

The pathological changes associated with endotoxemia in rabbits.

By

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Summary

Endotoxin is responsible for different body systems changes. This study designed by used rabbits, gave intravenous endotoxin doses to study the pathological findings.

The gorse pathological findings were characterized by petechial and ecchymotic hemorrhage in the pleura, kidneys swollen and pulpy.

The histopathological changes manifested by fatty degeneration, bile ducts hyperplasia and fibrosis of liver. Lungs showed edema, emphysema and bronchial dilatation, kidneys with dilated Bowman's spaces and dilated tubules. Spleen showed hyperplasia in red and white pulp, lymphoid tissues hyperplasia. Brain and meninges showed edema and congestion of meningeal membrane. Most organs showed diffuse thrombus formation, which was organized during 12-20th days.

Introduction:

Endotoxin may gain entrance to the circulation system from damaged gut epithelium, coliform mastitis in dairy cattle and coli form septicemia in newborn calves (1). The common treatment for endotoxemia with antibiotic can produce 2000 fold increase in the blood level of endotoxin as result of the bacterial wall lyses (2). Endotoxins usually activate kinin system (3), as well as inflammatory response syndrome (SIRS) cascade, in the absence of documented infection (4). So this study was aimed to determine the effect of pure endotoxin on the clinical findings and pathological findings.

Materials and Methods:

Vial of endotoxin (*Salmonella typhimurium* endotoxin, Difco-USA), dissolved with PBS 7.2 pH in a serial dilutions to reach 0.1mg /ml. (kept by freezing at -18°C).

Twenty adult healthy male rabbits, domestic breed were used, between 1.5 -1.9 Kg weight, divided into 4 equal groups, and given endotoxin intravenously in doses of 5, 15 and 20 µg/Kg body weight for groups I, II and III respectively, while group IV gave PBS as a control.

Rabbits clinically examined, then scarified at 4, 8, 12, and 16 and 20 days for gross and histopathological studies, tissue samples were taken from liver, lungs, kidneys, spleen, brain, heart, intestine and lymph nodes. Other samples were taken at day 20th from control group .All these samples were processed as in (5)

Results and discussion:

The obvious gross changes observed followed by endotoxin injection were foci of petechial hemorrhage in the liver at the early time of experiment, while during the following time changed to yellowish- white foci, indicated fatty changes (6), also petechial and ecchymotic hemorrhage in the subcapsular region of the lungs and the cut section showed foamy content were related to intraalveolar edema (7). Kidneys were swollen, soft in consistency due to hypertrophy (8).

The main histopathological changes have been seen in the liver, lungs, and kidneys. Their were degeneration, extensive congestion and thrombus formation. These lesions also associated with extensive infiltration of inflammatory cells, due to leukocytic recruitment, as effect of endotoxin on cell activation, and these recruited cells especially neutrophils were responsible for inducing parenchymal cell injury in the body organs (9). Endotoxin increased adherence between platelets and their adherence to endothelia of blood vessels, that due to expression of adhesion molecules and then thrombus formation (10).

The result showed vaculation of hepatocytes during 8th -20th days (figure-1), that was related to fatty changes, which occur under effect of endotoxin on hepatocytes, either by imbalance in the uptake of fatty acid by hepatocytes or over supply of triglycerides to hepatocytes and decreased fatty acid oxidation. The activated hepatocytes showed variable sized vacuoles in there cytoplasm that may displaced the nucleus to the periphery of the cell, which was sever in group III (11). Other lesion were bile duct proliferation, and fibrosis in liver (figure-2),which may related to the injury caused by TNF α , followed by repair of the damaged young hepatocytes (12) .

There was thickening of interstitial tissue of lung, which related to congestion, edema and neutrophils infiltration after endotoxemia (figure-3), these changes in response to increased blood pressure of microcirculation, as well as endotoxin cause subsequent increased capillary permeability (6). Also infiltration of neutrophils were lead to alveolar damage and development of emphysema (13), other lesion of lung included bronchial dilatation (Bronchiactasia), which occur in response to relaxation of smooth muscle fibers following endotoxemia, that in association with high level of nitric oxide (12), also the proliferation of bronchial epithelium especially during 8-16th days post injection was under nitric oxide effect (14).

