Study the physiological and histological changes in mice liver that infected with Candida albicans and the role of camel's milk to treatment

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Abstract

The present study used 16 albino mice Mus musculus that divide randomly to four groups (each group consist 4 mice), the first group was control group administrated only normal diet and water, the second group injected intraperitoneally LD50 (5x105 CFU/ml), the third group injected with infectious dose of Candida albicans (8x104 CFU/ ml), the fourth group injected with infectious dose and treated with camel milk with dose (1 ml). The GOT and GPT levels were increased and showed high significant changes ($P < 0.01$) in groups that administrated with LD50% and infected dose compared with control group but its back to normal when treated with camel milk. The microscopic examination showed many changes in liver of groups that injected with lethal and infectious dose from C. albicans including degeneration, fibrosis, infiltration of lymphocytes, congestion and thickening wall of central veins. The liver tissue in group that injected with infectious dose and treated with camel milk showed enhancement in the arrangement of hepatocytes and the liver tissues return to the normal form and structure. It was concluded from this study that camel milk has amply good effect on liver enzymes and tissue.

Introduction

Candida albicans is an opportunistic yeast pathogen that is found in the gastrointestinal and genitourinary tracts also on the skin as a normal flora [1,2]. C. albicans is a polymorphic organism: it has the ability to undergo morphological changes between the yeast form (with rounded cells and daughter buds that physically separate from the mother cell), the pseudohyphal form (which consists of chains of cells with different degrees of elongation that show constrictions between adjacent cells), and the hyphal form (which consists of long tubes with parallel sides and no constrictions) [3].

Infections with C. albicans increased mostly in the patients with immune system disorders and weak (as a result of cancer chemotherapy) or when the competing flora are eliminated and after surgical operation and tissue transplantation, [4, 5, 6], C. albicans colonizes and invades host tissues. Candidiasis is a common infection in

human and animals [1,7]. This study aimed to identify effect of C. albicans in the liver of albino mice and to identify pathological findings associated with experimental infection of this animal with C. albicans infection and to show the role of camel milk to treated the liver lesions that caused by C. albicans.

Materials & methods

Animal model

Sixteen adult albino mice (Mus musculus), (wt 25-28 g) obtained from the Public company of medicines manufacture and requirements medicals - Samara, Iraq, and kept on standard pellet diet and water.

Candida albicans culture

Candida albicans obtained from Mousle University/ college of science/ department of biology, and to ensure its purification, the following diagnostic methods had done including, cultural characteristics, microbiological examination, Germ tube\ formation chlamyolospores formation test. API candida Biomerieuxs, France).

Collect of milk sample

Milk samples were collected from camels (from Al-Hadam region, in the Tikrit city), this camels was in third month from period of milk production. The age, color, type of food and period of milk production for camel was reported. Milk was collected from camels by hand milking as normally practiced by the farmers. The samples were collected in sterile screw bottles and kept in cool boxes until transported to the laboratory.

Experimental of induced infection in mice

After determining the lethal dose for 50% (5x105 CFU/ml) [8] from animals and infectious dose (8x104 CFU/ ml) [9]. C. albicans were taken and injected intraperitoneally. The animals divided as following (each group consist four mice): the first group, control group administrated only normal diet and water, the second group administrated lethal dose for 50% of animals, the third group administrated infected dose from fungus, and the fourth group administrated infected dose from fungus and treated with camel milk with dose (1 ml and with ad libitum). In the LD50 group tow mice were dead after 24 hours. Mice in the infected group left for three days to induced the disease and to present symptom and then anesthesia and dissected the animals, in the infected and treated group, the animals left for three days to induced the disease and to present symptom and then treated with camel milk until seven days and then anesthesia and dissected the animals, where liver pieces are taken and kept in 10% buffered formalin.

