Assiut Vet. Med. J. Vol. 49 No. 98, July 2003

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HEMATOLOGICAL AND BIOCHEMICAL CHANGES ASSOCIATING CHRONIC LOCALIZED CHOLANGITIS AND SAWDUST LIVER IN CATTLE

(With 5 Tables and 3 Figures)

By

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(Received at 24/6/2003)

التغيرات الدموية والبيوكيميائية المصاحبة لإلتهاب القنوات المرارية الموضعي المزمن وكذلك الالتهاب الكبدى الدخنى شبيه نشارة الخشب فى الماشية

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استهدفت هذه الدراسة تقييم بعض التغيرات الدموية والبيوكيميائية المصاحبة لالتهاب القنوات المرارية الموضعي المزمن وكذلك حالات الالتهاب الكبدى الدخنى شبيه نشارة الخشب في الماشية. شملت الدراسة مجموعتين من عجول التسمين كان عددها ٦٩ حيوان ، تراوحت أعمارها بين ٢,٥ ، ٣ سنوات. شملت المجموعة الأولى ٣٥ من عجول التسمين البلدى المصرية والتى كانت معدة للذبح في مجزر أسـيوط بجمهورية مصر العربية. تم فحص الحيوانات قبل الذبح واخذت عينات دم منها لدراسة صورة الدم لها ، أما بعد الذبح فقد تم فحص الذبائح واخذ عينات من نسيج الكبد لعمل الفحوصات الهستوباثولوجية. ٢٠ من حيوانات هذه المجموعه كانوا بحالة صحية ظاهرية جيدة وبفحص هذه الحيوانات بعد ذبحها افاد الفحص عدم وجود اى تغيرات مرضيه باى من اعضائها ، واستخدموا كمجموعة قياسيه ضابطه. باقي حيوانات هذه المجموعه (١٥) بعضها كان يعانى من الضعف العام وشحوب الأغشية المخاطية والبعض الاخر شوهدت تعانى من درجات متباينة من الإسهال الذى تراوح قوامة بين الطرى والمائى فى بعض منها. فحصت هذه الحيوانات بعد ذبحها وافاد الفحص وجـود التهابات موضعية مزمنة في القنوات المرارية لكبدها. تكونت المجموعة الثانية من ٣٤ حيوان من ذكور الهولشتين اليابانى البالغة والتى كانت معدة للذبح في مجزر أساهـيكاوا بجزيرة هوكايدوا باليابان. كانت حيوانات هذه المجموعه سليمة ظاهريا، وبفحص هذه الحيوانات بعد ذبحها افاد الفحص عدم وجود اى تغيرات مرضيه باى عضو من اعضاء ٢٥حيوان منها، تم استخدامهم كمجموعة قياسيه ضابطه. اما باقي حيوانات هذه

