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# BRONCHOPNEUMONIA IN BUFFALO-CALVES IN ASSIUT GOVERNORATE I- STUDIES ON BACTERIAL CAUSES, CLINICAL, HAEMATOLOGICAL AND BIOCHEMICAL CHANGES ASSOCIATED WITH THE DISEASE

(With 8 Tables)

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## الإلتهاب الشعبي الرئوي في العجول الجاموسي في محافظة أسيوط ١ــ دراسات عن الأسباب البكتيرية والتغيرات الإكلينكية والدموية والبيوكيميائية المصاحبة للمرض

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يعتبر الإلتهاب الشعبي الرئوي واحداً من الأمراض *التي* تصيب قطعان الأبقار المختلفة خاصة العجول النامية *التي* تتعرض للعوامل الضاغطة وظروف الإيواء السيئة . استهدفت هذه الدراسة إلقاء الضوء على أهم العوامل الممهدة لحدوث المرض ومعرفة الصورة الأكلينيكية والأسباب البكتيرية الحقيقية له وكذلك دراسة التغيرات الدموية والبيوكيميائية المصاحبة للمرض. أجريت الدراسة على عدد ١٢٦ حيوان من العجول الذكور والإناث الجاموسي *التي* تتراوح أعمارها بين ٣ــ٩ شهور ، ٩٠ منهم كانت تعانى من مرض الإلتهاب الشعبي الرئوي بينما أثبتت الفحوصات الإكلينيكية والمعملية أن باقي الحيوانات وعددها ٣٦ رأساً كانت سليمة صحياً *وفي* حالة جيدة واستخدموا كضوابط للبحث . أظهر الفحص الإكلينيكي أن الحيوانات المريضة كانت تعانى من صعوبة *في* التنفس مصحوباً بكحة طرية وإفرازات أنفية تراوح قوامها *في* الحيوانات المختلفة بين المخاطية والمخاطية الصديدية . هذا بجانب فقدان الشهية والهزال والضعف العام . لوحظ أيضا وجود أصوات

تنفسية غير طبيعية عند فحص الرئتين، وبلغت نسبة النفوق بين الحيوانات المريضة ٦,٧% . أظهرت نتائج الفحص البكتيري إصابة بعض الحالات المريضة ٧٤,٧% بنوع واحد من البكتريا (عدوى فردية) ، بينما تم عزل أكثر من نوع من البكتريا في ١٩,٣% من الحالات (عدوى مختلطة) . تمثلت العدوى الفردية في عزل أجناس الميكروب القولوني (*E.coli*) في ٢٣,٠٨% والباستريلا مالتوسيدا (*P.multocida*) في ٢١,٥٤% ، والميكروب السبحي العنقودى (*Staph aureus*) في ١٥,٣٨% بالإضافة إلى بعض البكتريا الأخرى بنسب مختلفة . أما العدوى البكتيرية المختلطة فتمثلت في عزل الميكروب القولونى (*E.coli*) مع الباستريلا مالتوسيدا (*P.multocida*) في ٣٨,٨٨% والميكروب السبحى العنقودى (*Staph aureus*) مع الميكروب القولونى (*E.coli*) في ٢٢,٢% وبعض البكتريا المختلطة الأخرى بنسب مختلفة . تم عزل الميكروب القولوني (*E.coli*) والباستريلا مالتوسيدا (*P.multocida*) والميكروب السبحي العنقودي (*Staph aureus*) من البيئة المحيطة بالعجول (الأرضيات والهواء ومياه الشرب) . وقد تبين أن أرضيات المزارع التي تربى بها هذه العجول والمياه المستخدمة لسقى هذه الحيوانات هى أهم المصادر لإنتشار المرض. أوضحت نتائج تحليل الدم أن الحيوانات المريضة تعانى أنيميا شديدة تمثلت في انخفاض معنوى في قيم الهيموجلوبين (Hb) وحجم خلايا الدم المنضغطة (PCV) وكذلك العدد الكلى لكرات الدم الحمراء (RBCs)، بينما كان هناك ارتفاع معنوي في العدد الكلى لخلايا الدم البيضاء (WBCs) . أظهرت نتائج التحاليل البيوكيمائية لمصل الدم وجود انخفاض معنوى في مستوى البروتين الكلى والزلال (الألبيومين) مع وجود زيادة معنوية في مستوى الجلوبيولين . هذا وقد أظهرت التحاليل البيوكيمائية أن الالتهاب الشعبي الرئوي يؤثر على وظائف بعض الأعضاء الأخرى كالكبد والكلى ، والذي قد يعزى إلى انتقال الميكروبات من الرئة عن طريق الدم إلى هذه الأعضاء وإحداث بعض الآثار الضارة بها . اتضح هذا التأثير من وجود زيادة واضحة في مستويات بعض إنزيمات مصل الدم مثل إنزيم (ALT) وأنزيم (AST) وأنزيم (AP) وكذلك إرتفاع في مستوى البولينا والكرياتينين بالدم في بعض الحالات المريضة مما أدى إلى حدوث زيادة معنوية في قيم متوسطات هذه العناصر.

