قسم : الباثولوجيا .
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مرض الباستيرلا في الأرانب والعوامل المهيأة له

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تمت هذه الدراسة على خمسة وعشرون من ذكور وأرانب البوسكات تم احضارها من وحدة الحيوانات المعملية بجامعة أسيوط. بعد التأكد من خلوها من الأمراض البكتيرية والطفيلية قسمت الى خمسة مجموعات منفصلة كل منها خمسة أرانب .

تمت عدوى الحيوانات صناعيا اما بميكروب الباستيرلا وحدة أو بالاضافة الى ميكروب البورد يتلا أو ميكروب الدبلوكوكس او الميكروب العصوى القولوني وتركت المجموعة الخامسة بدون عدوى كضابط للتجربة .

لوحظت الأعراض ، أجريت الصفة التشريحية للحيوانات النافقة وأخذت عينات من القصبة الهوائية ، الرئة ، الكلية ، الامعاء ، الكبد ، المخ ، الفحص الميكروسكوبي .

فحصت العينات ميكروسكوبيا ونوقشت النتائج وقد تبين أن العوامل البكتيرية التي استخدمت في البحث تلعب دورا في شدة الاصابة بميكروب الباستيرلا .

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RABBIT PASTEURELLOSIS AND ITS INITIATING FACTORS

(With 2 Tables and 15 Figures)

By
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(Received at 4/12/1982

SUMMARY

The present work was carried out on twenty five male Boskat rabbits, obtained from the laboratory Animal Unit, Assiut University. The animals were divided into five groups and experimentally infected with Pasterurella multocida alone or associated with Diplococcus pnumonae, E.Coli or Bordetella bronchiseptiea. The clinical picture was recorded and post-mortum examination was carried out as well as reisolation of the inoculated strains was conducted. Trachea, Lung. Kidney, intestine, liver and brain were examined microscopically.

The present work indicated that such bacterial agents not only play an important role in initating rabbit pasteurellosis but also shared in the severity of infection.

INTRODUCTION

Rabbits, in addition to their nutrative value, are also used as experimental animals in medical and biological institutes for both diagnostic and research purposes. Pasteurellosis still represents the most common bacterial infection in rabbits and cause high lossses almost in all ages especially in those under adverse conditions.

In Egypt, isolation, identification and capsular typing of Pasteurella multocida in rabbits was studied by SAAD (1970), ZAHER, et al. (1976), EL-GHAWAS (1980) and in upper Egypt by YOUNES (1982).

A number of secondary microorganisms were isolated in association with Pasteurella multocida infection in rabbits (YOUNES, 1982) and it was persumed that lowering the resistance of animals caused and extension of the previously existing inflammatory process.

The present work was designed to elucidate the role of Diplococcus pneumonae, Bordetella bronchiseptica and E.coli in the pathogenesis of Pasteurellosis induced in rabbits.

MATERIALS and METHODS

Pasteurella multocida type A, E.coli O 25:K11 (L), Diplococcus pneumonae and Bordetella bronchiseptica strains isolated from rabbits by YOUNES (1982) were used in our study.

The present work was carried out on thirty male Boskat rabbits, 8-weeks-old, obtained from the laboratory Animal Unit, Assiut University. The animals were housed in seperate cages, fed standard ration supplemented with vitamins and minerals and were put under observation for a week before the experimental infection.

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Random samples of five animals were slaughtered, and subjected to post-mortum examination, also bacteriological and parasitological examinations were conducted. Twenty five bacteriologically and parasitologically free animals were removed and isolated for the experiment. The animals were divided into five groups, each consisted of five animals. The rabbits were experimentally infected as shown in Table (1). During the experimental period, the clinical picture was recorded and post-mortum examination was carried out shortly after death of the animals. Resolation of the inoculated strains was conducted. From all, dead and sacrificied animals, specimens for histopathological examination were taken from the lung, trachea, kidney, intestine, liver and brain. The samples were fixed in 10% neutral buffered formalin, embeded in paraffin, sectioned, stained with haematoxylin and eosin and examined.The control animals were sacrificied at the end of the experiment which lasted for and specimens for microscopical examination were taken.

