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Histological Study of the Liver and Kidney of Albino Mice *Mus musculus* Exposed to Lead

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ABSTRACT

The present study was performed to investigate the histological effects caused by lead in the liver and kidney of male albino mice *Mus musculus*. The study was conducted on 25 mice, the animals were divided into 5 equal groups. The first group was given distilled water and used as a control group. The second and third groups were orally administered (2 and 4 mg lead acetate /kg of body weight) for two weeks. The fourth and fifth groups were orally administered (2 and 4 mg lead acetate/kg of body weight) for four weeks.

The animals were anesthetized, the (liver and kidneys) were extracted for histological studies. Histological changes which observed in the liver were, vacuolation, fatty degeneration, necrosis in some hepatocytes, congestion within central veins, hemorrhage between hepatic cords and infiltration of inflammatory cells. In the kidney the changes noted were hemorrhage in the interstitial tissue with enlargement of epithelial layer lining renal tubules, necrosis of some tubules with hyalinization and focal inflammatory cells infiltration. In this study deleterious toxic effects observed in liver and kidney.

Key words: Lead, Histological changes, liver, kidney.

دراسة نسجية لكبد وكلى الفئران البيض السويسرية *Mus musculus* المعرضة للرصاص

الملخص

أجريت الدراسة لتقييم التغيرات النسجية لتأثير الرصاص على الكبد والكلية في ذكور الفئران البيض السويسرية *Mus musculus*. استخدمت 25 فأرة، قسمت الحيوانات إلى خمـس مجـاميع (5 فـأرة لكـل مجموعة). جرعت المجموعة الأولى ماء مقطر واعتبرت مجموعة سيطرة، في حين جرعت المجموعتـان الثانية والثالثة 2 و 4 ملغم خلات الرصاص لكل كغم من وزن الجسم لمدة أسبوعين. أما المجموعتان الرابعة والخامسة فقد جرعت 2 و 4 ملغم خلات الرصاص لكل كغم من وزن الجسم لمدة أربعة أسابيع . تم تخـدير الحيوانات وأخذ الكبد والكلى وعمل لها مقاطع لدراسة التغيرات النسجية، إذ لوحظ في الكبد تنجـي وتـنكس

دهني وتنخر في بعض الخلايا الكبدية ، واحتقان في بعض الأوردة المركزية ونزف بـين الخلايـا الكبديـة وارتشاح لخلايا التهابية. أما في الكلى كانت التغيرات النسجية نزف في النسيج الخلالي مع ازدياد في حجـم الخلايا المبطنة للنبيبات الكلوية وتنخر في بعض النبيبات مع تزجج وارتشاح بؤري لخلايا التهابيـة. بينـت الدراسة حصول تغيرات مرضية في الكبد والكلى.

INTRODUCTION

Lead had been a toxic problem for human beings from the earliest time (Hurst and Martin, 2004). Despite its recognized hazards, lead continue to have widespread commercial applications as in plumbing, paints, manufacture of lead acid batteries, soldering etc. (Kosnett, 2004). Exposure to lead occur mainly through respiratory and gastrointestinal systems in acute and chronic exposure (Goyer and Clarkson, 2001).

Lead may enter foods, if they are put into improperly glazed pottery or ceramic dishes, the use of lead solder in the food canning industry and soft drink cans. Lead also can leach into drinking water from lead-soldered joints or leaded pipes in water distribution systems of individual house (Loghman, 1997; Gidlow, 2004; ATSDR, 2005).

The ingested and absorbed lead stored primarly in soft tissues and bone, but the highest concentration of lead occur within the bone, teeth, liver, lung, kidney, brain and spleen (Plumlee, 2004; Mudipalli, 2007).

Both hepatotoxicity and nephrotoxicity are known to occur in persons with exposure to heavy metals. Autopsy studies of lead exposed humans indicate that liver tissue is the largest repository 33% of lead among the soft tissue followed by kidney cortex and medulla (Goswani *et al.*, 2005 ; Lyn, 2006).