## SUMMARY

Bronchopneumonia affects different animal herds specially growing calves exposed to stressors and bad hygienic conditions. The present study aimed to throw a light on the important clinical signs and bacterial causes of bronchopneumonia among buffalo-calves and to study also the haematological and serum biochemical changes associating the disease. The study includes 126 buffalo-calves of both sexes, 3-9 months old. Calves representative to farms in the different villages in adjacent to Assiut city, Assiut, Egypt. On the basis of clinical and laboratory examinations buffalo-calves were classified into two groups. 90 calves showed the clinical signs of bronchopneumonia (diseased group) and 36

calves were healthy and served as control. Severe dyspnoea, moist cough, harried respiration, cyanosis of the mucous membranes , anorexia, depression, mucoid to mucopurulant nasal discharge and abnormal lung sounds were the evident signs observed on pneumonic calves. Some complicated cases were died. Bacteriological isolates indicated that *E.coli, Pasteurella multocida, Staph aureus* and other bacteria were the common bacteria from nasal passage as a single infection in 74.7% of the examined diseased calves, however, *E.coli* with *Pasteurella multocida, Staph aureus* with *E.coli* and other bacteria were isolated as mixed infection in 19.3% of diseased cases. *E.coli, Pasteurella multocida, Staph aureus* and *Strept pyogenes* were isolated also from the surrounding environment (Soil, air and drinking water). All isolates of *Pasterulla multocida* and *Haemophilus somnus* were highly virulent to mice within 3-6 days after intraperitonial injection with $7.5 \times 10^6$ viable organisms. Haematological picture of the diseased calves revealed significant decrease in Hb concentration, PCV and RBCs count associated with leucocytosis . Serum biochemical analysis revealed significant decrease in both total protein and albumin levels with significant increase in the level of globulin. Elevation in the serum levels of urea, creatinine , AST, ALT and AP were found in some diseased cases which may be attributed to renal ischaemia and to the minute scattered abscesses observed in the kidneys and livers of some died cases. This elevation was reflected by a significant increase in the mean values of these parameters.

***Key Words:*** *Bronchopneumonia, Buffalo-claves.*

## INTRODUCTION

Respiratory affections particularly bronchopneumonia are major problems among calves, causing severe economic losses through reduction of weight gain, high morbidity and mortality rates (El-Sebaie *et al.*, 1984 & 1987; Abd El-Ghani *et al.*, 1990; Youssef *et al.*, 1992 and Barrett, 1998).

These affections are a complex syndrome attributed to several factors involving stressors, viral infection and bacterial infection . Regarding the most common basic causes of respiratory troubles a lot of works had been done. Pasteurella spp., Corynebacterium pyogenes as well as Staph. aureus, Streptococci and E.coli are claimed to be the main bacterial causes responsible for pneumonia in calves (Al-Allawy *et al.*,

1979; Elyas, 1982 and Umlauft *et al.*, 1987). E.coli, Klebsiella pneumoniae, Salmonella spp. and pasteurella spp. are also recorded to be the bacteria responsible for pneumonia in calves (Mosier, 1997).

There were empirical and some experimental evidences suggesting that environmental conditions specially cold, damp weather, dust, ammonia, over corwding, poor ventilation as well as poor hygienic measures may influence the occurrence and severity of respiratory diseases in calves (Bryson *et al.*, 1978; Bickert and Herdt, 1985 and Woldehiwet *et al.*, 1990).

