

قسم الباثولوجيا  
كلية الطب البيطرى - جامعة أسيوط  
رئيس القسم : د. د / محمد أبراهيم الشرى

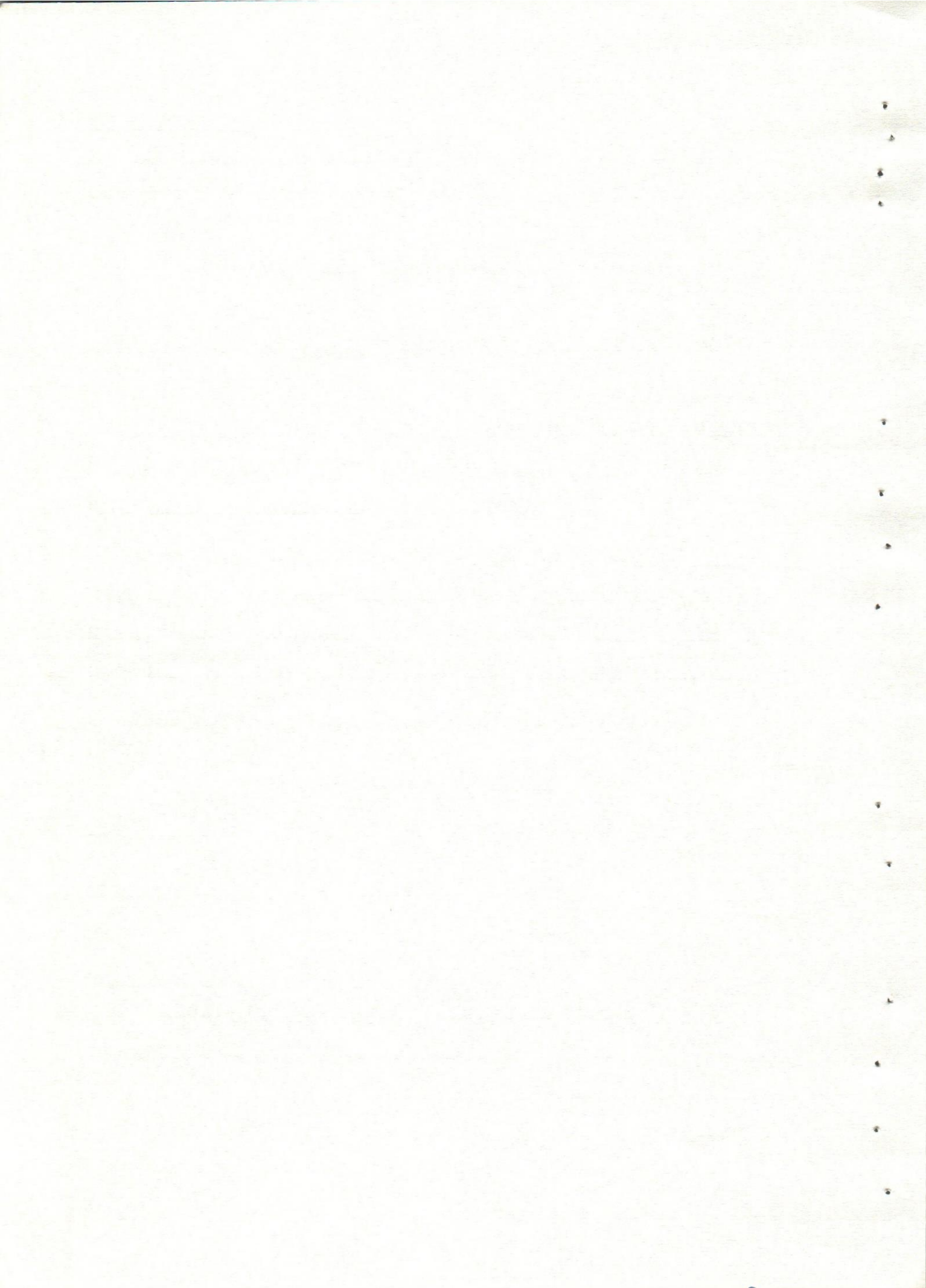
### دراسة على التهاب الحنجرة والقصبه الهوائية المعدى