Histological study

Fresh pieces of liver from each mice was cut out rapidly, fixed in 10% formalin and then dehydrated with ascending grades of ethanol. Dehydration was then followed by

clearing then tissue samples in two changes of xylene before being impregnated with three changes of melted paraffin wax, embedded and blocked out. Tissue sections thickness (5 um) were stained with haematoxylin-eosin [10].

Microscopic study and microscopic photograph

The microscopic investigation of liver sections involved the descriptive histology. A light microscope (Motic microscope) was used to perform the microscopic investigations of this study. Microscopic photograph was made using (Optica\Italy) microscope supplied with a special camera prepared for this purpose.

Results

Biochemical tests

GOT tests

The results of the present study showed significant changes (P>0.01) in level of GOT between groups. As shown in chart (1), the group B that administrated by injected with LD50 showed significant change compared with control group A. Also, group C that administrated by injected with infected dose showed significant change compared with control group. But in the group D, (group that administrated with infected dose for seven days and treated with camel milk), showed non-significant change compared with control group.

GPT tests

The results of the present study showed significant changes (P>0.05) in level of GPT between groups. As shown in chart (2), the group B that administrated by injected with LD50 showed significant change compared with control group A. Also, group C that administrated by injected with infected dose showed significant change compared with control group. But in the group D , (group that administrated with infected dose for seven days and treated with camel milk), showed non-significant change compared with control group.

Histological examination

A. Control group

The microscope examination showed normal structure of liver and demonstrated normal central vein, normal arrangement of hepatocytes and sinusoids (Fig.1).

B. group injected with LD50

The histological examination showed thickening wall of central vein and congestion of blood vessels and degeneration of hepatocytes and infiltration of lymphocytes with present fibrocytes (Fig.2).

C. group injected with infected dose

The microscope examination showed thickening wall of central vein and congestion of blood vessels and degeneration and necrosis of most

hepatocytes and severing infiltration of lymphocytes (Fig.3).

D. group injected infected dose and treated with 1ml from milk

The microscope examination showed more recovery for hepatocytes normal centravein without any damage. Also, the kupffer cells and sinusoids appear normal (Fig. 4).

Discussion

The physiological changes showed increased in the levels of GOT and GPT levels in the groups that injected with Candida albicans. These lesions come in accordance with previous search [11]. In this study, camel milk after administrated to the infected group lead to recovery the levels of GOT and GPT and back to the normal ranges that is in agreement with the results of previous search [12]. Histopathological changes in the liver that caused by Candida albicans showed degenerative changes, necrosis. These lesions come in accordance with previous researches [13, 14]. Camel milk act as antimicrobial, where in study about Camel milk effect to inhibition Streptococcus spp., Staphylococcus spp. found that the Camel milk has strong capacity to inhibit the growth of these bacteria [15], and There are many authors' reports that the camel milk has contains Proteins [16], Caseins [17], Fats [18], Lactose [15, 19], Mineral contents [15, 20], vitamins (D, E, A, C) [17], Immunoglobulins [21] and Lactoferrin [22], so suggested that the camel milk has antioxidant effects milk play important role in regeneration and repair the liver tissues. In study designed to show the role of camel milk to repair the liver of rats after administrated Carbon Tetrachloride, found the liver of carbon tetra chloride-intoxicated rats and treated with camel milk showed clear liver recovery characterized by a complete repaired of hepatocytes and the other liver tissues in most cases, so suggested camel milk may be used to protect against toxic effects of CCl4 and other chemical agents in liver [23]. That may be explains the role of camel milk against the toxicity of C. albicans. on the other hand, camel's milk has antioxidant effects as well as a strong capacity to trap free radicals. Also, camel's milk play important role in regeneration and repair the hepatocytes [24].