RESULTS

Clinical observations:

The animals in group one, which were infected with Pasteurella lepiseptica only, showed depression, rough fur, dull eyes, laboured breathing, frothy nasal discharge and only two animals showed diarrhea. Similar picture, although severe form, was also observed in group two, in which the animals were infected with Pasteurella and Diplococcus pneumonae. In group three where Bordetella bronchiseptica was used in addition to Pasteurella infection, Laboured breathing and the mucopurulent discharge were prominent. In addition to the respiratory symptoms aforementioned, severe diarrhea occurred in all animals of group four infected with both Pasteurella and E.coli.

In regard to the survival period after infection (Table 2), all animals of group one died within the fifth day after infection. Animals of group two and those of group three died within the third day after infection, but animals of group four died within the sixth day post-infection.

Macromorphological results:

In group one, the trachea was congested, inflammed and filled with a frothy blood-stained exudate. The lungs were congested and showed scattered, well demarcated reddish brown to reddish grey focal areas of consolidations. The pneumonic foci were usually bilateraly and mainly affecting the ventral portion of the cardiac lobes. The kidneys were slightly congested while the other organs showed no changes.

In group two, the post-mortum picture was similar to group one except, that, in the lungs, the areas of consolidations progressed to involve much of the entire cardiac lobes. On cut section the lung was firm and the bronchi were filled with thick exudate. The meningeal vessels were congested.

In group three, in almost all the animals, the pleural cavity contained varied amounts of clear sanguineous fluid. The lung was more severely affected when compared with the above two groups; the foci of consolidation could be observed in the crainioventral portions of the diaphragmatic lobes. The bronchial lumens were filled with slimy, yellowish grey exudate. Areas of atelectasis and others of emphysema could be also observed.

In group four, the trachea was inflammed but to a lesser degree than in other groups. Lung consolidation and bronchial exudations were restricted to few focal areas in the cardiac

lobes. The kidneys were swollen and pale in colour. The liver was slightly enlarged, congested and had rounded borders. The intestines contained a large amount of catarrhal exudate and showed a congested mucosa. These inflammatory changes could be seen only in the small intestine.

Micromorphological results:

In group one the trachea showed epithelial desquamation and severe hyperaemia (Fig. 1). The lungs revealed areas of fibrinous pneumonia in which the fibrin deposited in the alveolar spaces (Fig. 2). Many areas showed neutrophilic cellular infilterations. The interalveolar capillaries were greatly congested. In some areas varied numbers of alveolar macrophages could be observed (Fig. 3). The kidneys showed marked tubulonephrosis) manifested by degeneration of the epithelial lining of their tubules, the intertubular capillaries were severly congested and some of them were ruptured (Fig. 4).

In group number two, degenerative changes and desquamation of the epithelium associated with hyperaemia and haemorrhages of the mucous membrane could be observed in the trachea (Fig. 5). The lungs revealed the picture of fibrinous pneumonia similar to animals of group one. Moreover slight degree of bronchitis and bronchiolitis manifested by degeneration of their epithelial lining as well as peribronchial hyperaemia and oedema could be seen. The pulmonary ramifications showed intimal degenerative changes and perivascular oedema Fig. (6). Changes in the kidneys were identifical to those in group one. The brain showed congestion of the cerebral blood vessels associated with oedema in virchow Robin spaces and slight microgliosis (Fig. 7).

In the third group, the tracheal wall showed severe oedema and hyperaemia (Fig. 8). Areas of haemorrhages and mononuclear cell infilterations could be observed. In the lung bronchitis and bronchiolitis were marked in which the lumens were partially filled with cellular exudate containing mononuclear cells and desquamted epithelial cells. The rest of the lung parenchyma showed severe hyperaemia, oedema and inflammatory cellular infilterations (Fig. 9), in which polymorphnuclear leucocytes consisted the main type.

In the fourth group, microscopic examination of the trachea revealed moderate oedema, inflammatory cell infilteration and slight hyperaemia (Fig. 10). Animals of this group suffered from interstitial pneumonia (Fig. 11), in which the interalveolar septa were thickened due to accumulation of cellular inflammatory exudate. The predominant were mononuclears. In the kidneys, in addition to tubulonephrosis, slight interstitial reaction, degeneration of vascular endothelium and perivascular oedema could be observed (Fig. 12). Changes in the liver consisted of hepatic cells with marked pyknotic and karyorrhetic nuclei could be seen (Fig? 13). In the intestine, epithelial degeneration and desquamation in addition to submucosal hyperaemia and oedema (Fig. 14 a,b) resembled the main findings.

