

CLINICAL, HEMATOBIOCHEMICAL AND RADIOGRAPHICAL STUDIES OF CAPRINE PNEUMONIA

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Abstract: Fever, dullness, tachypnea, bilateral nasal discharge, cough, dyspnea, and abnormal lung auscultation were all symptoms of pneumonic goats. To evaluate the hematological, biochemical and pulmonary function changes in 45 pneumonic goats compared with 10 apparently healthy ones, whole blood and serum samples were collected. The results revealed that pneumonic goats had significantly lower ($P<0.05$) RBCs, Hb, and PCV levels than healthy goats. A decrease in lymphocyte count and an increase in WBCs and neutrophils were found to be significant ($P<0.05$) in pneumonic goats compared to healthy ones. Blood pH and pO_2 were significantly reduced ($P<0.05$) while pCO_2 , tCO_2 , and HCO_3 were significantly elevated ($P<0.05$) in pneumonic goats compared to healthy ones. Biochemically, K, MDA, Hp and SAA showed significant ($P<0.05$) increase, whereas Cl and TAC showed a significant ($P<0.05$) decrease in the pneumonic goats compared to control group. The echotexture of the pneumonic consolidation on ultrasound resembles that of the liver. The chest x-ray showed increased opacity with a cotton-wool-like look that may be more grey or white. According to the findings, the diagnostic techniques mentioned above are helpful in diagnosing goat pneumonia. So, this study aimed to evaluate some selected biochemical parameters and different diagnostic methods including ultrasonography and radiography in pneumonic goats.

Key words: goats; pneumonia; radiography; antioxidants; SAA

Introduction

Goats are the most versatile domestic animals, they may be found in a variety of climates, including those that are dry, humid, tropical, chilly, desert-like, and mountainous (1). Goats are important primarily because they consistently provide more meat, milk, fiber, and skin (2).

Small ruminant respiratory disorders are complicated, and the respiratory disease complex has a number of different etiological causes. In particular, when bacterial and viral illnesses are present together, unfavorable weather conditions

that cause stress frequently contribute to the onset and spread of such disorders (3).

The risk of respiratory disorders is greatest for small ruminants in hot, humid climates like those found in the Tropics (4). Transportation, weather changes, unfamiliar environments or feed, poor management and nutrition, and excessive stocking density are other stressors that contribute to the development of respiratory disease (5). The primary risk factor for transmission is overcrowding along with involuntary inhalation of polluted air with a variety of potentially harmful compounds (6).

The presence of fever (pyrexia), loss of appetite, nasal discharge (serous or mucoid), coughing, sneezing, signs of difficulty breathing (dyspnea), and abnormal lung sounds upon auscultation are

the typical clinical signs and responses of animals infected with respiratory diseases (7).

Acute phase proteins (APPs) are a group of blood proteins that vary in concentration in animals exposed to external or internal stresses, such as infection, inflammation or surgical trauma (8). Major APPs in goats include haptoglobin (Hp) and serum amyloid A (SAA) (9).

If at all possible, chest radiography should be utilized to determine the location, size, and extent of the lesion, followed by lung ultrasonography to confirm the diagnosis of pneumonia. After that, use ultrasonography to obtain more details about the identified lesions (10). So, this study aimed to evaluate some selected biochemical parameters and different diagnostic methods in pneumonic goats.

Material and methods

The study protocol was approved by the animal care committee of the Faculty of Veterinary Medicine, Zagazig University, Egypt No. ZU-IACUC/2/F/99/2022.

Animals

A total of 45 goats of both sexes (31 females and 14 males Baladi goats) aged between 3 months to 1 year belonged to different private farms in Sharkia governorate and the hospital of Faculty of Veterinary Medicine – Zagazig University, Egypt. This study started from April 2021 to March 2022. Goats were suffering from respiratory manifestations in addition to 10 goats from private farm were apparently healthy and used as control group.

A thorough clinical history, clinical examinations including general clinical examinations for temperature, pulse, respiration, mucous membranes, superficial palpable lymph nodes, rumen motility, heart rate, and physical examination of the respiratory system by percussion and auscultation were carried out on each goat (11).

