled and rounded ,mild infiltration of inflammatory cells (green arrows),mild hemorrhage in the renal tissue(red arrow) and few dilation of renal convoluted tubule(blue arrows).10XH&E.

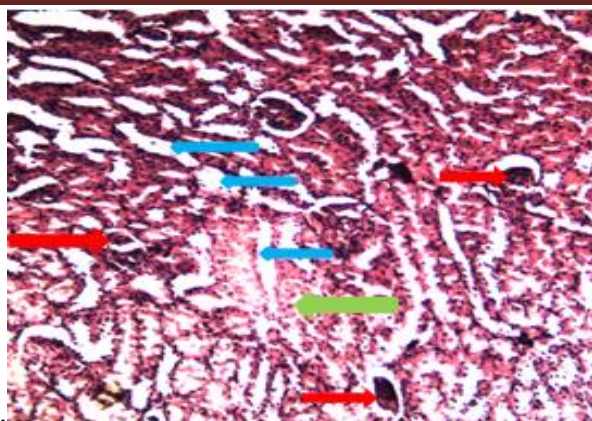


Fig. (9):T2 kidney: There is marked atrophy of glomeruli (red arrows) and dilation in renal convoluted tubule (blue arrows) with hemorrhage along the renal tissue (green arrow).10XH&E.

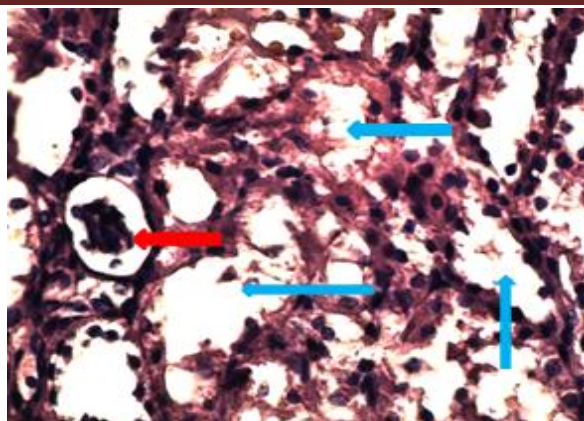


Fig. (10):T2 kidney: The glomeruli showed atrophy and small in size (red arrow) with marked dilation in the renal convoluted tubules (blue arrows), there is degeneration in the epithelial cells which lining the renal tubules.40XH&E.

Discussion:

This study was designed to determine the effect of Aspirin on the liver and kidney functions in the female albino rats.

A-Biochemical parameters

1-GOT, GPT, Uric acid and creatinine:

Our results showed that 3rd group, seemed a significant increase ($P \leq 0.05$) in the values of GOT, GPT, uric acid and creatinine compares with control group. The cause of these results due to the effect of high doses of Aspirin (20 mg/kg of B.W), in which Aspirin causes hepatic and renal toxicity (11). Owing to, Aspirin supervision mainly convinced vasoconstriction and smooth muscle degenerate in liver and kidney through release of the creation of different prostaglandins viz, PGE₂, PGD₂ and PGI₂ which are potent vasodilators (12,13). (14), showed that Aspirin caused significant increase in the activity of ALT and AST enzymes which probably indicate the hepatotoxic potential and tissue necrosis.

2-Total protein:

Our present results showed that 3rd group, appeared a significant decrease ($P \leq 0.05$) in the value of total protein compared with control group. This decrease agrees with (15). That decrease in serum total protein in hepatotoxicity states simply indicates the presence of para proteins or decreased antibody production. Retardance of cellular protein synthesis by salicylates has been described in (16,17). In (18), their results afford suggestion for a instrument,

instigated in the lumen of ER by which salicylates trigger inhibition of protein synthesis. Their results exhibit that inhibition of protein synthesis by salicylates associates with phosphorylation of eIF2 α . PKR-like kinase PERK was necessary to induce phosphorylation of eIF2 α and mediate reserve of protein synthesis in response to salicylates.

B-Histopathological changes

1-Liver:

Our present result of histopathological lesions in the livers of 3rd group rats showed infiltration of the inflammatory cells mainly macrophage and lymphocytes, congestion and haemorrhage in the hepatic tissue, the damage of liver due to aspirin was confirmed by elevated levels of biochemical parameters like AST, ALT and total cholesterol. The main causative factor of tissue damage is lipid peroxidation which is motivated by the reactive oxygen species (ROS) formation and this process is associated with the formation of malondialdehyde (MDA) because it is the by-product of lipid peroxidation (19,20,21,22). Our present findings of livers 2nd group rats showed slighter changes due to the animals of this group received fewer quantity of Aspirin (10mg/kg B.W) for 30 days. These results indicate, there is congestion of central vein and mild dilation of sinusoids, mild haemorrhage, proliferation of Kupffer cells and mild infiltration of inflammatory cells.

2-Kidneys:

Our present results of the histopathological lesions in the kidneys of 3rd group rats showed atrophy and small in size of the glomeruli with marked dilation in the renal convoluted tubules. There is degeneration in the epithelial cells which lining the renal tubules, also there is haemorrhage along the renal tissue. These findings were consistent with the results of previous studies (23), who exposed atrophic variations in the proximal and distal convoluted tubules in the cortex of Aspirin treated kidneys of rat. The provocative variations were observed in medulla and damaged tubular epithelial cells by ischemia due to vasoconstriction of renal arterioles. Production of prostaglandins

which is reserved by this drug and thus caused unopposed constriction of arterioles resulting in ischemia of tubules and epithelial cell death. Other workers also advised that Aspirin caused vacuolar deterioration of proximal tubules, focal tubular deterioration and considerably decreased proximal tubular per unit area, causing renal miscarriage (24,25). Our present findings of kidneys 2nd group rats showed slighter changes due to the animals of this group received fewer dose of Aspirin (10 mg/kg B.W) for 30 days. These results indicate, there is high cellularity of the glomeruli, slight access of inflammatory cells, mild bleeding in the renal tissue and few stretching of renal convoluted tubules.