Kidneys showed Bowman's space dilatation, other area of shrunken glomeruli and such glomeruli showed increase cellularity, that were related to immune complex deposition in glomerular basement membranes (11), other lesions were dilated tubules and some other tubules with swollen their epithelial lining, which revealed degeneration of these cells and developed cloudy swelling, these changes were related to ischemia following endotoxemia (7). There were highly pigmented casts accumulated in the lumen of renal tubules(figure-4), may due to hemoglobin casts, which occur in response to increased permeability for hemoglobin after its liberation from erythrocytes (intravascular hemolysis) (15).

Brain and meninges showed blood vessels congestion and edema, which causes thickening of meningeal wall (figure-5), that endotoxin increased permeability of meningeal blood vessels (16). Other findings such as perineuronal edema and focal gliosis, due to the effect of TNF α on brain tissue and subsequent increase of glial cells (7).

There was hyperplasia in the white pulp of spleen also peribronchial lymphoid tissue of the lungs (figure-6) and lymph nodes hyperplasia, especially during 12th -20th days, which revealed to endotoxin action as a mitogen to stimulate these lymphoid cells. Also Roitt, *et al.* (17) assessed the endotoxin mitogenecity for B cell region. The red pulp of spleen showed hypertrophy, related to endotoxin stimulated the extramedullary erythropoiesis; these changes were reported by previous authors in response to overwhelming infections (8).

Small intestine showed lesions such as increased cellularity of lamina propria and congestion of blood vessels, these changes were due to the effect of endotoxin on blood vessels and increased their permeability, but the intestinal mucosa showed no lesion, that endotoxin may not targeted the mucosa (18).

The pathological changes of this study were more prominent in group III rabbits, which given high dose of endotoxin then subsequent inflammatory process and organ injury (19).

Figure (1) Liver from group II;
there is generalized vacuolation of
hepatocytes (a) and sinusoidal
congestion (\Rightarrow) (H&E X200).



Figure (2) Liver from group III;
→ there is extensive fibrous tissue
proliferation in the portal area at 20th
day (Vangieson's stain X100).

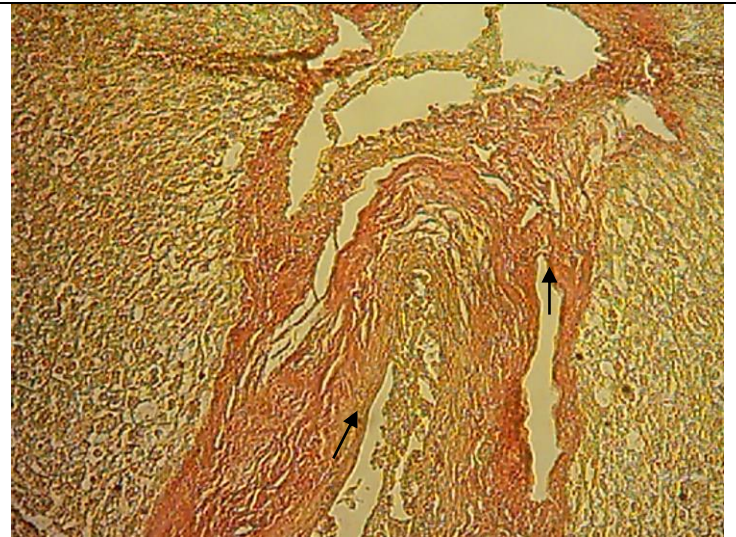


Figure (3) Lung from group III;
there is pulmonary edema and
congestion (a) in addition to dilatation
of alveoli (b) neutrophils infiltration
(\Rightarrow) at 4th day (H&E X100).