عبد اللطيف بيومى ، صلاح موسى\*، أبراهيم سكر\*، عماد نافع\*، ناهد جاد\*

تم تشخيص صورته غير نموذجية صابة بمرض التهاب الحنجرة والقصبه الهوائية المعدى فى قطيع من الدجاج الفيومى عمر ١٤ اسبوع . وظهرت الاصابة اعراضا تنفسية وبلغت نسبة الاصابة ١٥ ٪ ونسبة النفوق ٥ ٪ .  
وتم عزل الفيروس المسبب للاصابه وتعريفه . وقد احدث الفيروس بقع كبيرة باهته على الغشاء المشيمى الالنتونى لاجنة الدجاج . كذلك فقد تم احداث العدوى الصناعية فى طيور قابله للاصابة وظهرت اعراضا تنفسية وعممل الصفه التشريحية لها ظهرت التهابات مخاطية صديديه فى القناة التنفسيه وكذلك اظهر الفحص المجهرى وجود الاجسام الضمنية داخل أنوية الخلايا المصابة .

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\* قسم أمراض الدواجن كلية الطب البيطرى - جامعة أسيوط  
\* \* قسم البترولوجيا كلية الطب - جامعة أسيوط



Dept. of Pathology,  
Faculty of Vet. Med., Assiut University,  
Head of Dept. Prof. Dr. M.I. El-Sherry.

**INVESTIGATIONS ON A MILD ATYPICAL INFECTIOUS LARYNGOTRACHEITIS  
IN UPPER EGYPT**  
(With 4 Figs.)

By  
**A.H. BAYOUMI; S.A. MOUSA\*; L.H. SOKKAR\*; E.K. NAFIE  
and N.A. GAD\***

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**SUMMARY**

An atypical, mild form of infectious laryngotracheitis (ILT) was diagnosed in a flock of 14 week-old, Fayoumi chickens at Assiut Province. Infection took the form of respiratory distress, with 15% morbidity and 5% mortality rate. The virus was isolated and identified. The virus induced large pale lesions on the chorioallantoic membrane of chicken embryos. The disease was reproduced in susceptible chickens. Typical signs of respiratory distress were observed. On postmortem examination mucopurulent inflammation of the respiratory tract in addition to typical intranuclear inclusions were observed microscopically.

**INTRODUCTION**

Infectious laryngotracheitis, an acute disease of chickens characterized by signs of respiratory depression, gasping and expectoration of bloody exudate, has been reported from many parts of the world, HOFSTAD (1978). Lesions of ILT have been reported chiefly in the upper respiratory tract and conjunctiva, JORDAN, (1966) and in lungs and air sacs, SNOCYENBOS and OLESIUK (1971). The disease has been recently reported in Egypt among imported flocks, TANTAWI *et al.* (1983). The present work reports naturally occurring ILT infection in Fayoumi chickens at Assiut Province and observations on the experimentally induced infection.

**MATERIAL and METHODS**

**Flock history:**

During JUNE, 1983, respiratory signs were observed in a flock of 20,000, 14 week-old Fayoumi chickens at Beni-Mor governmental farm. Samples for the present study were taken, when a disease condition, characterized by gasping, nasal discharge, coughing and moist rales, was noticed (Fig. 1). A morbidity rate of 15% and mortality of 5% were recorded within one month. Lesions of mucoid to haemorrhagic inflammation in the larynx and trachea were observed.

**Viruses:**

- a- A homogenized chorioallantoic membranes (CAM) of susceptible chicken embryos infected with an egg adapted vaccinal strains of ILT virus was used as a reference virus. The vaccinal strain was supplied by TAD .. pharmaceutical werk GMBH, Cuxhaven, west Germany.

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\* Dept. of Poultry Diseases, Faculty of Vet. Med., Assiut University.