References:

- 1-Macdonald S. Aspirin used to be banned in under 16 years olds. *BMJ.*, (2002); 325 (7371):988.
- 2-McClatchy A, Kenneth D. *Clinical Laboratory Medicine*, Lippincott Williams and Wilkins. (2006); Pp. 288.
- 3-Tobassum N, Qazi MA, Shah MY. Curative Potential of Kashni (*Cichorium intybus* Lin) Extract against Carbon Tetrachloride induced Hepatocellular Damage in Rats. *Pharmacology Online*. (2010); 2: 971-978.
- 4-Gilman EA, Langman MJ, Cheng KK, Lancashire RJ. Effect of anti-inflammatory drugs on overall risk of common cancer: case-control study in general practice research database, *BMJ*; .(2000); 320, p1642-1646.
- 5-Luigi DL, Laura G, Francesco R, Carlo B, Domenico C. Aspirin, exercise and pituitary hormones, *Official Journal of the American College of Sports Medicine*; (2001); p2029-2035.
- 6-Collins E, Turner G. Maternal Fetal effects of regular salicylate ingestion during pregnancy, *Lancet*; (1975); 2, p335-339.
- 7-Starke RD, Smith SC, Blair SN, Bonow RO, Brass LM et al. AHA/ACC guidelines for preventing heart attack and death in patients with atherosclerotic cardiovascular disease, *The American College of Cardiology*; (2001);104, p1577-9.
- 8-Wang Z, Hasegawa J, Wang X, Matsuda K, Tokuda T, Miura Watanabe T. Protective effects of ginger against aspirin-induced gastric ulcers in rats. *Yonago Acta medica.*, (2011);54:11-19.
- 9-Luna LG. Processing of tissue, histologic staining methods of the aimed forces institute of pathology. 3rd Ed. McGraw Hill book comp. London, New York, Toronto, Sydney. (1968); pp:12 – 31.
- 10-Leech NL, Barrett KC, Morgan GA. *IBM SPSS for intermediate statistics*. 4th ed. Taylor and Francis Group. (2011); LLC. USA.
- 11-Sherifa KA. Hepatic and renal biochemical responses to the toxicological interaction between acetylsalicylic acid and diazinon in albino Rats, *J Egypt Soc Toxicol*; (2006); 35, p1-6.
- 12-Adnan S, Janabi Al, Ahmad M, Izohyri A, Fouad K, Rubayai. Pharmacological effects of low dose of aspirin on corpus luteum functions in mature cycling female mice, *Institute of the Embryo Research and Infertility Treatment*; (2005); 10 (2), p150-162.
- 13-Vane JR. Inhibition of prostaglandin synthesis as a mechanism of action of aspirin-like drugs, *Nat New Biol*; (1971); 231, p232-235.
- 14-Oyedeji KO, Bolarinwa AF, Adeyemo CO. Effect Of Aspirin on Haematological and Plasma Biochemical Parameters in Male Albino Rats. *IOSR Journal of Dental and Medical Sciences*. (2013); 2279-0861. Volume 3, Issue 5, pp 80-83.
- 15-George RK. *Biochemistry Laboratory*. Philadelphia, (2009); www.jbc.org.
- 16-Spohn M, McColl I. *Biochim Biophys Acta*, (1980); 608, 409-421.
- 17-Kwon G, Hill JR, Corbett JA, McDaniel ML. *Mol Pharmacol*, (1997); 52, 398-405.
- 18-Silva AM, Die Wang, Komar A, Castilho BA, Williams PRG. Salicylates trigger protein synthesis inhibition in a pkr like endoplasmic reticulum kinase-dependent manner. *JBC Papers in Press*. Published. (2007); <http://www.jbc.org/cgi/doi/10.1074/jbc.M609996200>.
- 19-Mohammed Mustafa Hashim Zayni. Protective Effect of Terfezia claveryi Extract on Gentamicin-Induced oxidative stress in Rats. *Kerbala Journal of Pharmaceutical Sciences*. (2012); No. 4.

-
- 20-Gutteridge J. Clinical chemistry. (1995); 41:1819-1828.
- 21-Requena JR, Fu MX, Ahmed MU, Jenkins AJ, Lyons TJ, Thorpe SR. Nephrology Dialysis Transplantation. (1996); 11:48-53.
- 22-Richter C. Chemistry and physics of lipids. (1987); 44:175.
- 23-Talat Y, Farzana Y, Ghulam SQ. To evaluate the role of diclofenac sodium (a NSAID) on renal parenchyma of young albino rats, Pak J Pharm Sci; (2008); 21(2), p98-102.
- 24-Gray R, Peto R, Collins R *et al.* Randomized trial of prophylactic daily aspirin in British male doctors, BMJ; (1980); 296, p313-16.
- 25-Clive DM, Stoff JS. Renal syndromes associated with non-steroidal anti-inflammatory drugs, N Engl J Med; (1984); 310, p563-572.