Reference

1. Berman, J. & Sudbery, P.E. . Candida albicans: a molecular revolution built on lessons from budding yeast. Natu. Rew., 3:918-930 , (2002) .
2. Yuthika, H. Samaranayake, P, and samaranayake, L. Experimental oral candidiasis in animal models . Clin. Microbial – Review, 14(2): 398 – 429 , (2001).
3. Sudbery P., Gow, N. and Berman, J. Trends Microbiol. 12:317-324.
4. Ruhnke, M. (2002) skin and mucous membrane infection In. R.A. Calderone (ed) Candida and

candidiasis . American society for microbiology,Washington D.C., P 307- 325 , (2004).

5. Ryan, K. J. and Ray, C. G. Sherries medical microbiology 4th ed . Chapter 48, Candida albicans,Pp. 661-663 , (2004) .
6. Vazques, J.A. and Sobel,J.D. Mucosal Candidiasis. Infect. Dis. Clin. N.Am, 16:793-820, (2000).
7. Miller, L. G., Hajjeh, R. A. & Edwards, J. E. Estimating the cost of nosocomial candidemia in the United States. Clin. Infect. Dis. 32, 1110 , (2001).
8. Reed. L. J. and muench. H. Asimple method of estimating fifty percent and end points Am. J. Hygiene, 24 , 493 – 499 , (1938) .
9. Berényi, R., Kovács, R., Domán, M., Gesztelyi, R., Kardos, G., Juhász, B., Perlin, D. and Majoros, L. J. Efficacy of single large doses of caspofungin in a neutropenic murine model against the "psilosis" group. New Microb., 37:355-362 , (2014).
10. Bancroft, J.D. and Stevens, A. Theory and practice of histological techniques. Edinburgh: Churchill Livingston. Pp: 482-502 , (1987).
11. Duro, D., Babayi H., Odeh O. E., Tijani A.Y. and Salawu O. A. Investigating the toxicity and antimicrobial activity of garcinia kola extracts. J. Res. Art. 4 (4):57-67.
12. Darwish, H. A., Naglaa R. A. and Amina M. Camel's milk alleviates alcohol-induced liver injury in rats. J. Food and Chemical Toxicology 50 :1377–1383.
13. Al- Jeboori, K.H. & Al- Harreery, S. A. H. Study on mycotic dissemination and pathology findings associated with experimental CANDIDA ALBICANS infection in mice. G.J.B.B., 2 (4): 534-538 , (2013).
14. Mohamed, B. J., AL- Hussain, R. A. and AL. Thwani, A.N. Study the inh inhibitory effect of Lactobacillus acidophilus isolated from yoghurt as probiotics on Candida albicans growth in vitro and in vivo. Iraqi J. Biotech. 9(2): 167-179 , (2010).
15. Hussein, H.J. Study to evaluation the Camel milk effect to inhibition Streptococcus spp., Staphylococcus spp. in patients with gingivitis. kufa med. J. 2(1): 20-27, (2011).
16. Konuspayeva, G., B. Faye and G. . Loiseau. The composition of camel milk: a meta-analysis of the literature data. J. Food Compos. Anal. 22: 95-101 , (2009).
17. Khaskheli, M., M. A. Arain, S. Chaudhry, A. H. Soomro and T. A. Qureshi. Physico-chemical quality of camel milk. J. Agri. Soci. Sci. 2: 164-166 , (2005).
18. Haddadin, M. S. Y., S. I. Gammoh and R. K. Robinson. Seasonal variations in the chemical composition of camel milk in Jordan. J. Dairy Res. 75: 8-12 , (2008).