Diseased goats showed symptoms of respiratory distress including nasal “serous or mucoid” and ocular discharge, rhinitis and congested mucous membranes, dry or moist cough, respiratory distress, cough, depression and inappetence.

Sampling

Using a jugular vein puncture, three blood samples were obtained from each goat. Two milliliters of blood were drawn freely from the jugular vein into EDTA blood collection tubes, and immediately analyzed. The hematological indices total red blood cells (RBCs), hemoglobin concentration (Hb), packed cell volume (PCV%), total leucocytic count (TLC), and differential leucocytic count were measured in these samples (12).

Additionally, 10 ml of blood from the jugular vein is allowed to flow freely into a clean, dry, and labelled centrifuge tube without the use of an anticoagulant for serum preparation (12). After that, it was preserved for the analysis of potassium, chloride, sodium, acute phase proteins (Hp & SAA), and MDA & TAC in a deep freezer set to -20°C .

Heparinized syringes were used to draw venous blood samples from the jugular vein. The blood was immediately examined using a blood gas analyzer for blood pH, partial oxygen pressure (pO_2), carbon dioxide pressure (pCO_2), and bicarbonate (HCO_3).

Ultrasonographic examination

Using the ultrasound machine (Esoate My lab, Neitherland with a 7 MHz linear probe), the lung area of each goat was examined for ultrasonographic changes of lung abnormalities (edema, congestion, or consolidation) (13).

Radiography

The radiography was taken to confirm the pneumonia in order to evaluate the various pulmonary function scores in pneumonic goats (14). Radiographs were performed using an X-ray machine (Pox-300 BT, Toshiba, Rotanode TM, Japan) from both dorsal and lateral views without tranquilizer (15). The exposure factors ranged from 45-50 kV and 50 mA-S with 70-75 cm as focal film distance (FFD). The films were manually processed in a darkroom.

Postmortem and histopathological examination

were applied if mortality occurred (six goats) or after emergency slaughter (three goats). For

histopathological analysis, small lung specimens with a thickness of 0.5 cm were taken from recently dead or emergency slaughtered goats with respiratory disorders. In order to protect the pulmonary alveoli's architectural integrity, lung tissues were inflated with formalin as previously done by Ghanem et al. (16).

Statistical analysis

A Student's T-test was used to statistically examine the data, and the findings were recorded as mean \pm SE. The values were considered significant at $P < 0.05$.

Results

Clinical findings and physical examination:

Anorexia, dullness, depression, dyspnea, nasal discharge, moist painful cough and dry cough were the most prevalent clinical symptoms on pneumonic goats (Table 1). Physical examination revealed that the pneumonic goats had fever (40.22 ± 0.13 °C), shallow rapid breathing (35.00 ± 1.14 breaths/min), an accelerated heart rate (94.80 ± 1.65 beats/min), redness of the ocular mucous membrane and conjunctiva, and crusts around the nasal opening. In contrast

to apparently healthy goats, lung auscultation revealed pronounced wheezing, crackling sound, moist rales, and exaggerated vesicular sound (Table 2).

Hematobiochemical changes

Upon hematology of pneumonic goats, RBCs count, Hb and PCV revealed significant ($P < 0.05$) decrease while WBCs and neutrophils revealed significant ($P < 0.05$) increase with a significant ($P < 0.05$) decrease in lymphocytes count. Monocytes and eosinophils revealed no significant changes. Serum mineral analysis revealed a significant ($P < 0.05$) decrease in Cl, a significant ($P < 0.05$) increase in K, and no significant changes in Na. TAC showed significant ($P < 0.05$) decrease while MDA, Hp and SAA levels showed significant ($P < 0.05$) increase in pneumonic goats when compared with control goats (Table 3).

Pulmonary function tests

In comparison to the control group, the pulmonary function tests of pneumonic goats revealed a significantly ($P < 0.05$) lower pH and pO_2 and a significantly ($P < 0.05$) higher pCO_2 , HCO_3^- , and tCO_2 (Table 4).

Table 1: Incidence of clinical signs in diseased pneumonic goats.