المجموعه (٩) فقد وجد بكبدها تتكرز في صورة حبيبات تشبه نشارة الخشب منتشرة في جميع أنسجة الكبد. ، تم أخذ عينات دم على مانع للتجلط و أخرى بدون مانع للتجلط للحصول على مصل للدم وكذلك عينات من نسيج الكبد من كل حيوان فى هذه المجموعة. أستخدمت عينات مصل الدم فى تقدير مستويات نشاط أنزيمات الأسبرتات أمينوترانزفيريز والجاما جلوتميل ترانسفيريز والأدينوزين ديأمينيز والجلوتامات ديهيدروجينيز والبروتينات الكلية والألبيومين والصفراء، واستخدمت عينات الدم الكلى وعينات نسيج الكبد لتعيين مستوى أنزيمى الجلوكوز ٦ فوسفات ديهيدروجنيز والجلوتثيون بيروكسيداز المتواجدين في كل من خلايا الدم الحمراء وانسجة الكبد لهذه الحيوانات. بعد اجراء الصفه التشريحيه للذبائح وفحص عينات الكبد مجهريا (هستوباثولوجى) أوضحت النتائج أن السبب الرئيسى فى حدوث التهاب القنوات المراريه الموضعى المزمن فى عجول التسمين البلدى المصرية فى هذه الدراسه يرجع الى إصابتها بالديدان الكبدية. هذا وقد أظهرت التحاليل الدموية لعينات الدم الكلى لهذة الحيوانات عدم تغيير صورة خلايا الدم الحمراء بها بينما حدثت زيادة معنوية فى العدد الكلى لخلايا الدم البيضاء مصحوبة بزيادة في النسبة المئوية للخلايا المتعادلة والخلايا الحامضية. أظهرت التحاليل البيوكيميائية وجود زيادة معنوية في أنشطة أنزيمات الأدينوزين دىأمينيز والجاما جلوتميل ترانسفيريز والجلوتامات ديهيدروجينيز والأسبرتيت أمينوترانسفيريز والصفراء في مصل دم الحيوانات التي تعانى من الالتهاب الكبدى الدخنى شبيه نشارة الخشب. لوحظ أيضا وجود زيادة معنوية في مستوي أنزيم الجلوكوز ٦ فوسفات ديهيدروجينيز في خلايا كبد هذه الحيوانات وتعزى هذه الزيادة إلى إرتفاع مستوى الشوارد المؤكسدة فى الجسم أثناء هذا المرض مما أدى إلي زيادة نشاط هذا الانزيم كوسيله من الجسم لانتاج المزيد من مضادات الاكسده للقضاء على هذه الشوارد. علي الجانب الأخر لم يحدث أي تغيير معنوي في مستوي أنزيم الجلوتاثيون بيروكسيداز سواء في خلايا الكبد أو خلايا الدم. من هذه الدراسة يمكن أن نستخلص أن زيادة العدد الكلى لخلايا الدم البيضاء مع زيادة نسبة الخلايا المتعادلة والحامضية قد يعد مؤشرا جيدا لاصابه الحيوانات بالتهاب القنوات المرارية الموضعى المزمن وأن قياس مستوى نشاط إنزيمات الأسبرتيت أمينوترانسفيريز والجاما جلوتميل ترانسفيريز والأدينوزين ديأمينيز والجلوتامات ديهيدروجينيز والبروتين الكلى والصفراء بالاضافه الى الصورة الهستوباثولوجيه لعينات بذل الكبد يساعد في تشخيص الالتهاب الكبدى الدخنى شبيه نشارة الخشب فى الماشية كما يعد قياس محتوى خلايا الكبد من انزيم الجلوكوز ٦ فوسفات ديهيدروجينيز من اهم ما يساعد فى تشخيص هذا المرض.

SUMMARY

This study aimed to evaluate some hematological and biochemical changes associating chronic localized cholangitis and sawdust liver in cattle. Two groups of fattening steers with a total number of 69 animals, 2.5 - 3 years old were investigated in this study. The first group included 35 fattening steers (Egyptian native breed of cattle), were subjected to slaughtering in Assiut`s slaughterhouse, Assiut Governorate, Assiut, Egypt. 20 steers of them were clinically healthy, of good condition and the post mortem examination revealed freeing of any organ of them from