The purpose of this study is to investigate the effect of different dose of lead on liver and kidney of albino mice using histological examination.

MATERIALS AND METHODS

Healthy male albino Swiss mice *Mus musculus*, three months age, weighting (25-30)gm were housed in plastic cages under controlled conditions of temperature (25 ± 2)°C and light (14h light: 10h dark) cycle. The animals received standard diet and water *ad libitum*. The animals were obtained from the animal house of medical college / University of Mosul.

The mice were divided into five groups (n = 5 per group). The first group was considered as a control and received only distilled water, the second and third groups were administered lead acetate (2 and 4mg/kg of body weight) respectively for two weeks. The fourth and fifth groups were administered lead acetate (2 and 4mg/kg of body weight) respectively for four weeks. All groups were orally administered lead acetate once daily by gavage needle.

On completion of experiments, animals were anesthetized with chloroform, the liver and kidneys were quickly removed, fixed in buffered neutral formalin for 48 hrs. The fixed organs were dehydrated in ascending series of alcohol, cleared in xylene and embedded in paraffin wax, then 4 – 5 μm thick sections were obtained by rotary microtome and stained with Harris hematoxylin and Eosin (Luna, 1968).

RESULTS

Fig(1) showing section for liver from the control animal. The Fig.(2) illustrated section of liver after exposing the mice to (2mg/kg) lead acetate for 2 weeks, showing vacuolation in some hepatocytes, fatty degeneration in other hepatocytes, congestion within central veins and some sinusoids. After 4 weeks of ingestion the same dose the hepatocytes showed more fatty degeneration, large area of hepatic lobule occupied within vacuolar degeneration, hemorrhage between hepatic cords in addition to the congestion of the central veins and sinusoids Fig.(3).

Fig(4) showing section for normal kidney from the control animal. After exposing the mice to (2mg/kg) for 2 weeks the kidneys showed mild lesions included hemorrhage in the interstitial tissue with enlargement of epithelial cells lining renal tubules, but after 4 weeks of ingestion the same dose, the lesions were showing more hyalinization of some renal tubules with necrosis of other tubules, patches of hemorrhage and inflammatory cells mainly mononuclear cells Fig(5).

In case of the second dose (4mg/kg), the histological changes of the liver after 2 weeks of exposure Fig(6) illustrating severe fatty changes, necrosis of hepatocytes, infiltration of inflammatory cells in the interstitial tissue in addition to the congestion of the central veins. The changes after 4 weeks of exposure for the same dose, were similar to those of the lesions at 2 weeks exposure but were more severe and included the whole lobule of the liver and the normal tissue was very scanty Fig(7).

The histological changes were noted in kidney after 2 weeks of exposure to (4 mg/kg) showed hemorrhage within interstitial tissue, proliferation of epithelial cells lining renal tubules filling the lumen in some tubules, other show hyalinization and necrosis Fig(8). After 4 weeks of exposure to the same dose the lesions were more severe included patches of hemorrhage, necrosis and hyalinization of renal tubule with glomerular cell tuft degeneration Fig (9) and focal inflammatory infiltration mainly mononuclear cells Fig(10).

DISCUSSION

Lead is one of the most common toxic metals and the most common route of exposure is by ingestion of lead contaminated food (Durgut *et al.*, 2008). Once it is absorbed from gastrointestinal tract, lead bounds to erythrocytes and widely distributed initially to soft tissues such as liver, kidney, brain and spleen (Kosnett, 2004), for this reason we select the liver and kidney to describe the histological changes after lead exposure. The liver via the portal vein is the first organ exposed to enternally absorbed nutrients and other xenobiotics. The liver is composed of highly active metabolic tissue containing huge complement of detoxification machinery system (US-EPA, 1986).

