Animal Health Research Institut,
Assiut. Regional Laboratory.

MORPHOLOGICAL AND AETIOLOGICAL STUDIES ON PNEUMONIA IN TURKEY: ORNITHOBACTERIUM RHINOTRACHEALE INFECTION

(With 8 Figures)

By

*SANAA A. EL-SHAMY; SOHAIR Z. HUSSEIN and A.H. BAYOUMI**

*: Dept. of Pathology, Fac. Vet. Med., Assiut University

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دراسات مورفولوجية على الالتهاب الرئوي في الرومي: الإصابة بميكروب الاورنيثوباكتريم رينوتراكيالى

سناء عبده الشامي ، سهير زين العابدين حسين ، عبد اللطيف حسن بيومي

ظهرت أعراض تنفسية شديدة في قطيع من الرومي في عمر ١٤ أسبوعا. تميزت هذه الأعراض إكلينيكيا بوجود ضعف عام وسائل رشحي من الأنف وكحة وصعوبة في التنفس وكانت نسبة الإصابة ٢٥% بينما وصلت نسبة الوفيات إلى ٩%. و لقد كانت العلامات المميزة عند التشريح احتقان في الجهاز التنفسي والتهاب فبريني هيستروفيلي في الرئة والأكياس الهوائية. وقد أمكن عن طريق الفحوص البيوكميائية والسيرولوجية عزل الميكروب المسبب لهذا المرض وهو الاورنيثوباكتريم رينوتراكيالى. وقد تم استبعاد بعض مسببات أمراض الجهاز التنفسي الأخرى.

SUMMARY

A turkey flock of 14 weeks of age suffered from a severe respiratory affection. Clinically it characterized by signs of depression, nasal discharge, coughing, dyspnea and gasping. The morbidity rate was 25% and the mortality rate reached 9%. On postmortem examination congestion of the upper respiratory tract, areas of lung consolidation and airsacculitis were the most characteristic lesions. The reaction was

mostly acute fibrinoheterophilic in nature. Pasteurella like organisms could be seen microscopically. The organism was identified biochemically and serologically as Ornithobacterim rhinotracheale. Other aetiologic agents for respiratory infections could be ruled out.

Key words: Pneumonia, turkey, ornithobacterium rhinotracheale.

INTRODUCTION

As in chickens, respiratory problems were also observed in turkey flocks (Joubert et al., 1999). Hafez and Sting (1999) reported that the respiratory disease condition are one of the most serious groups of diseases. Many aetiological agents were incorporated (Charlton et al., 1993; Hafez et al., 1993). Since 1994, Ornithobacterium rhinotracheale has been identified and described as one of aetiological agents of respiratory diseases in turkey (Vandamme et al., 1994; Van Beek 1994). Several strains were isolated from chickens and turkeys in many countries (Van Empel and Hafez, 1999; Hafez and Sting 1999). In the available literature, nothing was recorded about isolation or infection with Ornithobacterium rhinotracheale in Egypt.

The disease causes heavy losses through, respiratory failure, increased condamenation rate, and increased medication costs (Charlton et al., 1993, Hafez, 1994; Van Empel and Hafez, 1999 and Joubert et al., 1999). On postmortem examination, De Rosa et al. (1996) and Stephanie et al. (1998) described unilateral or bilateral lung consolidation and fibrinous serositis. Histopathologically fibrinous pneumonitis with heterophilic cellular infiltration in the parabronchi and air capillaries were described by De Rosa et al., (1996) and Stephanie et al., (1998). De Rosa et al., (1996) added hepatopathic alterations. The latter included individualization, coagulative necrosis of the hepatic parenchyma and vascular thrombosis. Joubert et al., (1999) noticed grossly whitish nodules ranged from 1-3 mm in diameter. Van Empel et al., (1999) observed pneumonic lungs only in mixed infections with Newcastle disease virus in his experimental study. Stephanie et al., (1998) reported the presence of small numbers of gram-negative bacteria in the exudative pneumonia in experimental reproduction of the disease. The author noticed microscopically the presence of multiple foci containing necrotic cellular material, macrophage and multinucleated giant cells.

The aim of the present investigation is to report and describe an outbreak of Ornithobacterium rhinotracheale infection in turkey flock at El-Minia Governorate, Egypt.