any pathological lesions and were kept as control. The rest (15 steers) were suffered general weakness, paleness of the mucous membranes and variable degrees of diarrhea (soft to watery). After slaughtering, post mortem examination revealed, thickining of the bile ducts of their livers with presence of adult liver flukes (*Fasciola* spp.) in their lumen (chronic localized cholangitis). Specimens of these affected livers were taken and submitted for histopathological examination. The second group composed of 34 clinically healthy fattening steers (Japaniese Holstein breed of cattle) subjected to slaughtering in Asahikawa slaughterhous, Asahikawa, Hokkaido – Japan. Post mortem examination revealed the freeing of any organ of 25 steers of these animals from any pathological lesions and they were kept as control. The remaining (9 steers) were found suffering from sawdust liver. The affected liver contains several minute yellowish foci or necrosis, as if the same numbers of sawdust granules scattered over it. Whole blood and blood serum samples with liver tissue specimens were collected from each of these animals. The blood serum samples were used for the determination of the activities of aspartate amino transferase (AST), gamma glutamyl transferase (γ-GT), adenosine deaminase (ADA), glutamate dehydrogenase (GLDH), total protein, albumin and total bilirubin. The whole blood samples and part of the liver tissue specimens were used for the determination of the activities of erythrocytic and hepatic glucose 6 phosphate dehydrogenase (G6PD) and glutathione peroxidase (GSH-Px). The other part of the liver tissue specimens was subjected for the histopathological investigations. Chronic localized cholangitis in fattening steers in this study appears as a result of *Fasciola* spp. infestation. The findings of histopathological examination of these cases revealed incresed amounts of portal connective tissue and fibrosed thickned bile ducts. Hematological investigations revealed insignificant change in red blood cell picture, however, there was a significant increase in the total leucocytic count with neutrophilia and eosinophilia. On the other hand, cases with sawdust liver showed significant increase in serum activities of ADA, γ-GT, GLDH, AST, bilirubin and hepatic G6PD.The increased level of hepatic G6PD activity may be attributed to the increased free radicals levels in cases of sawdust liver. Insignificant changes in the hepatic and erythrocytic GSH-Px activities were found in such cases. It could be concluded that; Leucocytosis with eosinophilia and neutrophilia in addition to specific clinical manifestation are good indicators for chronic localized cholangitis. Serum total protein, albumin and globulin levels are insensitive indicators for sawdust liver in cattle.

Measuring serum activities of AST, γ-GT, ADA, GLDH and serum total bilirubin levels are helpful in diagnosis of the sawdust liver. With the aid of liver biopsy, measuring of hepatic G6PD activity may be helpful in diagnosis of sawdust liver.

Key words: *Sawdust liver, Chronic localized cholangitis, liver affections, cattle.*

INTRODUCTION

Liver plays a central role in hemopieosis and synthesis of coagulation proteins. Liver diseases are associated with a broad range of hematological abnormalities. Anemia arises through multiple mechanisms, making hemoglobin metabolism is disturbed. Liver diseases cause alterations in red cell lipid metabolism, reduction in platelets number, defects in immune mechanisms and splenomegaly (Mehta and McIntyre, 1998). The hematological aspects in cases of chronic localized cholangitis associated with naturally occurring *Fasciola gigantica* infestation in cattle were investigated by Haroun and Hussein (1975) where reduced erythrocyte counts, decreased hematocrit values, increased mean corpuscular volumes, eosinophilia and decreased neutrophils, lymphocytes and monocytes counts were recorded. The only significant hematological change in cases of chronic localized cholangitis in cattle infested with *Fasciola magna* was eosinophilia present from 2^{nd} - 26^{th} week post-infection (Conboy and Stromberg, 1991).

Sawdust liver occurs frequently in fattened growing cattle. It may appears in clinically perfect healthy animals. The affected liver contains several minute yellowish foci or necrosis, as if the same numbers of sawdust granules scattered over it. They consist of collection of epithelial cells in a state of coagulative necrosis or in the process of disappearing, mingled with/or surrounded by sprinkling of neutrophils and lymphocytes (Smith *et al.*, 1972).

Glucose 6-phosphate dehydrogenase (G6PD) is the first enzyme in the pentose phosphate pathway (Deutsch, 1983). It catalyzes the oxidation of glucose-6-phosphate to 6-phosphogluconate, while concomitantly reducing $NADP^+$ to NADPH (Swenson, 1984). In cells experiencing oxidative stress, a continual supply of reducing equivalents in the form of NADPH is required (Martini and Ursini, 1996). G6PD is responsible for maintaining adequate level of NADPH inside the cell (Salvemini *et al.*, 1999).

Glutathione protect cells against reactive oxygen species by a variety of mechanisms, one mechanism is the glutathione peroxidase (GSH-Px) reaction in which hydrogen peroxide is reduced to water with the concomitant oxidation of glutathione (GSH) to the disulfide (GSSG). Since the amount of glutathione present in the cell is finite, GSSG must be recycled to the reduced GSH state to maintain protection against hydrogen peroxide. This is accomplished by glutathione reductase (GR), which utilizes electrons from the oxidation of NADPH to convert GSSG to GSH. GSH-Px and G6PD are the key enzymes involved in the protection of cells from oxidative damage (Khan *et al.*, 1987).