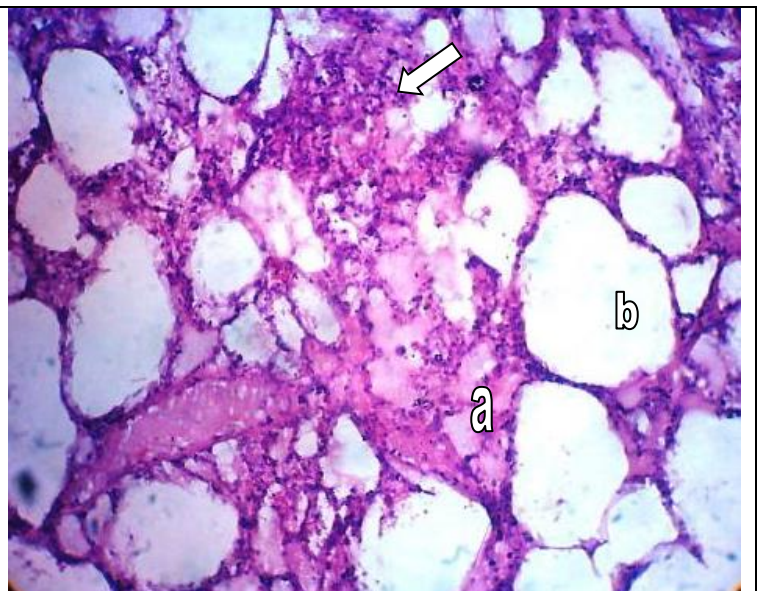


Figure (4) Kidney from group III; there is extensive congestion of the blood vessels and (→) presence of renal casts in the renal tubules (⇨) (H&E X40).

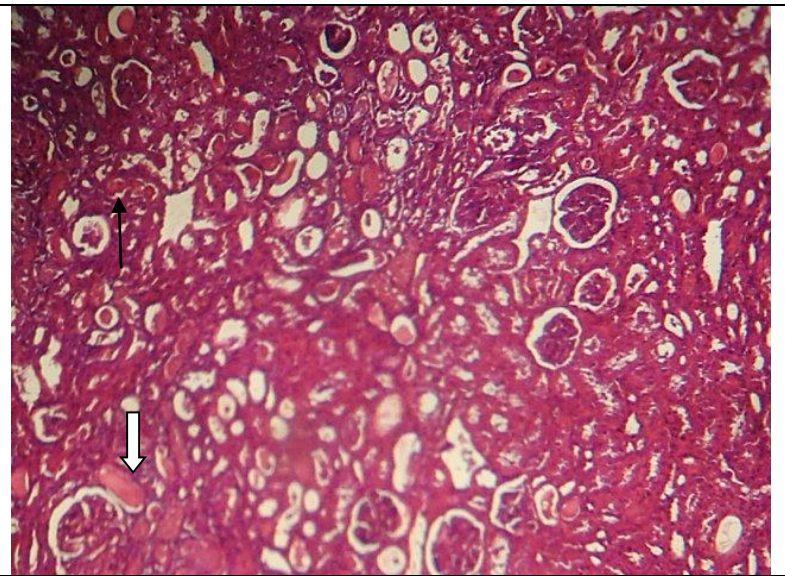


Figure (5) Brain and meninges from group III; showed thickening resulted from edema (→) and congestion (⇨) also perineuronal edema (H&E X100).