19. Konuspayeva, G., E. Lemarie, B. Faye, G. Loiseau and D. Montet. Fatty acid and cholesterol composition of camel's (Camelus bactrianus, Camelus dromedaries and hybrids) milk in Kazakhstan. Dairy Sci. Technol. 88: 327-340 , (2008).
20. Elamin, F. M. and C. J. Wilcox. Milk composition of Majaheim camels. J. Dairy Sci. 75: 3155-3157, (1992).
21. El-Agamy, E. I. and M. Nawar. Nutritive and immunological values of camel milk: A comparative study with milk of other species. In: Proc. 2nd International Camelid Conference: Agroecons. Camelid Farm, Almaty, Kazakhstan. 8-12 Sept. (2000).
22. El-Agamy, E. I. Camel colostrum. II. Antimicrobial factors. Proceedings of the Workshop on Camels and Dromedaries as Dairy Animal, Nouakshott, Mauritania. 24-26. pp: 177-180 , (1994).
23. Althnaian, T. Protective effect of camel milk against carbon tetrachloride hepatotoxicity in rats. Global Veterinaria 9 (5): 564-570 , (2012).
24. Salwa, M. Q. & Lina, A.F. Antigenotoxic and anticytotoxic effect of camel milk in mice treated with cisplatin. J. Biolog.Scien. 17:159–166 , (2010).

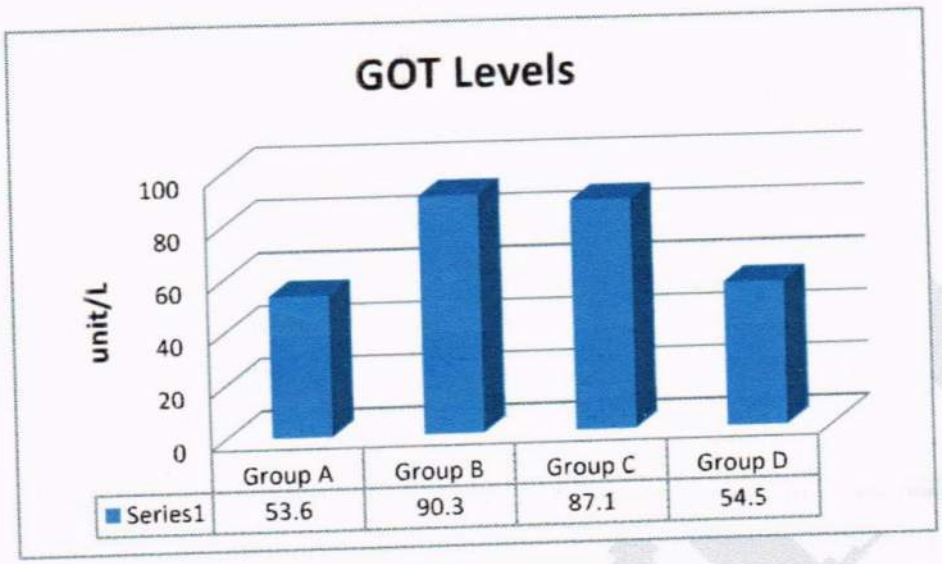

Chart (1)

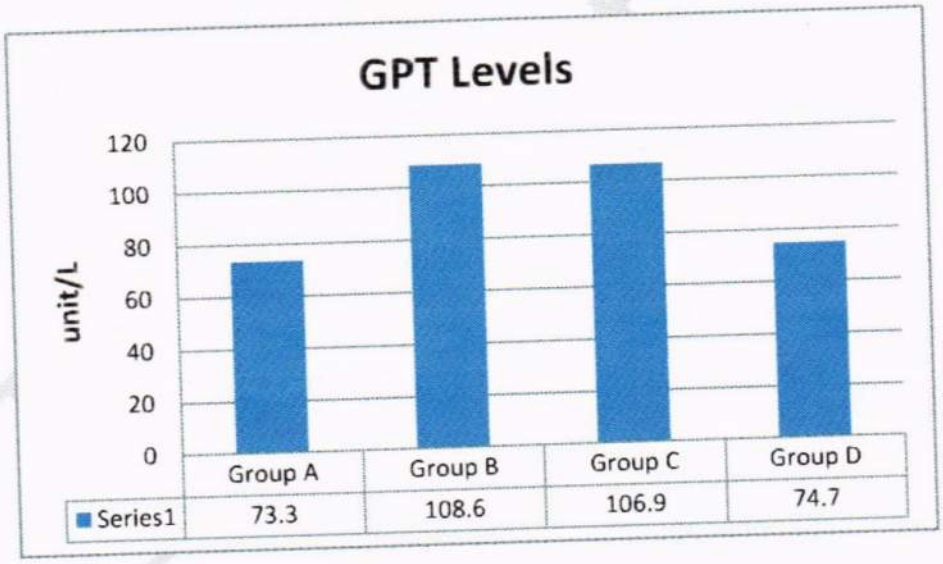

Chart (2)