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b- An isolate, designated 76/1983 was recovered from bacteria free suspension of larynx and trachea of dead, 14 week-old Fayoumi broilers with lesion suggestive to be ILT infection. The suspension was incubated in susceptible chicken embryos for 3 successive passages. Infected CAMS were homogenized, diluted 1:1 in normal saline, centrifuged and supernate was stored at 20 °C. The 50% embryo infective dose (E X D 50) was determined for both the reference virus and the isolate by the method of REED and MUNCH (1938).

**Hyperimmune serum:**

Antiserum against the vaccinal reference strain was prepared by intraocular instillation of 17- week-old white leghorn chickens, (with 10 EID 50, 14 days later). The birds were infected 4 times intramuscularly, (over a 2 week period). A virus dose of 10 EID 50 was used. Chickens were bled 12 days after the last injection and sera were collected and stored at 20 °C.

**Chicken embryos:**

10 to 11 day-old susceptible Fayoumi chicken embryos were inoculated by the dropped CAM method.

**Chickens:**

17 week-old white leghorn chickens were provided by the national poultry company at Assiut. All birds were checked for freedom of precipitating and neutralizing antibodies to LIT virus. They were used for preparation of antiserum and experimental infection.

**Agar-gel precipitin (AGP) test:**

Qualitatively, the microtechnique was carried out after NANSI (1958) to detect precipitins in sera of chickens and precipitinogen in infected CAMS.

**Virus neutralization (VN) test:**

VN test was carried out by the constant serum-virus dilution method in chicken embryos after CANNINGHAM (1966).

**Histopathology:**

Larynx, trachea and lungs of experimentally infected chickens were fixed in 10% neutral buffer formalin, embedded in paraffin, sectioned and stained with haematoxylin and eosin.

**Experimental infections:**

A total of thirty 17 week-old white leghorn chickens were divided into two groups (A & B), each of 15 birds. The group A was intratracheally infected with 10 EID 50 bird of the isolate 76/1983. Group B served as noninfected control. Each group was kept in separate unit under strict isolation. Birds were clinically observed for 10 days. Five birds from each group were subjected to autopsy at 3.5 and 10 days postinfection (P.i.). Larynx, trachea and lungs were subjected to histopathological examination. Egg inoculation was performed using pooled samples for virus reisolation.

**RESULTS**

Inoculation of susceptible chicken with the isolate 76/1983 revealed no deaths, but opaque thickened and elevated plaques with depressed central cores on the CAMS were noticed. Plaques were millet seed sized or even larger (Fig. 2).

Inoculation of the vaccinal reference strain resulted in stunting of inoculated embryos with formation of pinhead sized plaques on the CAMS (Fig. 3). The EID 50 for the isolate and the reference strain was 10 and 10 EID 50 10.1 ml respectively. Infected CAMS with 76/1983 isolate

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reacted positivity in AGP test against reference hyperimmune serum. Both the isolate and the vaccinal strain were neutralized by the hyperimmune serum with neutralization index of 3.1 and 3.6 log 10 respectively.

Results of experimental infection:

10 birds out of 15 of group A which were infected with the isolate 76/1983 showed marked depression and serous nasal discharge at the 2<sup>nd</sup> day P.i. Four birds out of 10 showed gasping and coughing 4 days P.i. Signs of illness disappeared at 8 days P.i. except in two birds which showed gasping, coughing and moist rales. Birds showed moderate congestion of the nasal, laryngeal and tracheo-broncheal mucosa on the third day P.i. Oedema and congestion of the conjunctiva and infraorbital sinuses were inconstant finding. On the fifth day P.i., the upper air passages including the laryngeal and tracheal laminae were in completely occluded by thickened, turbid, yellowish gray mucopurulent exudate. In two cases the exudate was admixed with blood and appeared dirty pink. At ten days P.i., mild mucopurulent exudations were noticed in four cases, while one case showed thick, tenacious and diphtheroid plugs in the larynx and trachea. Removal of these plugs left an eroded and reddened underlying surface. Histopathological alterations on the third day P.i. were manifested by epithelial cell swelling, nuclear chromatin margination and cytoplasmic proteinous dystrophic changes. Intranuclear inclusions were detected in all cases which were located in groups of cells (Fig. 4). The mucosa and submucosa showed hyperaemia, oedema and mononuclear cell infiltrations. Five days P.i., epithelial necrosis and desquamation were constant findings. Heavy mucosal and submucosal lymphocytic, histocytic and plasma cell infiltrations were evident. Intranuclear inclusions could be detected only in one case. At ten days P.i., the epithelial destructive alterations could be detected only in one case.