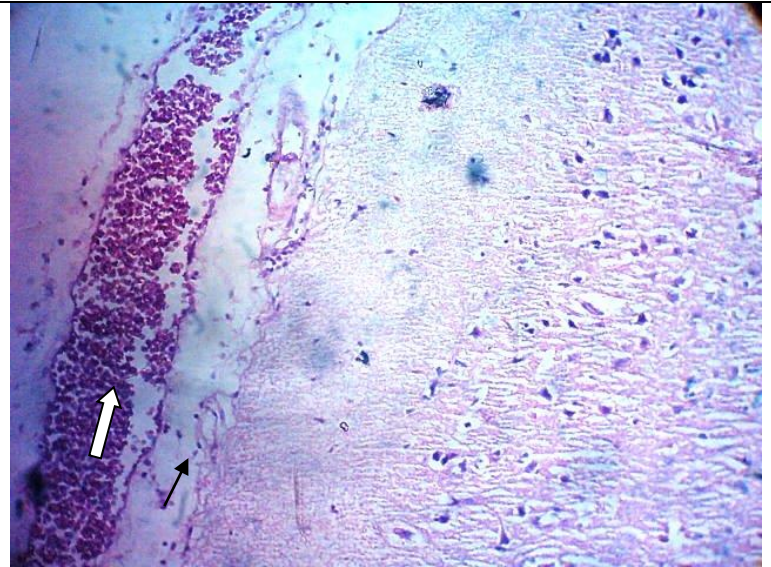
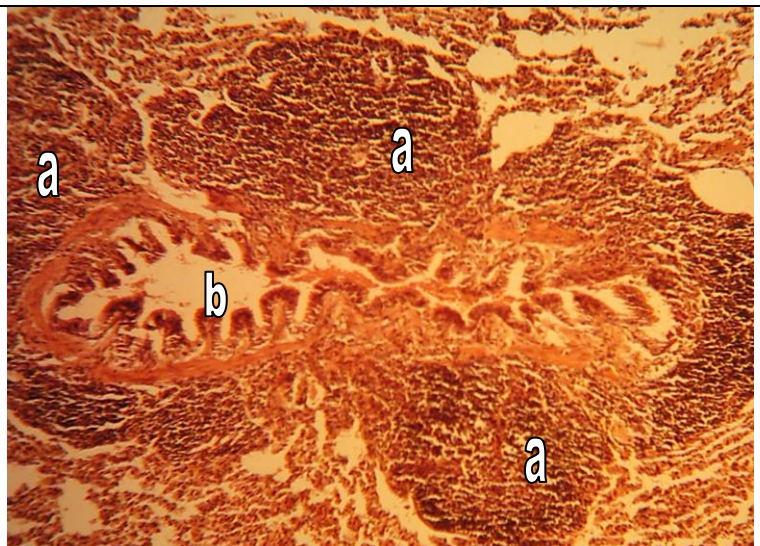


Figure (6) Lung from group III; there is extensive peribronchial lymphoid tissue hyperplasia (a) cause narrowing to the bronchus (b) (H&E X100).



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التغيرات المرضية المرافقة لوجود الذيفان الداخلي للدم

(endotoxemia) في الارانب

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الخلاصة

أن للذيفان الداخلي تأثيراً واسعاً على مختلف أجهزة الجسم ، لذا أعدت هذه الدراسة وبأستخدام أربعين أرنباً أعطيت بالحقن الوريدي للذيفان للكشف عن الأعراض المرضية. ظهر بعد حقن الذيفان تغيرات مرضية واضحة وتميزت عيانياً بحصول نزف حبري و كدمي على خلب الرئتين وان الكلى لينة الملمس ومنتفخة ، بالإضافة الى احتقان الأعضاء الأخرى . تميزت التغيرات النسجية بالتغير الدهني للكبد وكذلك تضخم قنوات الصفراء والتليف ، في حين أن الرئتين أظهرتا الوذمة والاحتقان وانتفاخ الاسناخ والتشنج الخلالي وأيضاً توسع القصبات . الكلى أظهرت توسعاً في حيز بومان وتوسع النبيبات. وكان النسيج الشبكي البطاني للطحال كثير التنسج، وكذلك اللب الأبيض له ، أما الأنسجة اللمفية الأخرى مثل النسيج اللمفي حول القصبات الهوائية والعقد اللمفية ولطح باير، جميعها أظهرت تضخماً في الأنسجة اللمفية. أظهر الدماغ والسحايا وجود وذمة واحتقان وكذلك زيادة في الخلايا الدبقية ووذمة ماحول العصبية، في حين أن أغلب الأعضاء الأخرى قد أظهرت احتقاناً وتخلق الخثرة داخل الاوعية الدموية، والتي كانت متعضية خلال الفترة من 12-20 يوماً.