Clinical sign	No. of affected goats	%
Fever	45	100
Cough	39	86
Nasal discharge	23	51
Decreased appetite	41	91
Congested mucous membrane	34	75.5
Cyanosed mucous membrane	11	24.5
Dyspnea	7	15

Table 2: Physical examination of diseased pneumonic goats compared with control

Physical examination	Control group (N=10)	Pneumonic group (N= 45)
Temperature (°C)	39.26 ± 0.08	$40.22 \pm 0.13^*$
Pulse (/minute)	82.60 ± 1.02	$94.80 \pm 1.65^*$
Respiration (/minute)	26.20 ± 0.58	$35.00 \pm 1.14^*$
Mucous membrane	Light rosy red and somewhat bluish, no secretion and no swelling	Congested m.m. in some cases and cyanosed in other cases
Appetite	Normal appetite	Anorexia
Depression	Animals were alert	Depression and recumbency were observed in late stages of pneumonia

Cough	Absence of cough	Some cases showed dry and nonproductive cough while others showed moist cough
Nasal discharge	Absence of nasal discharge	Scanty serous to mucoid in some cases, mucopurulent to purulent nasal discharge in other cases.
Dyspnea	No dyspnea	Some cases showed signs of dyspnea
Lung auscultation	Normal vesicular sound	In some cases, dry rales, moist rales, crepitation and wheezes were heard on auscultation. While, in others there were absence of vesicular sounds and audible heart sounds.
Lung percussion	Normal resonant sound	incomplete to complete dull sound

Data are presented as means \pm S.E. (S.E. = Standard error). * Means significantly different from control at $P < 0.05$

Table 3: Hematobiochemical changes of diseased pneumonic goats compared with control

Parameters (Units)	Control group (N= 10)	Pneumonic group (N=45)
RBCs ($10^6/ \text{mm}^3$)	6.09 \pm 0.03	4.82 \pm 0.17*
Hb (gm/dl)	12.21 \pm 0.05	11.19 \pm 0.16*
PCV (%)	33.09 \pm 0.35	27.07 \pm 0.52*
WBCs ($10^3/ \text{mm}^3$)	12.82 \pm 0.20	16.12 \pm 0.24*
Lymphocytes (%)	69.05 \pm 0.29	63.34 \pm 0.22*
Neutrophils (%)	21.14 \pm 0.18	26.26 \pm 0.30*
Monocytes (%)	4.65 \pm 0.19	5.00 \pm 0.08
Eosinophils (%)	4.72 \pm 0.07	5.49 \pm 0.12
Na (mmol/L)	144.40 \pm 0.18	143.92 \pm 0.32
K (mmol/L)	4.32 \pm 0.02	5.16 \pm 0.15*
Cl (mmol/L)	97.69 \pm 0.23	83.04 \pm 0.88*
SAA (mg/L)	88.60 \pm 1.01	122.61 \pm 0.81*
Hp (g/L)	2.98 \pm 0.10	7.03 \pm 0.09*
TAC (ng/ml)	15.04 \pm 0.08	12.95 \pm 0.33*
MDA (nmol/ml)	6.83 \pm 0.22	9.54 \pm 0.47*

Data are presented as means \pm S.E. (S.E. = Standard error). * Means significantly different from control at $P < 0.05$

Table 4: Pulmonary function test changes of diseased pneumonic goats compared with control

Parameters (Units)	Control group (N= 10)	Pneumonic group (N=45)
pH	7.43 \pm 0.01	7.30 \pm 0.01*
pCO₂ (mmHg)	38.42 \pm 0.14	48.11 \pm 0.33*
pO₂ (mmHg)	39.45 \pm 0.17	36.82 \pm 0.19*
tCO₂ (mmol/L)	19.92 \pm 0.31	25.23 \pm 0.35*
HCO₃ (mmol/L)	19.26 \pm 0.14	23.81 \pm 0.32*

Data are presented as means \pm S.E. (S.E. = Standard error). * Means significantly different from control at $P < 0.05$

Ultrasonographic Findings in normal and pneumonic goats:

The normal ultrasonographic picture of the lungs were typically aerated. This was evidenced by the presence of numerous parallel and regular reverberation artefacts underneath the uppermost hyperechoic linear image. Both pleural leaves were visible as a broad, smooth, hyperechoic line.