Virus was reisolated from larynx and trachea of all sacrificed at 3, 5 and 10 days P.i. Birds of the non infected group B showed no signs of illness or post mortum lesions and were free from any histopathological alterations.

### DISCUSSION

An ILT virus isolate was recovered from 14 week-old Fayoumi flock showing respiratory symptoms. Fifteen percent morbidity rate and five percent mortality rate were observed. Typical signs of coughing with expulsion of blood described by DELAPHANE (1945) could not be noticed. Mild forms of the disease were reported by RAGGI and ARMSTRONG (1960).

The isolate caused no death of inoculated embryos, but opaque millet seed sized plaques on the CAMS were noticed. Plaques appeared larger in diameter and size in comparison with those caused by the vaccinal virus strain. SRINIVASAN and MALLICK (1977) concluded that virulent strains produced larger pock lesions, while KRALI *et al.* (1977) reported that the changes induced in chick embryos were indistinguishable in less virulent strains from those produced by velogenic viruses.

The disease was successfully reproduced in susceptible chickens with incubation period of two days. Infected birds showed respiratory distress with congestion of nasal, laryngeal and tracheo-bronchial mucosae. Mucopurulent exudate was noticed occluding the upper respiratory passages. Intranuclear inclusions could be detected in all cases slaughtered three days post inoculations, and in one case out of the five birds slaughtered at the 5<sup>th</sup> day post inoculation. Similar findings were reported by WATRACH *et al.* (1959) and REYNOLDS *et al.* (1968).

Both the isolate and the reference virus strain were neutralized by the hyperimmune serum with neutralization index of 3.1 and 3.6 10 g respectively. Distinct immunologic strain have not been reported, although some variations in neutralizing abilities have been demonstrated between strains of varying pathogenicity, PULSFORD (1954).

In the present study pulmonary and air sac lesions reported by SNOEYENBOS and OLESIUK (1971) could not be detected, probably attributed to variations in the velogeneity of the causative viral agent. Although the present investigation had shed some light upon the presence of a mild, atypical ILT infection in upper Egypt. Such problem necesiate further screening on the epidemiology of the disease in Egypt.

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### DESCRIPTION OF FIGURES

- Fig. (1): Chickens showing respiratory signs.
- Fig. (2): Infected CAM showign large plaques.
- Fig. (3): Infected CAM showing small plaques.
- Fig. (4): Tracheal epithelium showing hyperplastic changes and intranuclear inclusions.

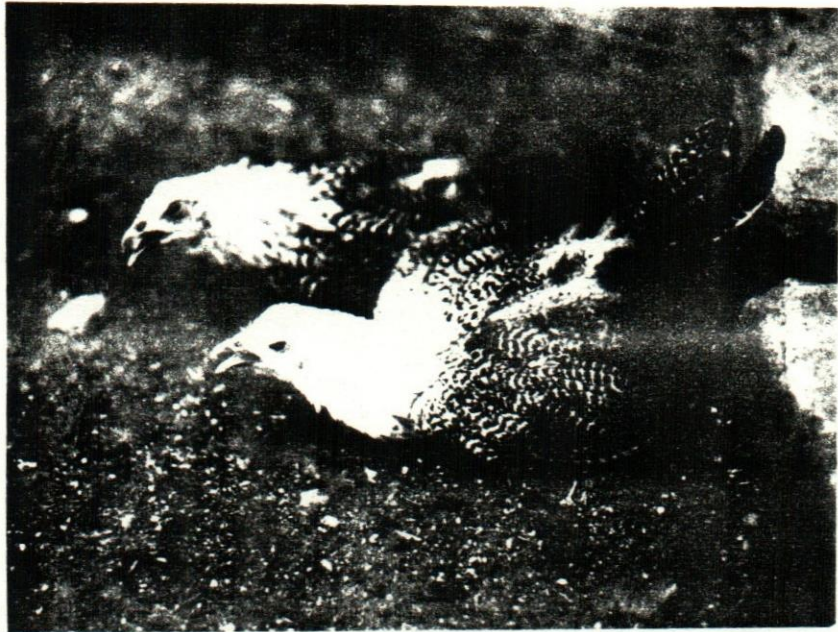


Fig. (1)