The most common findings in liver were fatty degenerative changes, necrosis of the parenchyma of hepatic lobule and a loss of normal architecture of the hepatocytes.

These observations are in agreement with (Piasek *et al.,* 1989 ; Kojima *et al.*, 2005; Durgut *et al.*, 2008), these changes may be due to that lead acetate was shown to decrease cytochrome P_{450} content (US- EPA, 1986). Also lead induces mitogenic response in the rodent liver (Calabrese and Baldwin, 1992). Lead acetate was found to induce glutathione – S – transferase in rat liver (Suzuki *et al.*, 1996).

Lead exposure causes renal injury, the absorbed lead is primarily excreted by the kidney (Hurst and Martin, 2004). The main microscopic change were noticed in this study

are enlargement of epithelial cells lining renal tubules (proximal tubules), with hyalinization, necrosis of some tubules, these changes are in agreement with other reports (Greenberg, 1990; Vvskocil *et al.*, 1995; Loumbourdis, 2003 ; Durgut *et al.*, 2008).

These morphological changes could be due to rapid selective accumulation of lead in the cytosolic fraction of proximal tubular epithelium (Hascheck and Rousseaux, 1998), also lead damage membrane associated enzymes such as sodium – potassium pump which result in renal tubular injury (Plumlee, 2004).

Exposure to high lead levels can produce renal tubular damage, it has been suggested that renal damage could be related to cumulative lead dosage (Gidlow, 2004).

The glomerular tuft degeneration within 4 weeks treated group with 4 mg may be due to filtration of lead across the glomeruli (Sageb *et al.*, 2001). The inflammatory reaction including hemorrhage and inflammatory cell infiltration which we noticed in renal tissue and hepatic tissue of both treated groups, this in agreement with (Piasek *et al.*, 1989 ; Greenberge, 1990; Durgut *et al.*, 2008). Lead could impair the immune system (Zhao *et al.*, 2004).

In conclusion lead causes some deleterious effects observed in liver and kidneys in very high doses of lead administration with some similarities to chronic oral administration of low doses of lead.

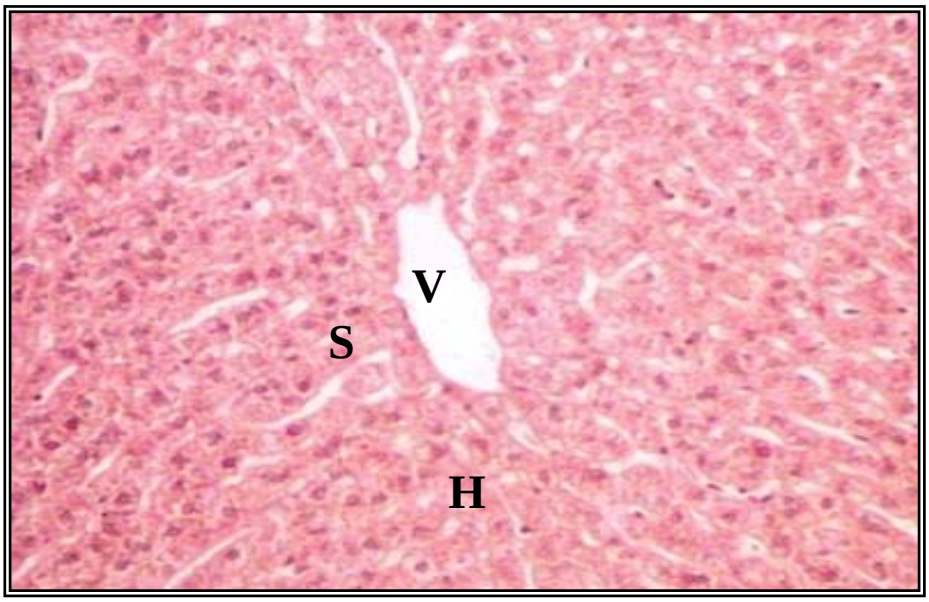

Fig.1: Light micrograph of mouse liver from control animal, showing normal structure, central vein (V) hepatocytes (H) and sinusoids (S) H&E. X 280.