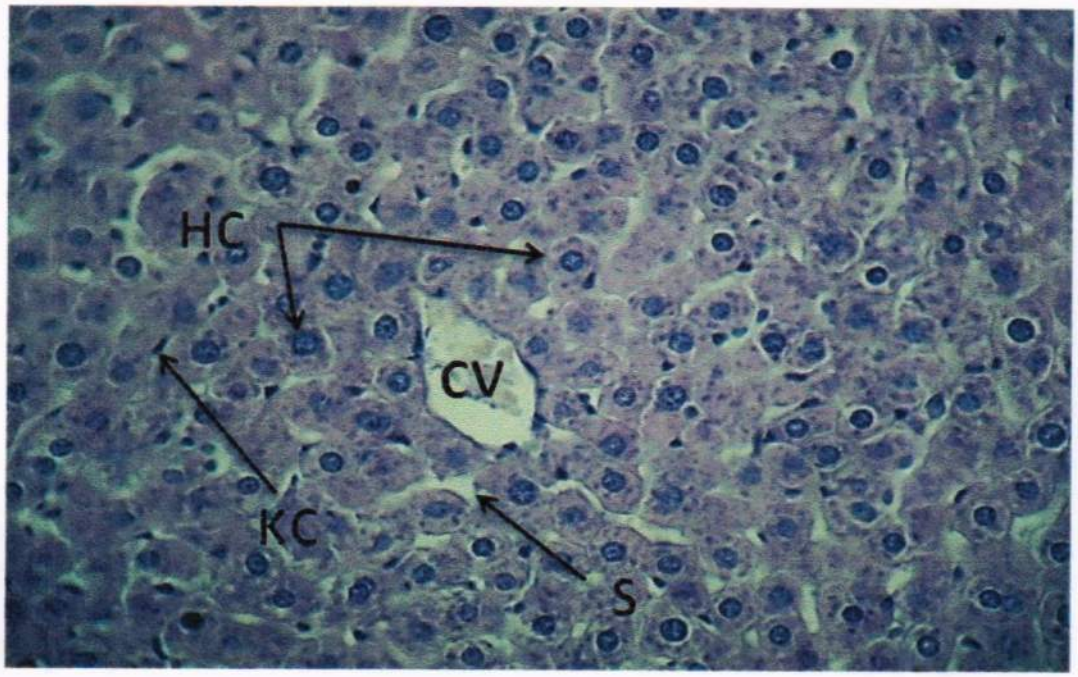

Figure (1): Liver of control group showed normal central vein (CV), normal hepatocytes (HC), normal sinusoids(S) and normal kupffur cells (KC) H&E 400X.