MATERIAL and METHODS

Twenty dead turkeys and ten diseased ones of 14 weeks age were obtained from El Minia Governorate. The living birds were examined carefully and the clinical symptoms were recorded. The birds were slaughtered and postmortem examination was conducted for the dead and slaughtered birds. The gross pathological alterations were described and samples from heart blood, lungs and livers were taken for virological, bacteriological, serological and fungal examinations (Cruickshank et al., 1980). Tissues were plated on 5% sheep blood agar and MacConkey's agar and incubated at 37°C in 7.5% Co2 for 48 hours. Swabs of trachea were plated on modified Frey's agar and inoculated into Frey's broth for mycoplasmal isolation attempts, Frey et al., (1968). In addition samples of lung from birds were cultured on sabouraud dextrose agar and incubated at 37°C for 24 hours followed by incubation at room temperature for 4 weeks for fungal isolation attempts. For virus isolation, pooled samples of lungs were homogenized, filtered and 0.1ml was inoculated into the chorioallantoic sac of 11-days-old embryonated eggs of chickens. The embryos were inspected at 5 days of incubation and hemoaglutionation test was conducted on the allantoic fluid.

For serological examination serum samples from living turkeys were tested for antibodies to Newcastle disease virus by hemoaglutination test and to avian influenza by agar gel precipitin test. Liver and Lung tissue specimens were prepared for histopathological examination. Tissue sections were stained by Harris's Haematoxylin and eosin, periodic acid schiff. Giemsa stain, Gram stain and Gridley's stain (Bancroft and Stevens, 1982).

RESULTS

Aetiological findings:

No mycoplasma or fungi could be recovered from any of the examined tissues. On sheep blood agar pin point small colonies were seen after 24 hours incubation. After 48 and 72 hrs the colonies were larger and convex. No colonies could be seen on MacConkey's agar.

Gram stained slides showed Gram-negative pleomorphic pasteurella like organisms, biochemically, the organism was catalase negative, oxidase positive, gelatin negative, lactose positive, inositol negative, urease positive and indol negative.

The inoculated embryos had no gross changes and no viruses could be detected by hemoaglutination of allantoic fluid.

All the taken serum samples were negative for antibodies to avian influenza and Newcastle disease virus.

Clinical symptoms:

All the living birds showed unthriftiness. emaciation weakness, reduced body weight and showed respiratory manifestations. The latter included nasal and occular watery discharge, Coughing, gasping, mucous expectoration, moist rales and dyspnea. The morbidity rate reached 25% and the mortality rate was up to 9% in the flock as was reported by the veterinarians.

Macroscopic finding:

On dissection of both dead and slaughtered birds congestion of the upper respiratory passages was consistent finding. The lungs of all birds were congested, heavy oedematous and in 70% of the cases areas of consolidation were noticed. The consolidated areas were either unilaterally or bilaterally located. In 30% of these pneumonic lungs greyish white nodules were seen. The nodules varied in size from pin head to 3 mm in diameter. The air sacs were turbid, thickened and in some birds showed fibrinous like deposits. No gross changes could be seen in the viscera.

Microscopic findings:

Examination of the lungs revealed varying degrees of acute fibrino heterophilic exudative pneumonitis. The lungs were extremely hyperemic, the alveoli were nearly filled with fibrin threads, heterophils and few numbers of mononuclear cells, lymphocytes and macrophage. In areas with large amounts of fibrin, few numbers of heterophils were present and vice versa. Most of heterphils were degranulated and many of them showed nuclear chromatorrhexis. Similar exudate was present also in the parabronchi. The fibrinoheterphilic exudate was present also in all air ways including bronchi atria, air capillaries and between air capillaries and atria. In the air ways the degranulated and necrosed heterophils were intermingled with the fibrin threads Fig 1,2. In three cases large randomly distributed areas of necrosis were seen, Fig. 3. The

center of which was sequestrated and separated from the surrounding lung parenchyma. The heterophils showed degranulation and nuclear chromatorhexis. The center showed the acidophilic granular cellular debris. In two cases liquifactive necrosis of lung parenchyma involving the smaller air ways could be seen Fig. 4. In giemsa stained sections pasteurella like organisms were seen in the fibrinous exudate of the air ways. Either relatively large branched chains Fig. 5a or scattered singly Fig. 5b. In all the examined sections the vascular ramfications were surrounded by inflammatory exudate, Fig. 6. The larger blood vessels are frequantly distented and showed also severe vacuolation of the smooth muscle cells of the tunica media Fig. 7. The examined livers showed diffuse mild proteinous and fatty dystrophic changes. In addition clusters of the bacteria could be seen, Fig. 8. The air sacs showed the same inflammatory reaction seen in the lung sections.