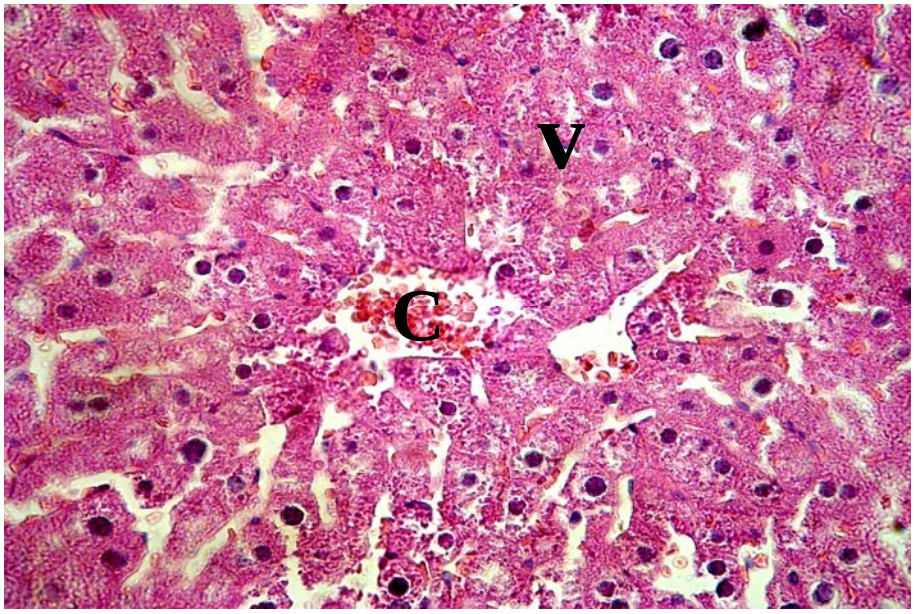

Fig. 2: Light micrograph of mouse liver treated with 2 mg pb/2weeks showing vacuolation of hepatocytes (V) with congestion of central vein (C). H&E. X 450.

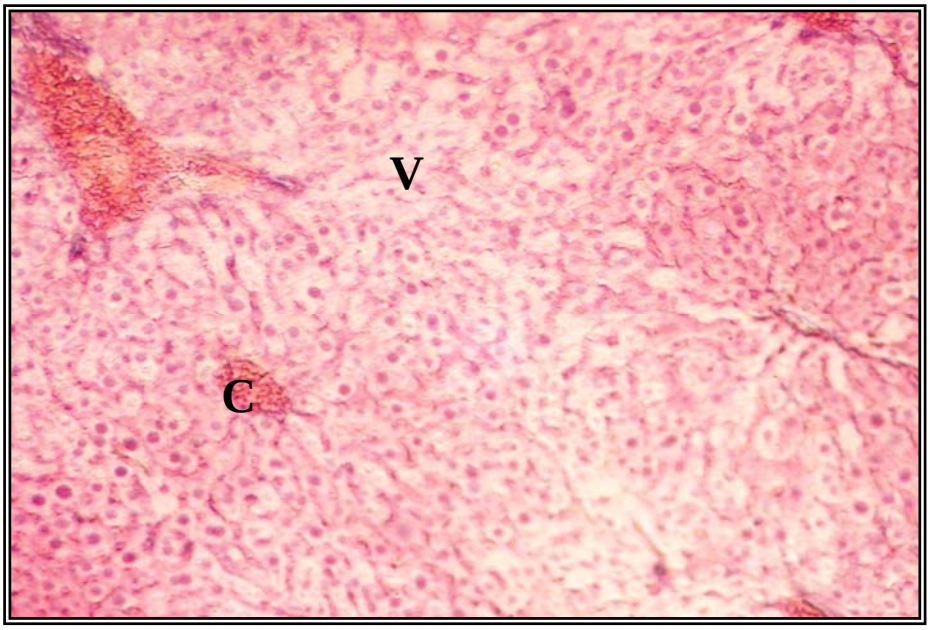

Fig .3: Light micrograph of mouse liver treated with 2mg pb/4weeks, showing vacuolar degeneration of hepatocytes (V) with congestion of central vein (C). H&E. X265.