This study aimed to; investigate some hematological and biochemical changes associating chronic localized cholangitis and sawdust liver in fattening steers. It is also aimed to evaluate the state of the oxidative stress associating the affection of fattening steers with chronic localized cholangitis and sawdust liver

MATERIALS and METHODS

I- Animals: This study was divided into two parts according to the area of study;

The first part was done in The Arab Republic of Egypt. A total number of 35 fattening steers (Egyptian native breed of cattle) were subjected to slaughtering in Assiut`s slaughterhouse, Assiut Governorate, Egypt. The animals were examined clinically before slaughtering according to Rosenberger (1990). Blood samples were collected in clean dry centrifuge tubes containing disodium salts of EDTA for complete blood picture. Post-mortem examination was carried out and liver tissues specimens were collected and kept in 10 % neutral buffered formalin for histopathological studies.

The second part was done in Japan, with a total number of 34 clinically healthy fattening steers (Japanese Holstein breed of cattle) were subjected to slaughtering in Asahikawa slaughterhouse, Asahikawa, Hokkaido – Japan. Case history of each animal before slaughtering was studied and then blood samples (whole blood and blood serum samples) were collected in clean and dry tubes. After slaughtering each animal was examined for presence of any pathological lesions. Of the total number, 25 animals were clinically healthy, free from any post-mortem pathological lesions and were kept as control, the remaining (9) were found suffering from sawdust

liver. Liver samples were collected from all animals under study. Blood and liver tissues samples from control animals and those with post-mortem pathological lesions in the liver were subjected to laboratory investigation.

II- Samples and adopted methods:

A) Whole blood with anticoagulant (disodium salts of EDTA):

- About 3 ml whole blood samples were collected from each animal of the first group in dry clean tubes and were used for complete blood picture according to Coles (1986).
- About 6 ml whole blood samples were collected from each animal of the second group and kept directly in cold container containing some ice pieces and were used for making the hemolysate according to the following steps:

1- Blood samples were centrifuged at 3500 rpm for 15 min at 4°C.
2- Directly after centrifugation the plasma and buffy coat were drawn off.
3- After that, the packed cells were washed one with ten volumes of cold saline.
4- The packed erythrocytes were divided into two parts in sterile ependorff tubes.
   - One part for determination of erythrocytic GSH-Px activity; the RBCs were hemolysed by adding 4 volumes of cold deionized water.
   - The second part was used for determination of G6PD activity of erythrocytes (RBCs were hemolysed by mixing 0.05 ml of the washed cell suspension with 0.5 ml of lysing solution (0.02 % digitonin containing NADP, 15 µmol/l; dissolve 16 mg digitonin in 80 ml deionized water, filter (Whatman # 1), then add 1 mg NADP) and then the hemolysates were stored at –70 °C till subjected to analysis.

Intracellular erythrocytic G6PD (U/g Hb.) activity was measured spectrophotometerically according to the method described by Deutsch (1983).

Intracellular erythrocytic GSH-Px (U/g protein) activity was determined by using test kits supplied by Sigma - Aldrich (Paglia and Valentine, 1967).

B) Whole blood samples without anticoagulants: were collected from each animal of the second gorup for obtaining blood serum samples for the determination of blood serum levels of total protein, albumin,

total bilirubin levels and serum activities of aspartate amino transferase (AST), adenosine deaminase (ADA), glutamate dehydrogenase (GLDH) and γ- glutamyl transferase (γ-GT) by using test kits supplied by Boehringer Manheim GmBH diagnostica.

C) **Liver tissues specimens:** Liver samples were collected from each animal under study and divided into two parts; one part (1 cm x 1.5 cm x 5 mm thickness) were preserved in 10 % neutral buffered formalin till subjected to histopathological examination, the other part were preserved directly after collection in ice and was prepared as follow: homogenize the sample in 4 volumes (per weight tissue) of cold buffer e.g. 50 mM TRIS-HCL, pH 7.5. Centrifuge at 3500 rpm for 30 minutes at 2-8°C, The supernatant fluid containing the enzyme was collected and stored in ice if they assayed at the same day or frozen at –70 °C if they analyzed later. These samples were used for measuring hepatic activities of G6PD and GSH-Px.