Pneumonic lung consolidation was visible as a homogenous or heterogeneous hypoechoic to echoic structure without reverberation artefacts. Due to acoustic enhancement from pleural exudates and the presence of comet-tail artefacts, the visceral pleura appeared thicker and more hyperechoic than normal (Figure 1).

Radiographic findings in normal and pneumonic goats:

Lung radiodensity changed in accordance with lung inflation during radiography, with expiration processes having a higher radiodensity than inspiration processes. Goats with pneumonia had considerable opacity on the radiograph. Opacity appeared as high gray- or white-density areas with the look of cotton wool. The cranioventral region was mostly affected (Figure 2).

Postmortem findings in pneumonic goats:

Upon gross examination of six recently dead and three emergency slaughtered pneumonic goats, trachea revealed frothy serous blood-tinged exudate (Figure 3 A). The lungs were found to

have hard, congested, consolidated, or hepatized parts (Figure 3 B, C & D).

Histopathological findings in pneumonic goat:

Microscopically, fibrin threads and leukocytes infiltration with eosinophilic substances (serous exudates) in the alveoli of some cases (fibrinous bronchopneumonia). Other cases showing thickening of interlobular septa by fibrin threads with alveoli filled with alveolar macrophage (interstitial pneumonia). Some cases showed thickening in the pleura by fibrin thread and leukocytes infiltrations with presences of thrombotic mass inside pulmonary blood vessels (Figure 4).