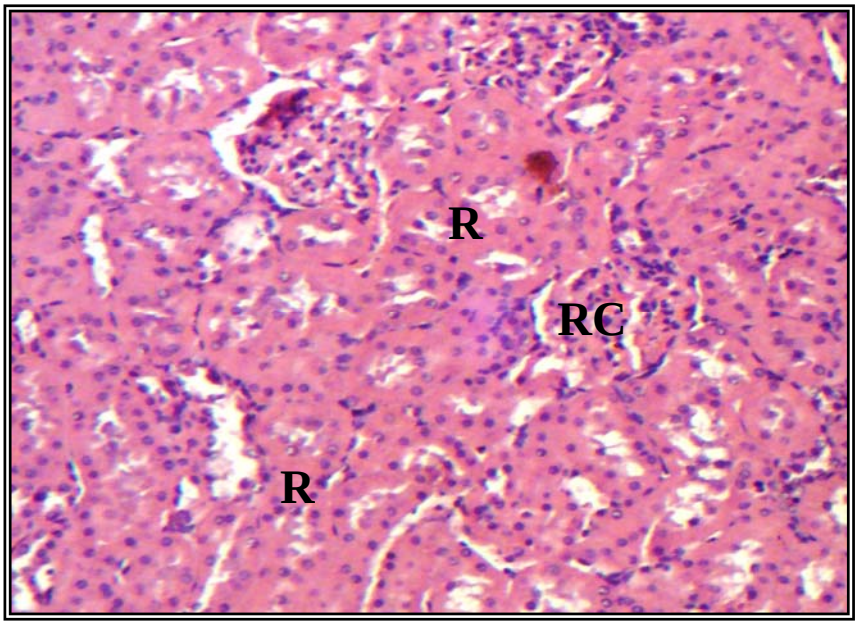

Fig. 4: Light micrograph of mouse kidney from control animal showing normal renal tubules (R) and renal corpuscles (RC) H&E. X 115