III- **Statistical analysis;** of the obtained data was done by window program (Statlets 2000, version 2.01, by StatPoint LLC. Englewood Cliffs, N. J.).

RESULTS

Clinical findings:

1- **Clinical examination of the first group** revealed that 20 animals were clinically healthy, of good condition, showed no abnormal clinical signs (control). The rest (15 steers) were suffered general weakness, paleness of the mucous memmbranes and variable degrees of diarrhea (soft to watery).

2- **Clinical examination of the The second group** revealed that there were no abnormal clinical signs observed on animals suffering from sawdust liver.

B. Gross and histopathological findings:

1. Chronic localized cholangitis:

Affected areas of the liver showed; thickened bile ducts with presence/or absence of mature forms of *Fasciola species*. However, the remaining parts of the liver tissues were healthy and free from any pathological lesions. The thickened bile ducts contained brownish fluid, some bile ducts were mineralized (Fig. 1). The histopathological examination showed that the increased portal C.T. was dense,

collagenous and contained few chronic inflammatory cells, bile duct proliferation was also detected (Fig. 2).

2. Sawdust liver:

There were several minute yellowish foci or necrosis, as if the same numbers of sawdust granules scattered over the liver and characterized histologically by presence of multifocal area of necrosis with neutrophilic infiltration (Fig. 3).

C. Hematological findings:

Statistical analysis of the recorded results showed non significant change ($p>0.05$) in total red blood cells count, Hb. concentration and in PCV, MCH and MCHC. However, there were significant increases ($p<0.01$) in total leucocytic count with neutrophilia and eosinophilia in cases of chronic localized cholangitis (Table 1 and 2).

D. Biochemical findings:

The results showed highly significant increase ($p<0.01$) in serum activities of ADA, γ-GT, GLDH, AST and in serum bilirubin levels. There was a significant increase ($p<0.01$) in hepatic G6PD activity and insignificant changes in hepatic and erythrocytic GSH-Px activities. Also, there were insignificant changes in serum total protein, albumin and globulin levels in cases with sawdust liver (Tables 3, 4 and 5).

DISCUSSION

Clinical findings:

In the present study, animals with chronic localized cholangitis were suffered general weakness, paleness of the mucous memmbranes, variable degrees of diarrhea (soft to watery) and moderate body condition score. Affected areas of the liver showed; thickened bile ducts with presence of mature forms of *Fasciola species*. Several authors recorded simillar observations (Haroun and hussein, 1975; Pearson, 1990 and Behm and Sangster, 1998). Infestation with *Fasciola* lead to acute or chronic hepatic insufficiency and infectious necrotic hepatitis may develop associated with the clasical signs of fascioliasis which are ; loss of weight, pallor of the mucosa and diarrhea (Radostitis *et al.*,1994)

Clinical examination of the second group revealed that there were no abnormal clinical signs observed on animals suffering from sawdust liver. Smith *et al.* (1972) recorded simillar observations.

Gross and histopathological findings:

In cases with chronic localised cholangitis the liver was manifested grossly by thickening of the bile ducts in a localized area,

thick fibrous wall and calcium deposits in some cases. Cholangitis and bile ducts hyperplasia may be attributed to the toxic metabolite of *Fasciola* and its mechanical irritation to the bile ducts epithelium (Jubb *et al.*, 1993). These findings are in agreement with previous studies by Behm and Sangster (1998) and Maclachlan and Cullen (1995). The localization of the lesion in small area of the liver may be attributed to the infestation of the animal with small number of *Fasciola* metacercariae and to the presence of resistance mechanism to reinfection with *Fasciola* in cattle. This resistance against challenge flukes begins at or soon after penetration of the liver (Doy and Hughes, 1984).