An increase in the levels of blood serum glucose and total protein in association to bronchopneumonia in calves was recorded by (El-Sheikh *et al.*, 1994 and Saleh and El-Bably, 1998), however a reduction in the levels of these parameters were recorded by (Al-Allawy *et al.*, 1979 and El-Sherbini *et al.*, 1996). Elevation in the blood serum levels of urea, creatinine, alkaline phosphatase (AP), aspartate amino transferase (AST), and alanine amino tranferase (ALT), were reported by (Abdou *et al.*, 1989; Youssef *et al.*,1992; El-Sherbini *et al.*,1996 and Saleh and El-Bably, 1998).

This study aimed to: Throw a light on the most important clinical, haematological and some biochemical changes associated with bronchopneumonia among buffalo-calves in Assiut Governorate, Egypt.

Isolation and identification of the different bacteria that may be responsible for the disease.

## MATERIAL and METHODS

### I. Animals:

A total number of 126 buffalo-calves, 3-9 months old of both sexes were involved in this investigation. The calves were belonged to individual farms in different villages in adjacent to Assiut city, Assiut, Egypt. These animals reared in unhygienic conditions, the sTables were neglected, partially roofed and the floors were muddy, continuously damped and heavily soiled with manure particularly during winter months. 90 calves were diseased and showing signs of bronchopneumonia however the other 36 animals were healthy and used as control.

Careful clinical examination of both diseased and healthy calves were carried out according to Rosenberger *et al.* (1979) and Radostits *et al.* (1995). All examined calves were free from either external or internal parasites.

**II. Samples:**

**Nasal swabs:** Sterile swabs for bacterial samples were collected from the nostrils of both healthy and diseased calves, the cotton swabs were pushed as far as possible into one nostril then transferred to sterile nutrient broth in tubes (Woldehiwet *et al.*, 1990). The isolation, purification and identification of the bacterial isolates were carried out according to Carter (1984) and Baily and Scott (1990).

**Blood samples:** Two blood samples were collected from both control and diseased buffalo-calves, the first was taken on anticoagulant and used for the determination of total erythrocytes ($RBC_S$ T/1), leucocytic counts ($WBC_S$ G/l) and haematocrit value (PCV%) according to Coles (1986).

The second blood samples were taken without anticoagulant for obtaining blood sera for determination of total protein (g/*l*), albumin (g/*l*), urea (g/dl), creatinine (g/dl), glucose (mg/dl), AST. (U/1), ALT. (U/1) and AP. (U/1). These parameters were estimated sepectrophotometrically by means of test kits supplied by Boehringer Mannheim Diagnostica-Germany.

**Soil samples,** were taken from each examined animal farm for the isolation and identification of the bacteria that my be existed and affecting the animal health. Approximately 100 gm of soil were taken from different sites of the stalls of the examined farms and putted into 1000 ml sterile broth and incubated at 37°C overnight. Isolation and identification of pathogenic micro-organisms in the media were carried out according to Clegg *et al.* (1983).

**Water samples:** In sterile capped bottle approximately 10 ml of water were collected from water troughs utilized for drinking of buffalo-calves and examined for bacterial contamination according to Clegg *et al.* (1983).

**Air samples,** were collected from different sites of calf stalls by using settle plate method (Cruickshank *et al.*, 1965). Isolation and identification of the bacteria from air samples were carried out according to Jones (1987).

**Laboratory animal inoculation:** A total number of 125 white mice weighting 25-30 gms were used to investigate the pathogenicity of isolated *Pasteurella multocida* and *Haemophlius somnous*, five mice were used for each isolates according to Carter (1984).

**Post mortum findings of died cases:** Post-mortum examinations were carried out for 3 of 6 died buffalo-calves and abnormal gross lesions were recorded.

**Statistical analysis:**

The obtained data were statistically analyzed by means of Software Computer Statistical Program (Spsswin, 1995).