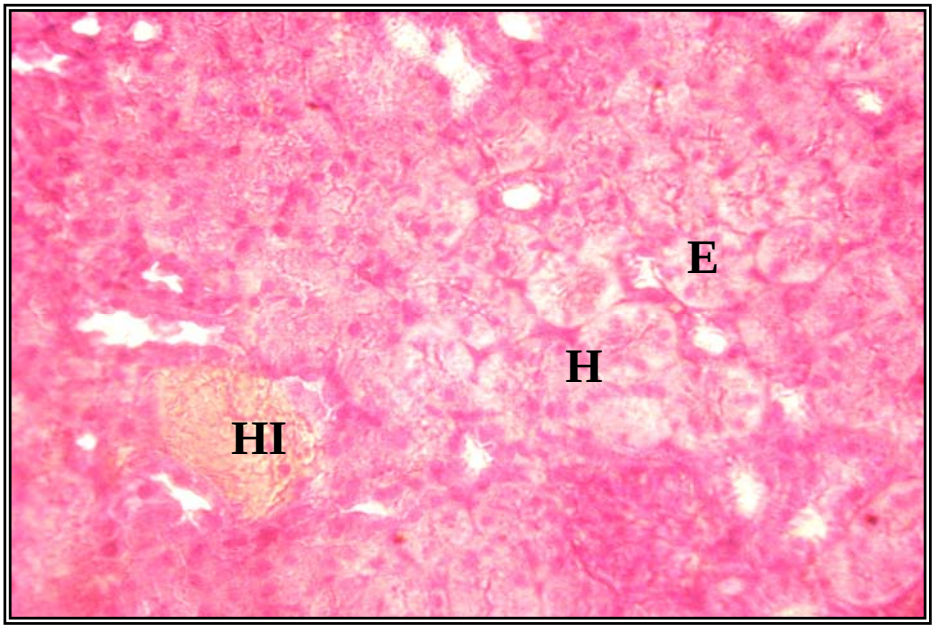

Fig. 5: Light micrograph of mouse kidney treated with 2mg pb/4weeks, showing enlargement of epithelial cells lining the renal tubules(E) hyalinization of some renal tubules (H) hemorrhage in the interstitial tissue (HI) H&E. X 90

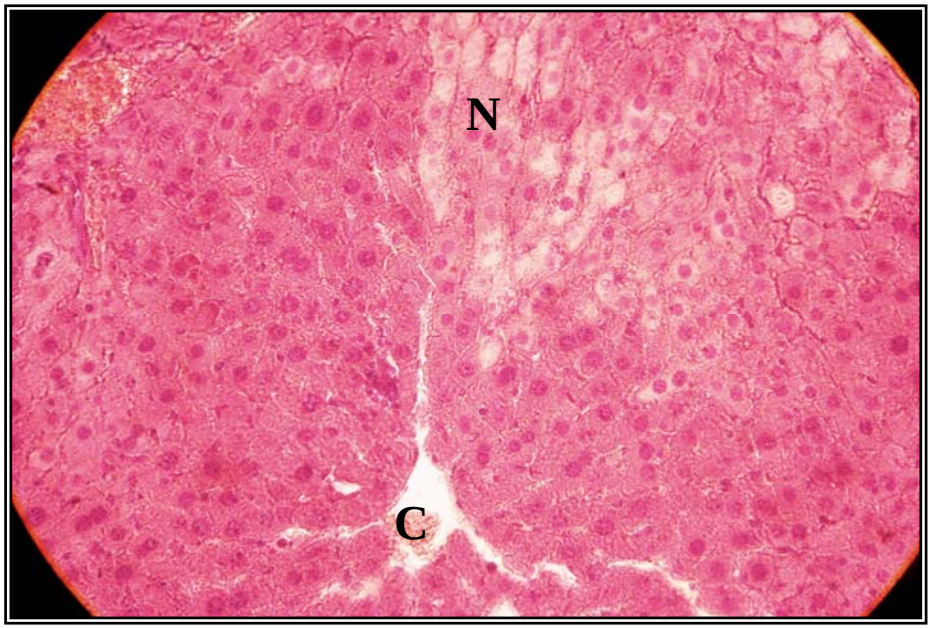

Fig 6: Light micrograph of mouse liver treated with 4mg pb/2weeks, showing necrosis of hepatic cells (N) congestion of central vein (C). H&E X325.