In the fifth group, which was used as controls, neither clinical nor morphological alterations could be observed except in one animal, its liver showed minute, focal greyish white spots which on microscopic examination revealed hyperplastic cholangitis (Fig. 15).

DISCUSSION

As it is, reported by (WATSON et al., 1975, SPANOGHE and OKERMAN 1978 and HIME 1979), the infectious and non infectious agents which act as stressful conditions were necessary for overt the rabbit snuffles to appear. The present work was designed to elucidate the role of Bordetella bronchiseptica, Diplococcus pneumonae and E.coli which were isolated by YOUNES (1982) in association with Pasteurella multocida as infectious agents which may deminish

the animal resistance.

For the present study, twenty five, male Boskat rabbits, about 800 - 1,000 gm in weight were obtained from the laboratory animal unit, Assiut University. These were selected to exclude if possible, previous attacks of rabbit pasteurellosis. The rabbits were exposed to cultures of either Pasteurella multocida alone or associated with, Diplococcus pneumonae, Bordetella bronchiseptica or E.coli.

Symptoms of respiratory affection were a prominant features in all groups. More or less, these clinical signs were also describred by (OSTLER, 1961, ZAHER et al., 1976 and YOUNES 1982). Deaths occurred between 1st. and 6th. day-post-infection, the mortality rate was 100% and this indicates that the locally isolated strains were highly pathogenic for suceptible rabbits. Nearly in all groups, the microscopic lesions of dead rabbits were restricted to the respiratory tract. In group three, the lung was more severely affected when compared with the other groups. Not only in character but also in distribution where the pneumonic foci extended also to involve the crainioventral portions of the diaphragmatic lobes. This could be probably due to the synergestic effect of the two microorganisms especially that both have been associagted with pneuomonia (WISSNER 1960, SATO and NAMIOKA 1967, FLATT and DUNGWORTH 1971 and SPANOGHE and OKERMANN 1978). The involvement of the tracheal, bronchial and peribronchial as well as involvement of the lung alveolar tissue indicates that the invection having entered by the bronchial route. Microscopically it was not easy to differentiate pneumonia produced by either organism, since the characterstics of the lesion were more or less similar and this also was observed by WATSON et al., (1975). Similar lesions were also noted in the naturally occuring disease (LÖLIGER, ALBERTI and MATTHES 1972, HIPPE 1979). In the vicinity of the pneumonic foci, they were almost areas of emphysema and others of atelectasis. The formers were due to the decreased space occupied by nieghbouring alveoli which were either atelectatic or consolidated. The atelectatic areas could be attributed not only due to the plugging of the air ramifications by masses of exudate but also due to the pressure of the consolidated areas. The presence of the large cells with eccentrically situated nuclei and a rather extensive cytoplasm in varied numbers, some of these cells must be monocytes from the blood, others doubtles came from the reticuloendothelial system and there is a considerable evidence that many of them are proliferating alveolar epithelium. In the fourth group, interstitial pneuomonia was observed, this could probably be due to the relative prolonged survivial time which gave the chance for the interstitial inflammatory reactions.

The brain lesions of group two in the present study could be assumed either to the toxins produced by pneumocci (DICK and GIMMEL 1971, BURNNET and SCHUSTER 1073) or due to Pasteurella multocida, Khera, Pandurangarao and Mall (1971).

The vascular degenerative changes observed in the second and fourth groups could be attributed to the toxins of Diplococcus pneumonae and E.coli respectively. In the fourth group, the inflammatory process with its alterative and exudative phenomenas were observed in the intestines and could be assumed to the E.coli infection, PRESCOTT (1978).

The hyperplastic cholangitis seen in one animal of the control group was due to infect with coccidia.

The present work indicated that bacterial agents as Bordetella bronchiseptica, Diplococcus pneumonae and E.coli not only play an important role in iniating rabbit pasteurellosis but also shered in the severity of infection.

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DESCRIPTION OF FIGURERS

Fig. (1): Showing epithelial desquamation and hyperaemia of the trachea. H & E. (X400).
Fig. (2): Lung showing fibrinous pneumonia. H. & E. (X400).
Fig. (3): Lung showing alveolar macrophages. H. & E. (X400).

Fig. (4): Kidney showing tubulonephrosis and focal haemorrhage. H. & E. (X250).