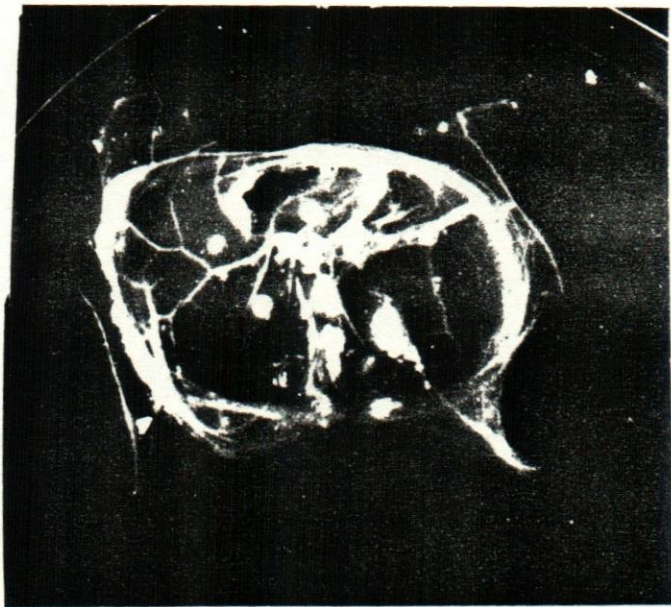
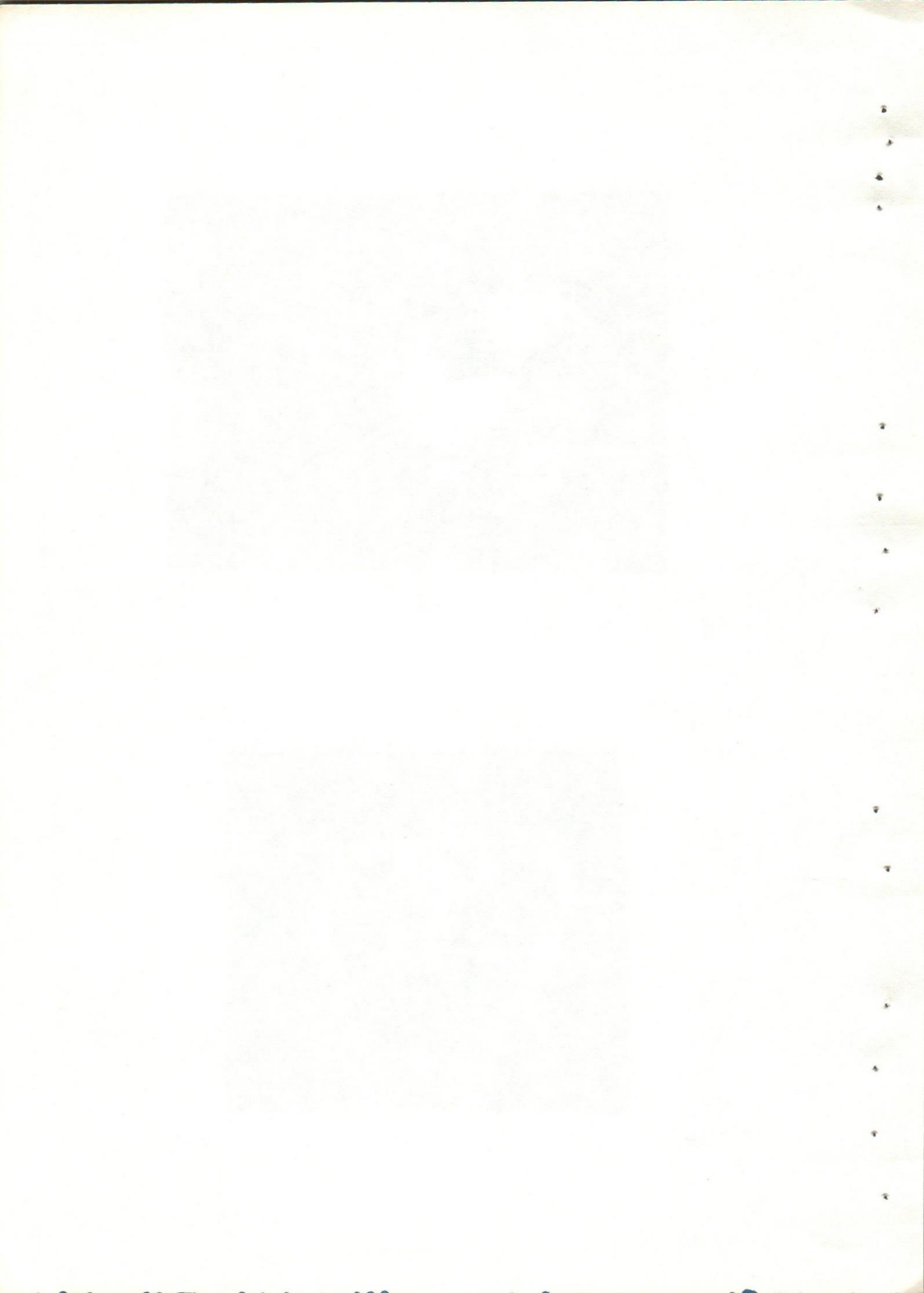