## RESULTS

Table 1: Clinical Signs observed on examined buffalo-calves.

| Groups | No. of Calves | Main clinical signs | Post-mortum findings |
|---|---|---|---|
| Diseased Buffalo-claves | 90 | - Inspiratory dyspnea (difficult inspiration, nostrils are widely distended and mouth was opened, extension of the head and neck and abduction of the fore limbs) painfull moist cough, mucoid to mucopulent nasal discharge.<br>- Congested and cyanosed mucous membranes<br>- On auscultation, abnormal chest sounds were heard at different areas of the lungs (harsh and exagerated vesicular sounds, crackles or moist rales). Muffled sound as well as cripitant rales were evident in advanced cases.<br>- Various degree of anorexia with progressive emaciation and lately recumbancy occurs<br>- Some complicated cases were died and the mortality rate reached 6.7%. | - The mucous membranes of the upper respiratory tract particularly nose were congested, haemo-rrhagic and covered with mucopurulant exudate.<br>- Presence of serofibrinous or purulent exudate in the bronchiole, lobular con-gestion or hepatiza-tion .<br>- Some cases show gelatin-ous exudate in the inter-lobular septa, pleurcy with shreds of fibrin present between the lobule.<br>-Bronchial and mediastin-al lymph nodes were swollen, oedematous and congested.<br>- Scattered petichae on serosal surface of the heart and mediastinum.<br>- Presence of minute scattered abscesses in the kidneys and liver tissues of some died cases. |
| Clinical Healthy Control | 36 | - Clinically healthy. No special findings. | — |

**Table 2:** Percentage of positive cases that exhibiting bacterial isolates of both healthy and diseased buffalo-calves.

| Animal | No. of Examined cases | No. of positive cases | % |
|---|---|---|---|
| Healthy calves | 36 | 5 | 13.9 |
| Diseased calves | 90 | 78 | 86.7 |
| **Total** | **126** | **83** | **65.87** |

**Table 3:** Incidence of single and mixed infection with bacteria in both healthy and diseased buffalo-calves

| Condition of buffalo calves | Single infection cases | | Mixed infection cases** | | Total No. of +ve cases |
|---|---|---|---|---|---|
| | No. | %* | No. | %* | |
| Healthy calves (36) | 3 | 3.6 | 2 | 2.4 | 5 |
| Diseased calves (90) | 62 | 74.7 | 16 | 19.30 | 78 |
| **Total** | **65** | **78.3** | **18** | **21.7** | **83** |

** The percentage was calculated in relation to the total number of the +ve cases (83).*
*** Mixed infection with two micro-organisms*

**Table 4:** Percentage of various micro-organisms isolated as single infection of both healthy and diseased buffalo-calves.

| Bacterial species isolated single form | Examined buffalo-calves | | | |
|---|---|---|---|---|
| | Healthy buffalo-calves | | Diseased buffalo-calves | |
| | No. of isolates | % | No. of Isolates | % |
| E.coli | 1 | 1.54 | 15 | 23.08 |
| P. multocida | 1 | 1.54 | 14 | 21.54 |
| Staph aureus | 1 | 1.54 | 10 | 15.38 |
| Strept pyogenes | 0 | 0 | 8 | 12.31 |
| Haemophilus somnus | 0 | 0 | 6 | 9.23 |
| Coryne bacterium ovis | 0 | 0 | 4 | 6.15 |
| Strept pneumoniae | 0 | 0 | 3 | 4.62 |
| Klebsiella pneumoniae | 0 | 0 | 2 | 3.08 |
| **Total** | **3** | **4.62** | **62** | **95.38** |

*The percentage was calculated according to the total number of cases with single infection (3+62=65).*

**Table 5:** Percentage of various micro-organisms isolated as mixed infection of both healthy and diseased buffalo-calves.

| Bacterial isolates | Examined buffalo-calves | | | |
|---|---|---|---|---|
| | Healthy | | Diseased | |
| | No. of Positive cases | % | No. of Positive cases | % |
| P. multocida +E.coli | 1 | 5.56 | 7 | 38.88 |
| Staph aureus + E.coli | 0 | 0 | 4 | 22.22 |
| Staph aureus + Strept pyogenes | 0 | 0 | 2 | 11.11 |
| Strept pneumoniae + E.coli | 1 | 5.56 | 2 | 11.11 |
| Kl.pneumoniae +Corynebacterium ovis | 0 | 0 | 1 | 5.56 |
| **Total** | **2** | **11.12** | **16** | **88.88** |

*The percentage was calculated in relation to the total number of mixed cases (2+16=18).*