Fig. (5): Trachea showing epithelial desquamation, hyperaemia and haemorrhage. H. & E. (X250).

Fig. (6): Pulmonary vessel showing intimal degeneration and perivascular oedema. H. & E. (X400).

Fig. (7): Brain showing congestion and oedema. H. & E. (X250).

Fig. (8): Trachea showing oedema and hyperaemia. H.& E. (X250).

Fig. (9): Lung showing bronchiolitis. H. & E. (X400).

Fig. (10): Trachea showing oedema. H. & E. (X250).

Fig. (11): Lung showing interstitial pneumonia. H. & E. (X160).

Fig. (12): Kidney showing tubulonephrosis, vascular endothelial degeneration and perivascular oedema. H. & E. (X400).

Fig. (13): Liver showing karyorrhetic and pyknotic nuclei. H. & E. (X250).

Fig. (14): Showing enteritis. H. & E. (X250).

Fig. (15): Liver showing hyperplastic cholangitis. H. & E. (X400).

Table (1): Showing the animal grouping type of infection, its route and dosage

Animal group	Number of animals	type of infection	Route and dose
G.One	5	Posteurella multocida A	Intranasal &X10 $^{9+}$
G.Two	5	Pasteurella multocida A + Diplococcus pneumonae	Intranasal 5X10 $^{9+}$ Intraperit. 1 ml. broth cult. ++
G.Three	5	Pasteurella multocida A + Bordetella brnchiseptica	Intranasal 5X10 $^{9+}$ Intranasal 3.5X10 $^{10+}$
G.Four	5	Pasteurella multocida A + E.coli 025 K 11 (L)	Intranasal 5X10 $^{9+}$ Oro-gastric 4X10 $^{10+++}$
G.five	5	Non infected controls	

+ : Watson et al. 1975. ++ : El-Ghawas, 1980. +++ : Cantey and Blake, 1977.

Table (2): Showing the survival period of animals after infection

Animal group	Type of Infection	day-post infection	Number of Dead animals
First	Pasteurella multocida A	3 rd. 4 th. 5 th.	1 1 3
Second	Pasteurella multocida A + Diplococcus pneumonae	1 st. 2 nd. 3 rd.	1 2 2
Third	Pasteurella multocida A+ Bordetella bronchiseptica	1 st. 2 nd. 3 rd.	2 2 1
Fourth	Pasteurella multocida A + E.coli	3 rd. 4 th. 5 th. 6 th.	1 1 2 1

Fig. (1)

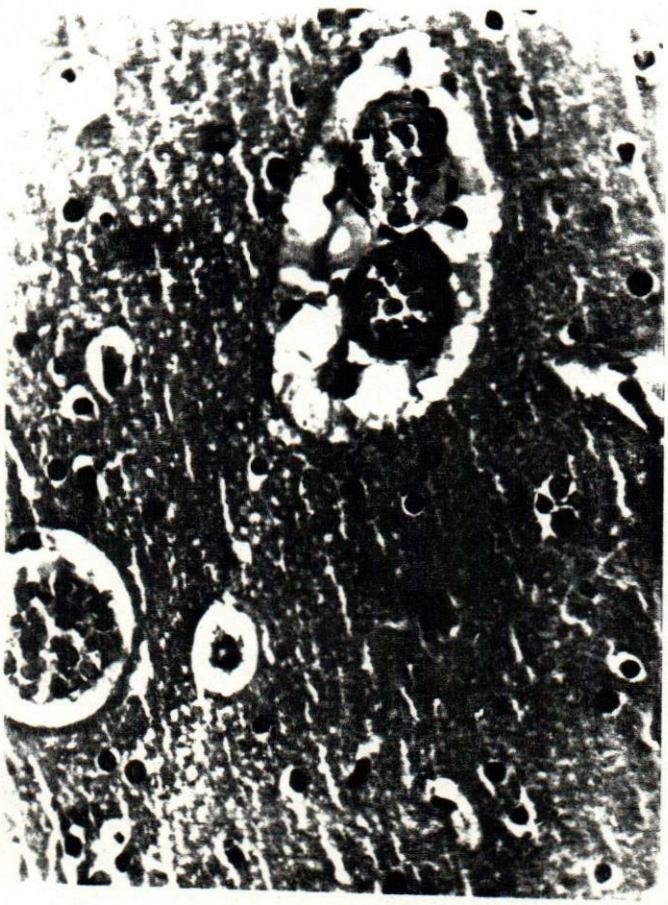
Fig. (2)