DISCUSSION

Ornithobacterium rhinotracheale has only recently recognized as a respiratory pathogen in chickens and turkeys in many countries (charlton et al., 1993; Hafez et al., 1993; Hafz, 1994; Hinz et al., 1994; Van Beek, 1994, Murray et al., 1995 and De Rosa, 1996). It was considered as a secondary respiratory pathogen but Stephanie et al., (1998) reported the first successful experimental production of the disease syndrome associated with Ornithobacterium rhinotracheale in turkeys. Charlton et al., (1993), Hafez et al., (1993) and Hinz et al., (1994) had fulfilled the three postulates of koch. In the avialable literature neither isolation of the organism or diagnosis of Ornithobacterium rhinotracheale infection in turkey was recorded in Egypt. This report record and describe the infection in turkeys for the first time.

In the present investigation Ornithobacterium rhinotracheale could be isolated and identified from turkeys with respiratory symptoms. Morphological and cultural characteristics of the organism were similar to that described by De Rosa et al., (1996); Stephanie et al., (1998) and Chin and Droual (1997). The organism could be parely isolated and cultured as it was conducted by Stephanie et al., (1998), while De Rosa et al., (1996) could not isolate or reproduce the disease.

Clinically turkeys showed respiratory distress. In the literature variable degrees of respiratory signs were reported (Hafez et al., 1993; Dudouyt et al., 1995, Odor et al., 1997 and Joubert et al., 1999).

Severity of clinical symptoms and subsequently the mortalities may be influenced by bad management inadequate ventilation, density, poor hygiene and high ammonia levels, Van Empel and Hafez (1999). The turkey flock showed mortality rate reached 9%. This is inaccordance with that reported by Hafez and sting (1996) and Chin and Droual (1997) who said that it ranges from 2 to 11% in chicken and turkey.

On post mortem examination congestion of the upper respiratory tract, unilateral or bilateral lung consolidation with the presence of greyish white nodules and fibrin deposition of air sacs were the characteristic findings. Similar lesions were described in natural and experimental Ornithobacterium rhinotracheale infections (Opengart et al., 1995; De Rosa et al., 1996; Stephanie et al., 1998; Joubert et al., 1999 and Hafez and Sting, 1999). Although, Back et al., (1998) and Nagaraja et al., (1998) postulated the vertical transmission of infection however no lesions could be seen in the examined ovaries and oviducts. This type of transmission was explained by Van Emple et al., (1997) as cloacal contamination of eggs.

On microscopic examination of the lung tissues, fibrinoheterophilic pneumoma with necrosis, only in 10% of examined cases was noticed. The severity of lesions and the relative high mortality rate are probably due to that these turkeys had no previous exposure to Ornithobacterium rhinotracheale during their growing period. In addition these severe lesions are similar to the experimental studies carried out by Stephanie et al., (1998); De Rosa et al., (1996) and Ryll et al., (1996), which have been completed using the intratracheal route of inoculation using young ages of turkeys.

In contrast to the results obtained by Roepke et al., (1998) and Sprenger et al., (1998), neither enteritis nor arthritis could be seen in this study. This could be probably related to the strain of Ornithobacterium rhinotracheale and duration course of the disease.

Because of the resemblance of the lesions obtained with those reported in pasteurellosis. The differential diagnosis in this study was based on the absence of vascular thrombosis, presence of necrotic foci only in 10% of examined cases, isolation and identification of Ornithobacterium rhinotracheale. In addition many other actiologic

agents for respiratory manifestations such as Mycoplasma, Chlamydia, Newcastle disease virus, Avian influenza virus and Aspergillosis were ruled out.