In cases with sawdust liver; the liver appears containing several minute yellowish foci or necrosis, as if the same numbers of sawdust granules scattered over the it and characterized histologically by presence of multifocal area of necrosis with neutrophilic infiltration.It was reported that sawdust liver seem to have unknown etiology, vitamin E deficiency has been suggested as the cause (Smith *et al.*, 1972). Laboratory results of the same authors are in agreement with the gross and histopathological findings observed in the present study.

Hematological findings:

The blood picture in cases of chronic localized cholangitis showed insignificant decrease ($p > 0.05$) in the total erythrocytic count and hemoglobin concentration and insignificant changes in the MCV, MCH and MCHC. However, there were a significant increases ($p<0.01$) in the total leucocytic count, neutrophils and eosinophils percent. The insignificant changes in the red blood cell pictures may be attributed to the localization of the lesion in certain and small area of the liver. Leucocytosis in cases of chronic localized cholangitis associated with fascioliasis was reported previously by Behm and Sangster (1998). The authors referred leucocytosis to the inflammatory changes produced by the parasite during migration. The same authors added that neutrophilia usually accompanies cases of chronic localized cholangitis in cattle. Significant increase in the eosinophil counts in cases of *Fasciola* worm infestation were reported previously by Haroun and Hussein (1975) and Conboy and Stromberg (1991).

Biochemical findings:

The increased hepatic G6PD activity in cases of sawdust liver may be attributed to the increased free radical levels in the hepatic tissues. Free radical stress can lower NADPH and increase NADP concentrations that stimulate the hexose monophosphate shunt (G6PD) (Miller *et al.*, 1993). Many studies under conditions of oxidative stress,

have shown that G6PD of the pentose phosphate pathway are routinely elevated (Tian *et al.*, 1998). G6PD may have a dominant role in the control of output of Glutathione (GSH) (Salvemini *et al.*, 1999), which is required for the reduction of H_2O_2 via GSH-Px (Agar and Suzuki, 1982 and Sarkar *et al.*, 1998). Free radicals serve as mediators of tissues destruction and also act as a chemoattractants for neutrophils (Stohs, 1995). This may explain the neutrophilic infiltration of the necrotic foci in cases of sawdust liver. Because superoxide (O_2^-) and hydrogen peroxide (H_2O_2) used by phagocytic cells to kill bacteria (Miller *et al.*, 1993), self-destruction of neutrophils during inflammation results in the release of free radicals (Stohs, 1995). It was reported that leucocytes contain higher activities of GSH-Px (Richard and Lawrence, 1979)). In fact the activity of hepatic GSH-Px might be decreased due to the toxic effect of free radicals and appeared normal on measurements due to the homogenization of the liver tissues that containing large numbers of neutrophilic infiltration.

Aspartate aminotransferase is widely distributed in many tissues being especially abundant in the liver, myocardium and skeletal muscles. Its activity increased rapidly following necrosis, disease or traumatic damage to these tissues, where the enzyme diffuse across the damaged cell wall and enter the blood (Benjamin, 1970). Liver is the major contributor of γ-GT to plasma and it is localized both in the hepatocytes and in the biliary tree and its richest distribution in the epithelium of the bile ductules (Blei, 1994). It was reported that serum γ-GT activity is specific for liver disorders in cattle (Mullen, 1976, Bouda *et al.*, 1980 and Pearson, 1990). GLDH is considered as one of the best enzyme for hepatocellular damage (Braun *et al.*, 1986). ADA activity was significantly elevated in the serum in cases of acute liver injury (Kurata, 1995) and may be used as a predictive index for the inflammatory changes in the liver (Yasuda *et al.*, 2001). Increased in total bilirubin level was reported to be specific for hepatic dysfunction in cattle (West, 1991). Albumin is synthesized in the liver and liver disease especially chronic cases reduces its level (Joan, 1982). The author added that general increase in globulin levels occur in chronic inflammatory states.

It could be concluded that; serum total protein, albumin and globulin are insensitive indicators for sawdust liver. Measuring serum activities of AST, GGT, ADA, GLDH and serum total bilirubin levels are helpful in diagnosis of the investigated cases. Increase hepatic G6PD activity in cases of sawdust liver may be attributed to increased free

radical levels. With the aid of liver biopsy, measurement of hepatic G6PD activity may be helpful in diagnosis of sawdust liver.