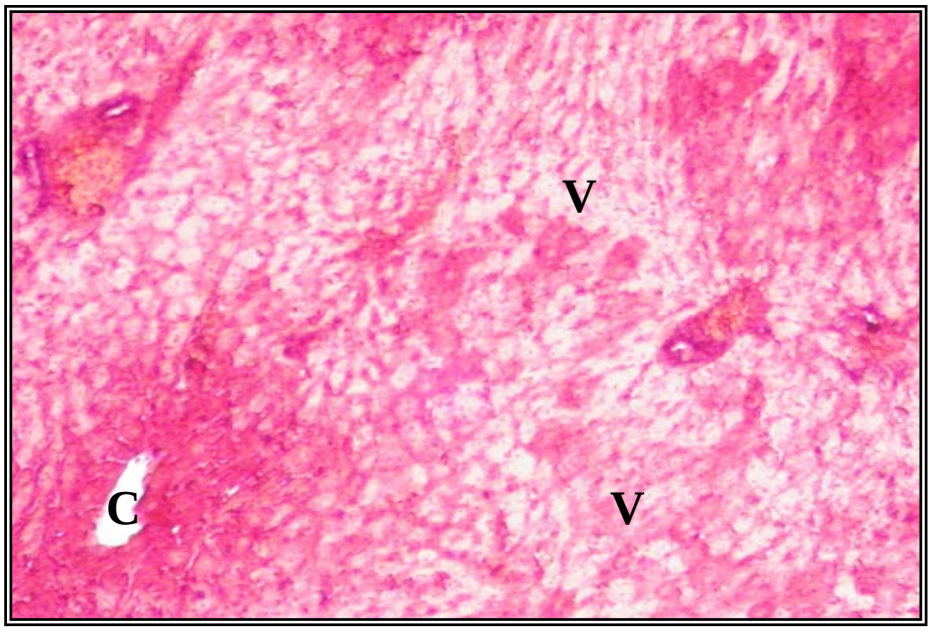

Fig .7: Light micrograph of mouse liver treated with 4mg pb/4weeks, showing more vacuolar degeneration of hepatic cells (V) and central vein(C)H&E X 265.

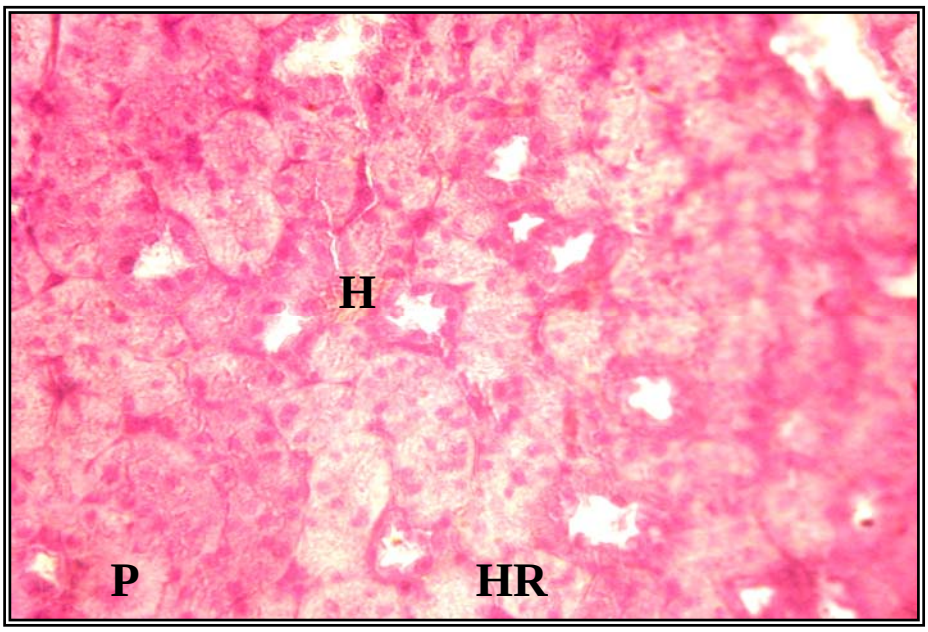

Fig. 8: Light micrograph of mouse kidney treated with 4mg pb/2weeks, showing hemorrhage in the interstitial tissue (H) proliferation of epithelial cells lining proximal tubules (P) hyalinization and necrosis of renal tubules(HR) H&E. X90.