Fig. (3)

Fig. (4)

Fig. (5)

Fig. (6)



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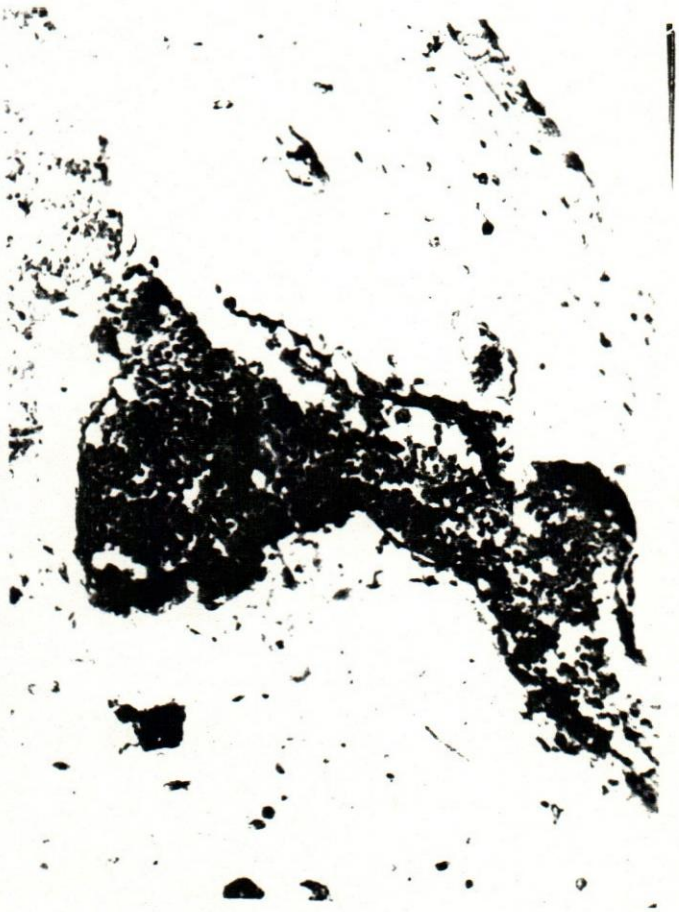

Fig. (8)

Fig. (9)

Fig. (10)

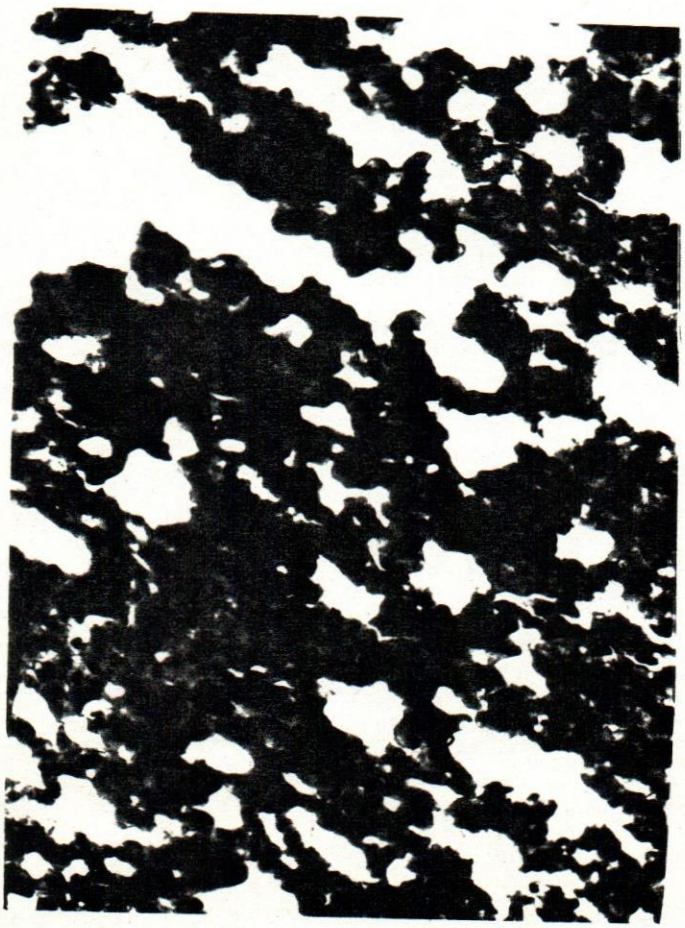

Fig. (11)