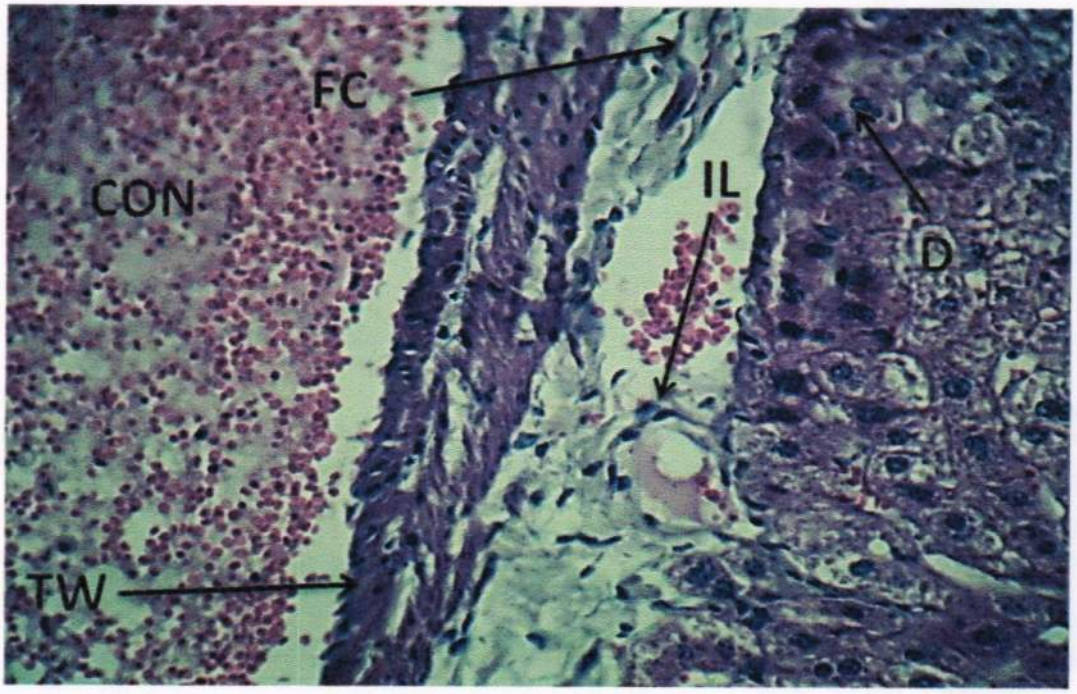

Figure (2): Liver of LD50% dose group showed thickening wall (TW) of central vein, congestion (CON), degeneration (D) hepatocytes and lymphocytes infiltration (IL) with fibrocytes (FC) H&E 400X.