**Table 6:** Percentages of the bacterial isolated from the surrounding environment of examined buffalo-calves

| Samples | No. of examined samples | No. of Infected Samples | % of infected samples | Percentages of isolated organisms | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | E.Coli | | P.mul-tocida | | Staph aureus | | Strept Pyogenes | |
| | | | | No. | % | No. | % | No. | % | No. | % |
| Soil | 28 | 24 | 85.7 | 17 | 70.8 | 2 | 8.3 | 3 | 12.5 | 2 | 8.3 |
| Water | 28 | 12 | 42.9 | 4 | 33.3 | 4 | 33.3 | 3 | 25 | 1 | 8.3 |
| Air | 28 | 7 | 25 | 2 | 28.6 | 3 | 42.9 | 1 | 14.2 | 2 | 14.2 |
| **Total** | **84** | **43** | **51.2** | **23** | **53.5** | **9** | **0.9** | **7** | **16.3** | **4** | **9.3** |

**Table 7:** Values of some haematological parameters of both healthy and pneumonic buffalo-calves

| Haematological Parameters | Healthy buffalo-calves | Diseased buffalo-calves |
|---|---|---|
| | $\bar{X} \pm SD$ | $\bar{X} \pm SD$ |
| Hb, g/l | 126.7 ± 4.2 | 89.0** ± 3.8 |
| PCV % | 34.55 ±0.78 | 23.85** ± 0.81 |
| R.BC$_s$, T/L | 8.30 ± 0.32 | 5.16** ± 0.25 |
| W.B.C$_s$, G/L | 9.28 ± 0.76 | 15.95** ± 1.92 |

** ***High significant (P>0.01).***

**Table 8:** Values of Biochemical parameters in both healthy and diseased buffalo-calves

| Parameters | Healthy buffalo-calves (control) | Diseased buffalo-calves |
|---|---|---|
| | $\bar{X} \pm SD$ | $\bar{X} \pm SD$ |
| Total protein, g/ *l* | 78.2 ± 0.80 | 68.9** ± 0.50 |
| Albumin, g/ *l* | 42.9 ± 0.40 | 21.7** ± 0.30 |
| Globulin, g/ *l* | 35.3 ± 0.20 | 47.2** ± 0.50 |
| A/G, Ratio | 12.1 ± 0.20 | 4.5** ± 0.10 |
| Urea, mg/dl | 36.394 ± 3.652 | 69.782** ± 4.906 |
| Creatinine, mg/dl | 1.335 ± 0.012 | 2.592** ± 0.081 |
| Glucose, mg/dl | 63.25 ± 4.60 | 43.75** ± 2.23 |
| AST , U/L | 58.45 ± 5.08 | 94.28** ± 3.62 |
| ALT , U/L | 18.715 ± 1.850 | 31.554** ± 1.220 |
| ALP, U/L | 23.80 ± 2.38 | 44.75** ± 3.22 |

*** High significant (P>0.01)***

## DISCUSSION

Bronchopneumonia is one of the most serious problems among calves causing high mortality in feedlot (Jensen *et al.*, 1976). The diseased buffalo-calves in this study suffered severe dyspnoea weakness and rough hair coat. The animals showed widening of the nostrils, mouth breathing, abduction of the fore limbs, extension of the head and neck, and increase in the heart and respiratory rates. Mucous membranes were congested and cyanosed. Bilateral mucopurulent nasal discharge, moist cough and tearing were also evident. Cryptation and rales were heard with the auscultation at different areas of the lungs. Percussion revealed dull sound in some areas of the lungs in some cases. Similar finding were previously described by Youssef *et al.* (1992); Abd El-Fattah *et al.* (1993) and El-Sheikh *et al.* (1994). Mortality rate reached 6.70 %, which are nearly similar to that recorded by Abd El-Ghani *et al.* (1990) and Healy *et al.* (1993).

Bacteriological investigation of nasal swabs (Table 2) revealed that 5 of 36 samples (13.9%) collected from healthy calves and 78 of 90 samples (86.7%) collected from pneumonic calves were positive for pathogenic microorganisms. 62 of 78 positive samples (74.7%) were

found exhibiting single types of bacteria (single infection) while the remaining 16 cases (19.3%) were found having mixed infection (Table 3). *E.coli* and *Pasteurella multocida*, represent the most common Gram negative bacteria isolated form pneumonic buffalo-calves with a percentage of 23.08 and 21.54% respectively. On the other hand Staph aureus was isolated form 15.38% of these cases and represents the most common Gram positive bacteria.

*Streptococcus pyogenes, Haemophilus somnus, Corynebacterium ovis, Streptococcus pneumoniae* and *Klebsiella pneumoniae* were isolated only from nasal swabs obtained from buffalo-calves suffering from bronchopneumonia (Table 4). These results were in agreement with that recorded by Corboz (1982) and Ismail *et al.* (1993).