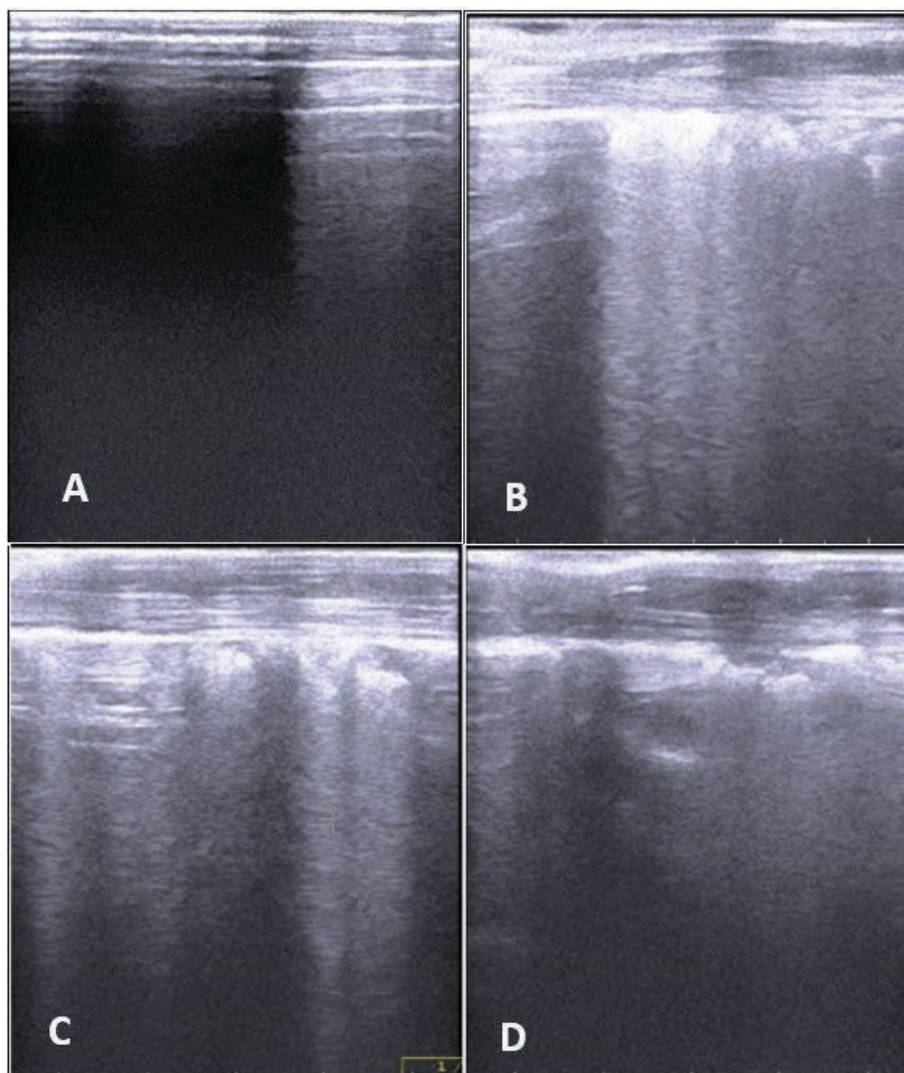


Figure 1: Ultrasonographic imaging of lung. A: normal lung with reverberation artifacts, B, C& D: Absence of reverberation artifacts, consolidation and presence of comet-tail artifacts

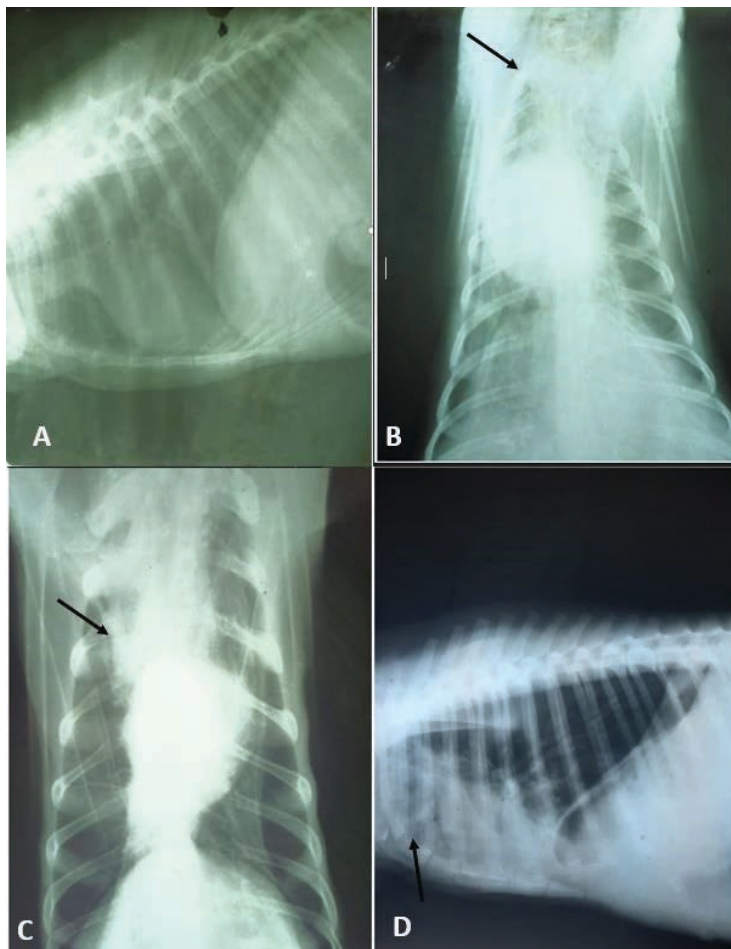


Figure 2: Radiographic imaging of lung. A: Normal radio-density of lung (lateral view) B, C & D: Increased radio-opacity of the lung (B & C: Dorsal views and D: Lateral view)