Fig. (12)

Fig. (13)

Fig. (14a)

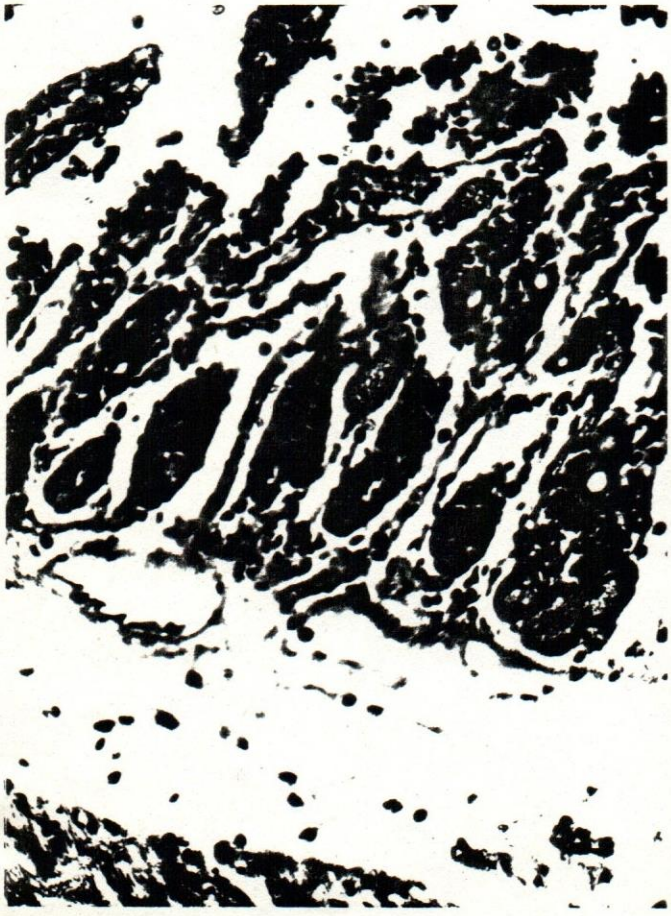

Fig. (14b)

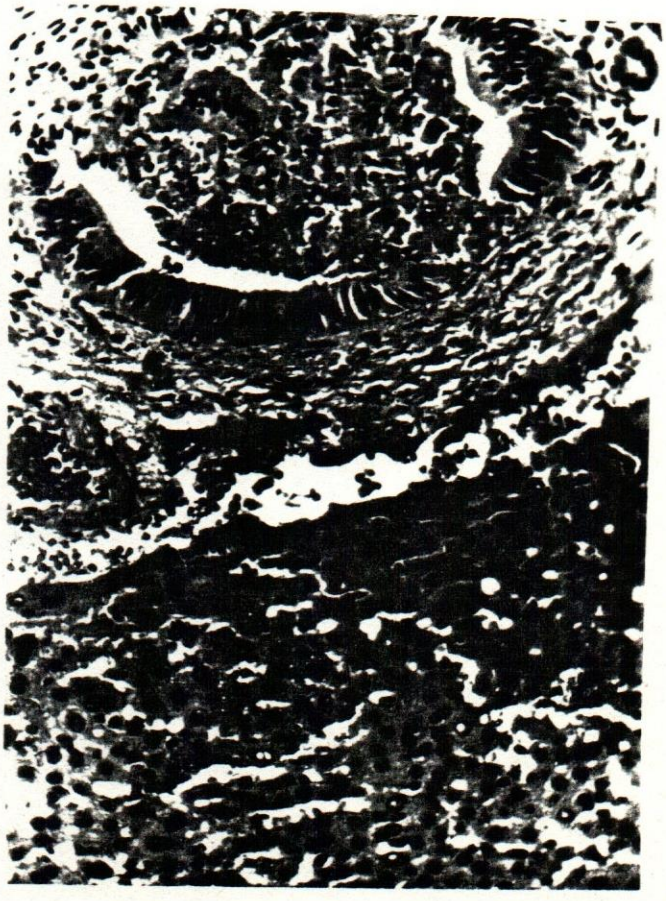

Fig. (15)