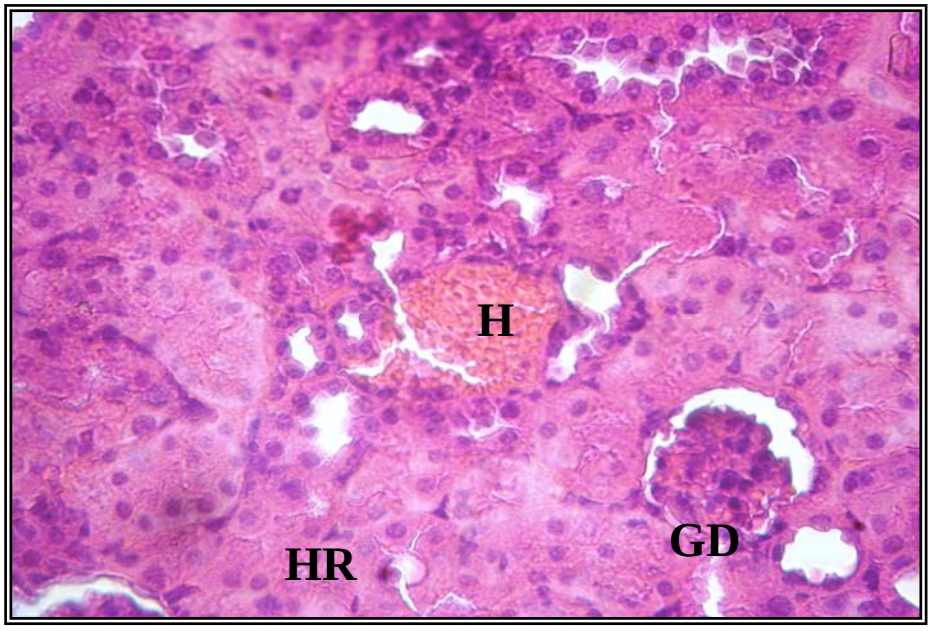

Fig.9: Light micrograph of mouse kidney treated with 4mg pb/4weeks showing hemorrhage in the interstitial tissue (H) hyalinization of renal tubules (HR) glomerular tuft degeneration (GD). H&E X130.

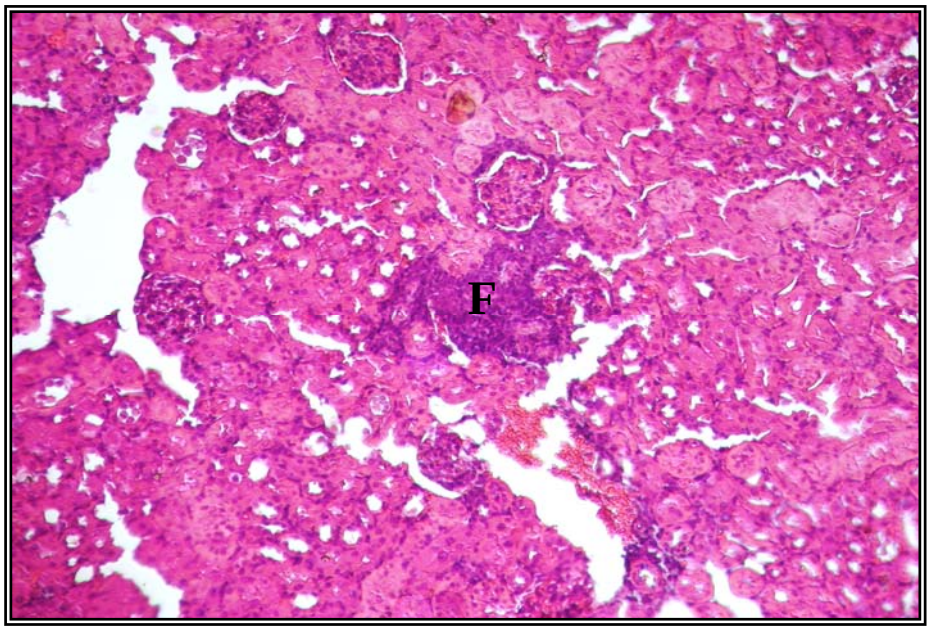

Fig.10: Light micrograph of mouse kidney treated with 4mg pb/4weeks, showing focal inflammatory cell infiltrated in renal tissue (F) H&E. X 75

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