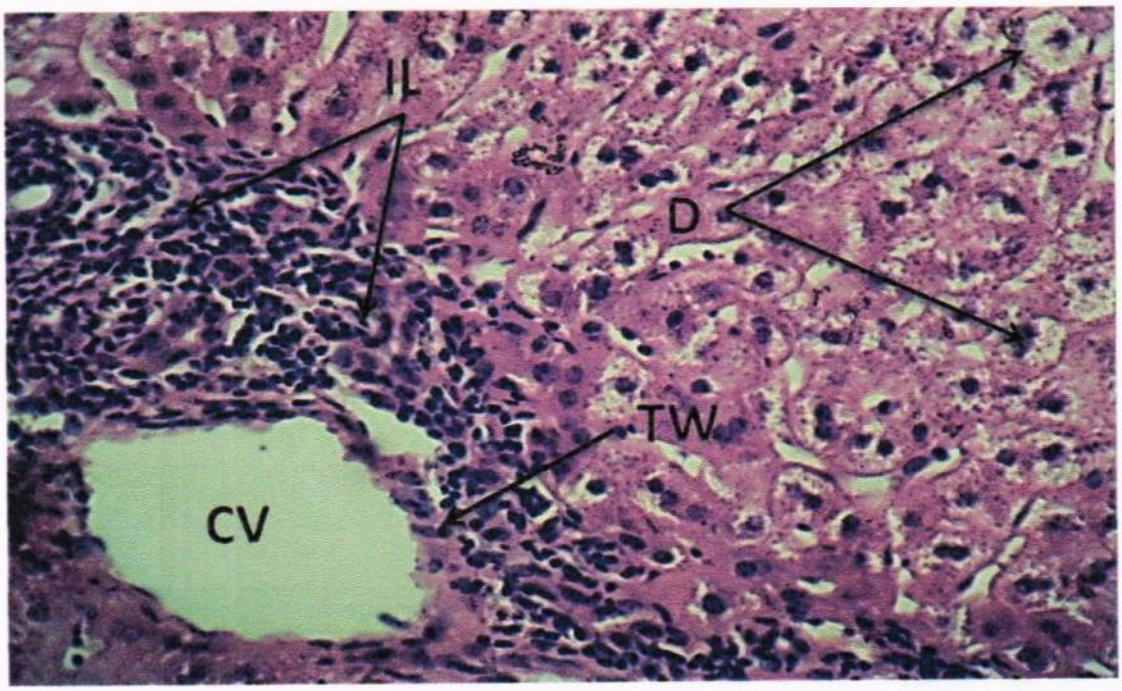

Figure (3): Liver of ethanol for twenty one days showed thickening wall (TW) of central vein (CV), degeneration (D) of hepatocytes and lymphocytes infiltration (IL) H&E 400X.

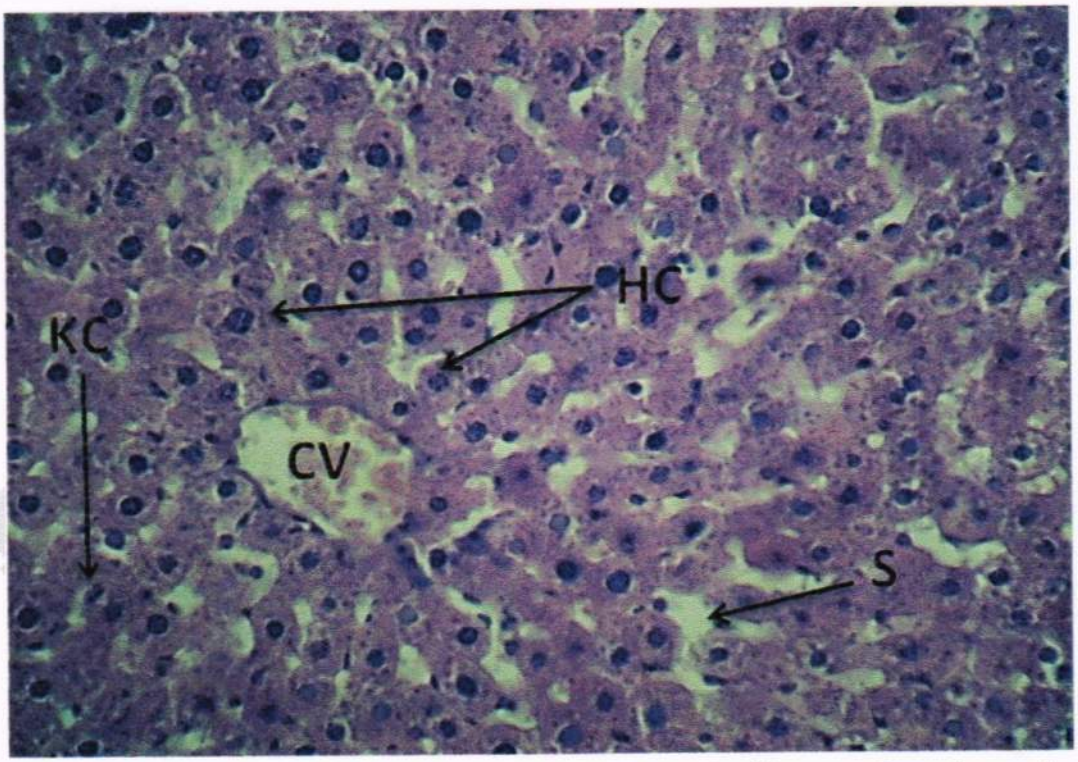

Figure (4): Liver of infected dose and treated with camel milk group showed normal central vein (CV), normal hepatocytes (HC), normal sinusoids(S) and normal Kupffer cells (KC) H&E 400X.