Fig. (2)





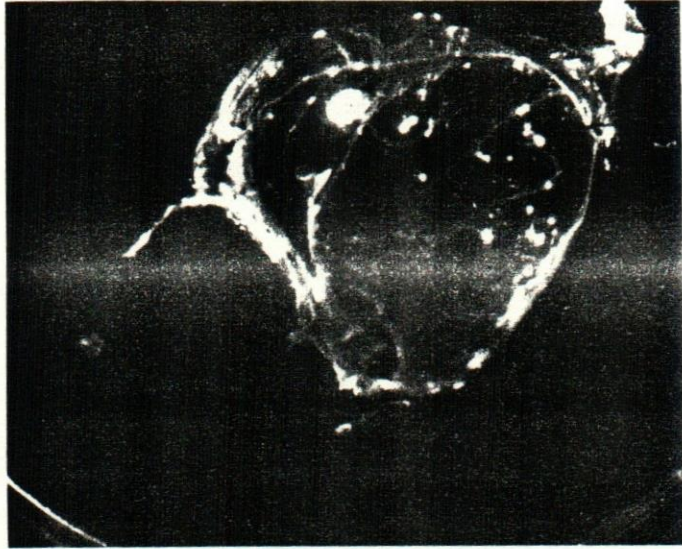


Fig. (3)

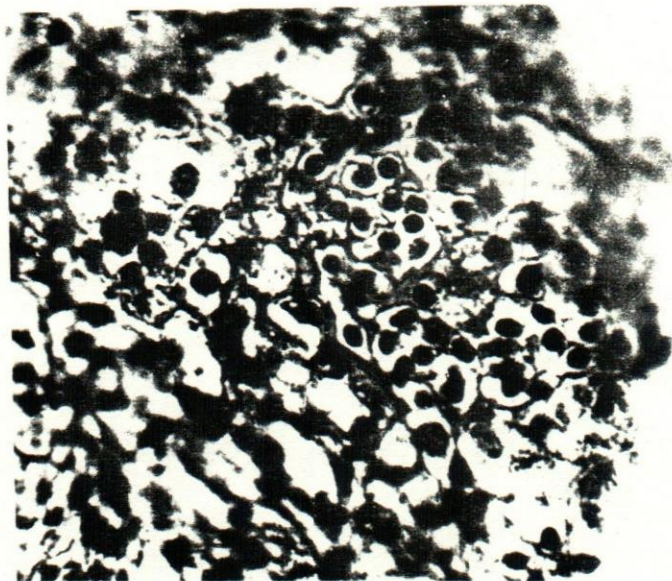


Fig. (4)



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