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Table1: Mean ($\bar{X}$) and standered deviation (Sd) values of total RBCs count, Hb. Concentration, PCV, MCV, MCH and MCHC in control and diseased cattle.

Animal Group		Total RBCS count(T/l)	Hb. Conc. (g/l)	PCV (%)	MCV Fl	MCH (pg)	MCHC (%)
Control No.= 20	$\bar{X}\pm$Sd	7.06±0.50	116.7± 6.80	32.7±2.22	46.47±3.94	16.54±0.55	35.59±2.73
	Range	6.18–7.95	106.0-128.0	28.00–39.00	40.25–55.01	15.63–17.54	31.28–40.00
Chronic localized cholangitis No. = 15	$\bar{X}\pm$Sd	6.72 ± 0.5 NS	110.9. ± 7.0 0 NS	32.4 ± 1.45 NS	48.32 ± 2.64 NS	16.51 ± 0.75 NS	34.23 ± 1.61 NS
	Range	6.23 – 7.82	102.0 – 126.0	30.00 – 36.00	43.47 – 51.98	14.78 – 17.84	30.58 - 37.05

Range: Minimum – Maximum No.: Number of examined animals. NS: Non-significant.

Table 2: Mean ($\bar{X}$) and standered deviation (Sd) values of total leucocytic count, neutrophils, lymphocytes, eosinophils, monocytes and band cell percents in control and diseased cattle.

Animal Group		Total leucocytic count (G / l)	Neutrophils (%)	Lymphocytes (%)	Eosinophils (%)	Monocytes (%)	Band cells (%)
Control No.= 20	$\bar{X}\pm$Sd	8.12 ± 1.99	35.80 ± 4.54	57.25 ± 4.73	2.55 ± 0.88	2.25 ± 0.85	2.15 ± 0.87
	Range	4.30 – 11.20	28.00 – 45.00	48.00 – 65.00	1.00 – 4.00	1.00 – 4.00	1.00 – 4.00
Chronic localized cholangitis No. = 15	$\bar{X}\pm$Sd	11.60 ± 3.17**	56.13 ± 7.31**	29.53 ± 4.48**	9.26 ± 4.43**	2.66 ± 0.89 NS	2.40 ± 0.91 NS
	Range	7.40 – 20.30	39.00 – 70.00	23.00 – 41.00	3.0 – 16.0	1.00 – 4.00	1.00 – 4.00

Range: Minimum – Maximum No.: Number of examined animals. NS: Non-significant.
** :Highly significant ($p < 0.01$).

Table 3: Mean ($\bar{X}$) and standered deviation (Sd) values of total protein and its fractions in control and diseased cattle

Parameters / Groups		Total Protein (g/L)	Albumin (g/L)	Globulin (g/L)	A/G ratio (%)
Control No. = 25	$\bar{X}\pm Sd$	64.30 ± 3.80	39.10 ± 4.90	25.20 ± 4.90	1.6 ± 0.5
Sawdust liver No. = 9	$\bar{X}\pm Sd$	65.35 ± 3.36 ^{NS}	40.01 ± 5.49 ^{NS}	25.34 ± 4.12 ^{NS}	1.63 ± 0.39 ^{NS}

NS: Non-significant.

Table 4: Mean ($\bar{X}$) and standered deviation (Sd) values of serum enzyme activities and total bilirubin levels in control and diseased cattle.

Parameters / Groups		AST (U/L)	GGT (U/L)	ADA (U/L)	GLDH (U/L)	Total Bilirubin (µmol/l)
Control No. = 25	$\bar{X}\pm Sd$	59.56 ± 13.98	23.74 ± 5.37	7.44 ± 2.69	16.12 ± 8.44	4.25 ± 1.79
Sawdust liver No. = 9	$\bar{X}\pm Sd$	137.0 ± 81.1**	39.03 ± 12.03**	11.47 ± 7.28*	39.67 ± 21.96**	11.91 ± 4.31**

NS: Non-significant. *: Significant ($p < 0.05$). ** : Highly significant ($p < 0.01$).