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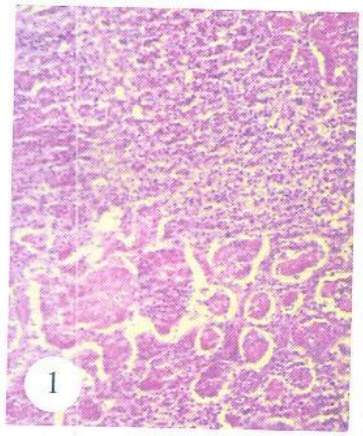
Fig. 8: Liver showing necrobiotic changes and clusters of bacteria.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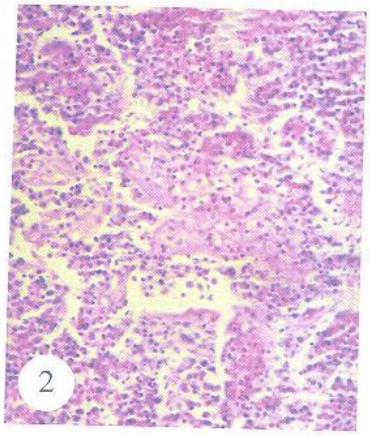
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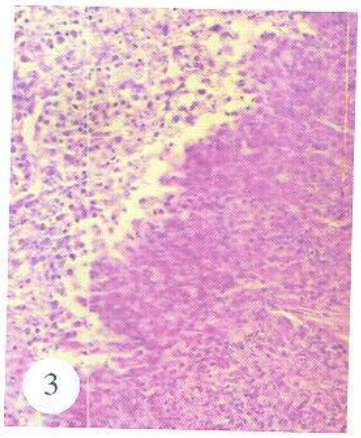
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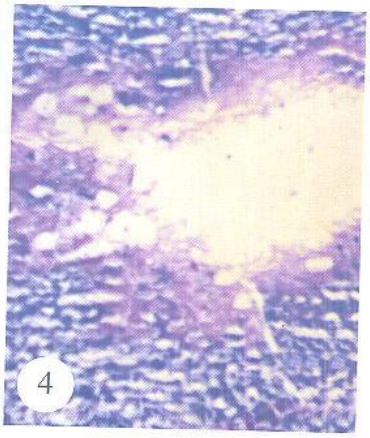
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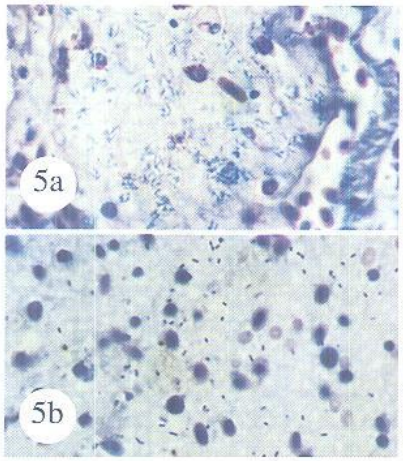
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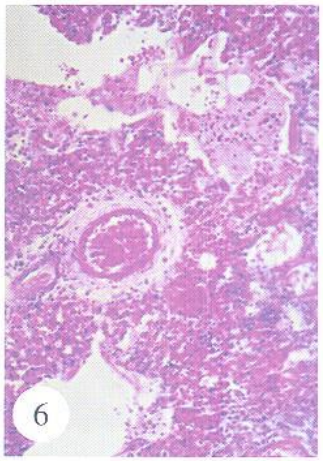
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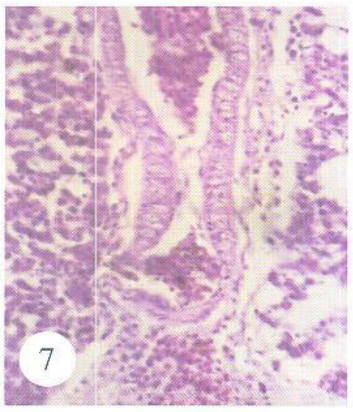
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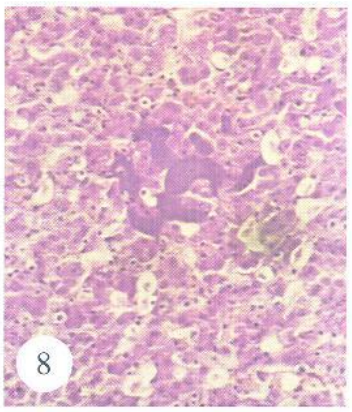
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