The isolated bacteria as mixed infections included *Pasteurella multocida* with *E.coli* in 7 cases (38.88%), Staph aureus with E.coli in 4 cases (22.2%), *Staph aureus* with *E.coli* in 4 cases (22.2%), *Staph aureus* with *Streptococcus pyogenes* in 2 cases (11.1%), *Streptococcus pneumoniae* with *E.coli* in 2 cases (11.1%) and *Klebsiella pneumoniae* with *Croynebacterium ovis* in one case (5.5%) (Table 5). These findings in general were partially coincided with that obtained by Weisbecker *et al.* (1979), Slocombe *et al.* (1984) and Saleh and El-Bably (1998). On the other hand *Pasteurella multocida* and *E.coli* were the colonies isolated from nostrils of healthy buffalo-calves in a percentage of 5.56% and represents devastating environmental challenge as these pathogenes mostly expressed their full pathogenicity in the adverse environmental condition.

Bacteriological investigation of the surrounding environment (soil, air and water) revealed that 43 of 84 examined samples (51.2%) were contaminated with pathogenic microogranisms (Table 6). Soil samples exhibited the highest percentage of isolated organisms (85.7%) followed by water samples (42.9%) then air samples (25%). The astonishing high recovery rate of organisms from the surrounding environment of examined buffalo-calves may be attributed to the unsatisfactory hygienic conditions prevailing in these farms particularly the muddy floor which is constantly damp and heavily soiled with manure. Wills *et al.* (1978) proved that the prolonged exposure of calves to cold and damp conditions reduced the ability of calves to clear up the potential pulmonary pathogens due to reduction in pulmonary oxygen tension and may reduce the ability of lungs to resist colonization by pathogenic organisms. *E.coli* appears to be the most important organisms isolated in

high percentage from the surrounding environment of buffalo-calves followed by *Pasteurella multocida, Staph aureus* and *Strept. pyogenes* in percentage of 53.5, 20.9, 16.3 and 9.3% respectively (Table 6). Nearly similar findings were reported by El-Sayed *et al.* (1992) and Saleh and El-Bably (1998). Soily floor may act as a potential reservoir of several pathogens incriminated as caustive agents of bronchopneumonia in buffalo-calves particularly *E.coli, Staph aueus, Pasteurella multocida* and *Streptococcus pyogenes* which were isolated in a high percentage of 70.8, 12.5, 8.3 and 8.3% respectively.

Improper and unsanitary water troughs represents another devastating route for spreading of these pathogens among buffalo-calves in the examined farms. *E.coli, Pasteurella multocida, Staph aureus* and *Strept pyogenes* were isolated in percentage of 33.3, 33.3, 25 and 8.3% respectively. Similar findings were investigated by El-Olemy *et al.* (1989), Woldehiwet *et al.* (1990) and Saleh and El-Bably (1998).

Air born infection played, also, an important role in transmission of infection among buffalo-calves in the examined farms. Pasteurella multocida, *E.coli, Staph. aureus* and *Strept. pyogenes* were isolated from air samples of the examined farms in significant percentage which are 42.9, 28.6, 14.2 and 14.2% respectively. These findings were agree with that recorded by Weisbecker *et al.* (1979) who suggested that air pollution affects the duration and severity of calf pneumonia rather than incidence of the disease.

The pathogenicity of isolates of *Pasteurella species* and *Haemophilus somnus* to white mice revealed that all the isolated bacteria from the diseased buffalo-calves were highly pathogenic to mice after intraperitoneal injection with $7.5\times10^6$ viable organisms. Acute septicemia and deaths of the injected mice occured within 3-5 days. This agreed with the result obtained by Selim *et al.* (1998).

The role of housing as predisposing to severe outbreaks of respiratory diseases among buffalo-calves has been stated by Parker (1967) and Bryson *et al.* (1978). In the examined farms in this study the high levels of ammonia, the excessively high relative humidity, the extreme temperature variations and the poorly ventilated houses were considered the stress factors which adversly affecting the mucociliary action of the affected calves and leads to higher incidence of respiratory diseases specially bronchopneu-monia.