Table 5: Mean ($\bar{X}$) and standered deviation (Sd) values of the activities of erythrocytic and hepatic G6PDH and GSH-Px in control and diseased cattle.

Parameters Groups		Erythrocytic G6PDH (U/g Hb)	Erythrocytic G6PDH (U/g protein)	Hepatic GSH-Px (U/g Hb)	Hepatic GSH-Px (U/ g Protein)
Control No. =25	$\bar{X}\pm Sd$	7.58 ± 1.28	2.28 ± 0.48	172.48 ± 30.66	24.32 ± 4.77
Sawdust liver No. = 9	$\bar{X}\pm Sd$	7.30 ± 1.17 ^{NS}	4.95 ± 1.37**	171.05 ± 40.68 ^{NS}	27.44 ± 6.98 ^{NS}

NS: Non-significant. ** : Highly significant ($p < 0.01$).

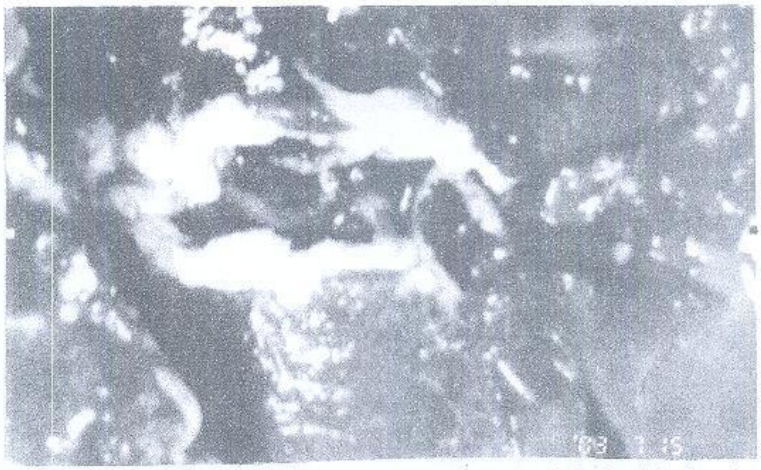

Figure (1): Liver of cattle in cases of chronic localized cholangitis showing fibrosed and thickened bile ducts.

Fig. (2): Cattle liver in cases of chronic localized cholangitis. The increased portal C.T. is dense and chollagenous. (H. & E. X 100).

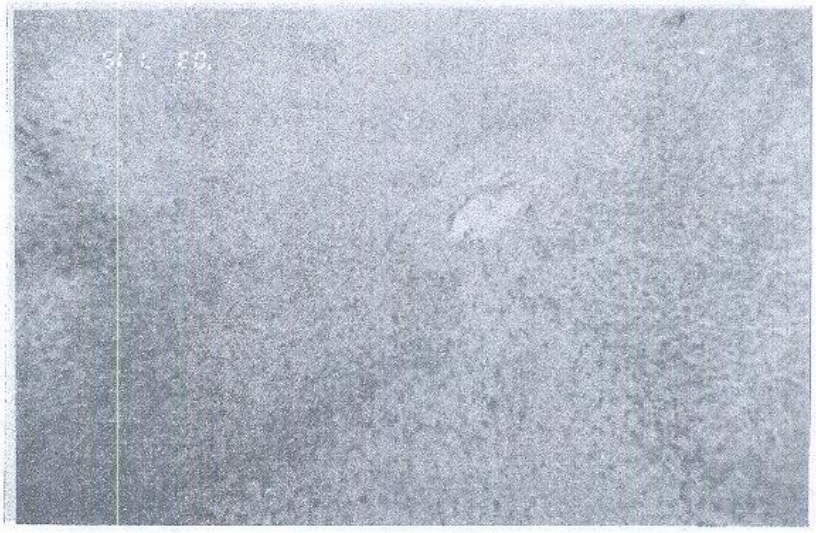

Fig. (3): Liver of cattle in cases of sawdust liver showed multifocal area of necrosis with neutrophilic infiltration (H. & E. X 100).