Hematological investigations revealed that total erythrocytic count (RBCs), hemoglobin concentration (Hb) and haematocrit (PCV) were

significantly reduced (P>0.01) in the diseased buffalo-calves. On the other hand highly significant elevation was found in the total leucocytic count (Leucocytosis) of these calves. These results were coincided with those of Medway *et al.* (1969), Doxey (1971), Al-Allawy *et al.* (1979) and Abd El-Raof and Hassan (1999). The most important basic cause of anemia may be due to the failure of bone marrow cells or the deficiency of the raw materials needed for cell production (Coles, 1986). The leucocytosis was attributed to the bacterial infection and the inflammatory lesions which act as prompt stimuli causing the increase in total white blood cells count and more production of mature and immature neutrophils (Doxey, 1971).

Biochemical studies and the determination of serum levels of total proteins, albumin, urea, creatinine, glucose, AST, ALT and AP revealed the following:

**a)** Highly significant decrease (P>0.01) in blood serum levels of total protein and albumin of buffalo-calves suffered bronchopneumonia (Table8). The obtained data were in agreement with those previously obtained by Youssef *et al.* (1991 and 1992) , Blum *et al.* (1996) and Abd El-Raof and Hassan (1999). Such reduction was attributed to the anorexia of the diseased calves and the inability of their liver to synthetize protein. Hassan *et al.*(1984) and Coles (1986) attributed the reduction of proteins to the stress factors and the general unthriftiness which may affect worsely the hepatic parenchyma resulting in failure of protein synthesis. West *et al.* (1968), Hoe (1969) and Doxey (1971) suggested that certain bacteria or bacterial toxins cause increased capillary permeability and permit escape of plasma proteins in tissues, so the osmotic pressure of proteins is increased in the tissue fluids and at the same time decreased in the blood.

There were a significant increase in the serum globulin levels of the diseased calves which could be attributed to the stimulation of immune system by the infectious agents to produce high amounts of immunoglobulins (Youssef *et al.*, 1992). The increased level of globulin with the reduced level of albumin result in significant decrease in albumin/ globulin ratio (A/G ratio). Affonso *et al.* (1960) and Varley (1976) stated the same results.

**b)** Significant increase (P>0.01) was found in blood serum levels of urea and ceratinine of diseased buffalo-calves (Table 8). Similar results were previously recorded by El-Sheikh *et al.* (1994) and Radostits *et al.* (1995). This elevation in the blood urea nitrogen and creatinine levels

could be attributed to the increased protein catabolism and decreased renal blood flow in association to pneumonia.

**c)** There were a reduction in the values of blood serum glucose of the diseased calves when compared with healthy ones (Table 8). This reduction may be attributed to the anorexia and the depraved metabolic processes (Coles, 1986 and Duncan and Prasse, 1986).

**d)** Significant increases ($P>0.01$) were observed in the activities of serum transaminases (AST and ALT) and alkaline phosphatase (AP) in diseased buffalo-calves when compared with those healthy ones (Table 8) . These data coincided with those of Al-Allawy *et al.* (1979); Aly (1986) and Youssef *et al.* (1991 and 1992). The use of enzymes estimate in blood serum for detection of acute and chronic damage of cells is one of the main diagnostic procedure in clinical medicine (Kaneko and Cornelius, 1970 and Amstutz 1980). Leakage of enzymes into extracellular fluid and hence into blood stream provides a senstive index of deterioration of plasma membrane. Several factors cause leakage of intercellular enzymes as depletion of its glucose, so combined effects of hypoglycemia interferes with tissue enzymes and degenerative changes reducing functional activities.

Elevation in the serum levels of AST, ALT, AP, urea and creatinine were found in some diseased cases which may be attributed to renal ischemia and to the degenerated cells of the kidney and livers caused by the observed minute scattered abscesses in these two organs (PM, Table 1). Kaneko and Cornelius (1970), Rosenberger (1978) and Anistutz (1980) reported similar results.

Finally, it could be concluded that bronchopneumonia in calves is an infections /environment complex disease problem of ecoomical importance manifested by a wide variety of clinical, haematological and biochemical disturbances. Careful early clinical examination, identification of the causative agents and serum biochemical analysis lead to correct diagnosis, treatment and control of such respiratory disorders. The adequate hygienic measures prevailing in the farms including proper housing, adequate ventilation, hygienic disposal of sewage, cleaning and disinfection of calf houses as well as proper management of calves are factors that can reduce the degree of exposure of calves to disease producing agents.

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