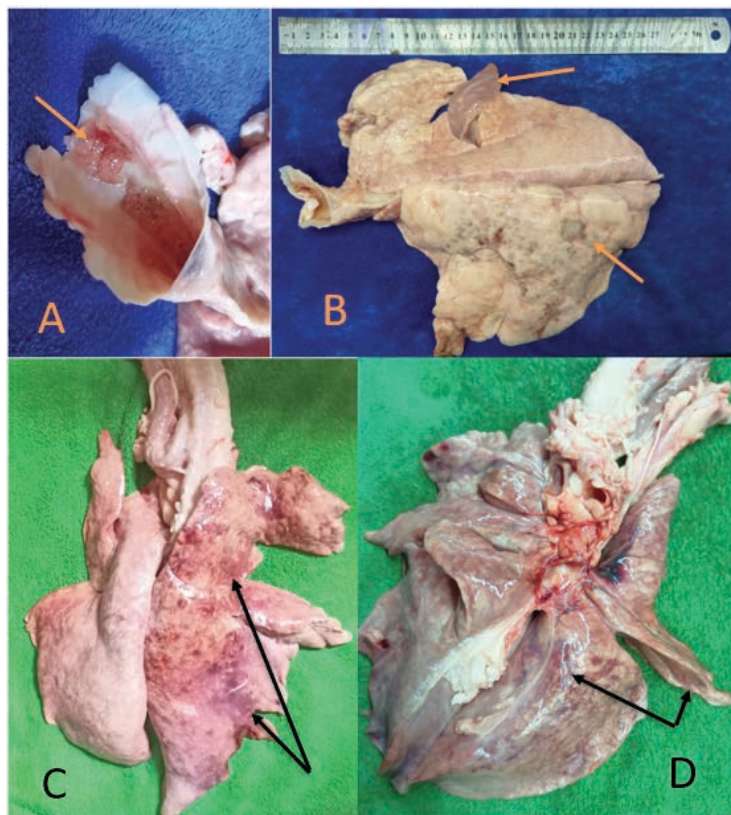


Figure 3: Trachea and lungs of goat. A: frothy serous exudate in trachea. B, C, D: congestion and consolidation of the pneumonic lung (B: lesions in right middle and left caudal lobes, C: lesions in right lung and D: lesions in left lung)

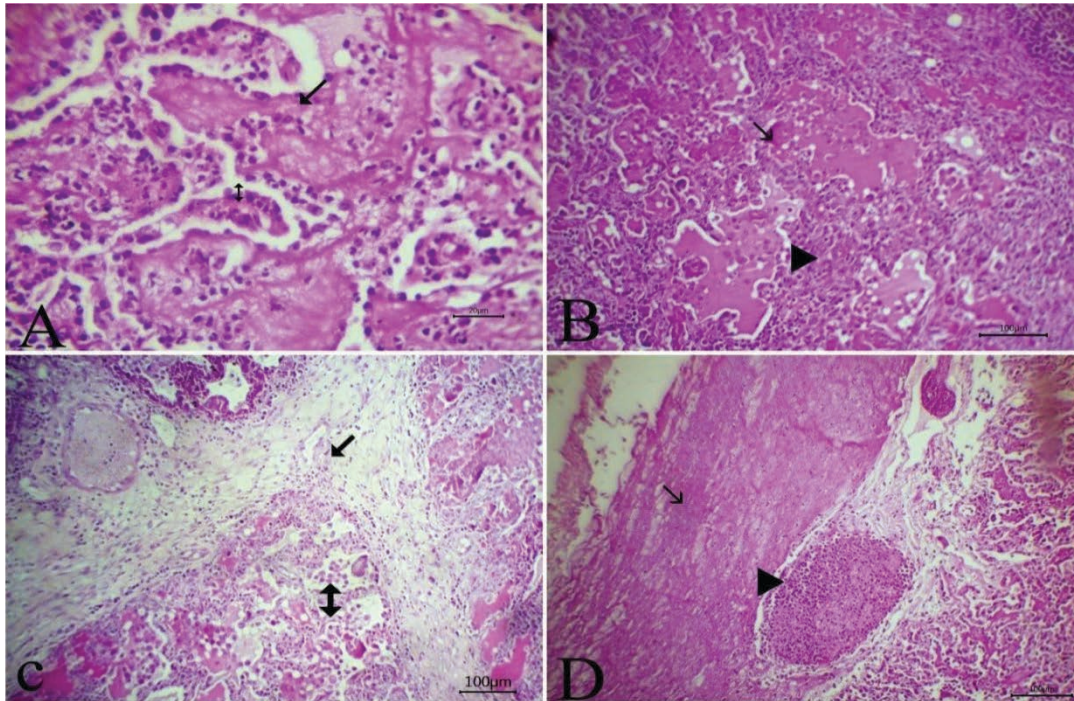


Figure 3: photomicrograph of the lung A: showing alveoli filled with fibrin thread (arrow) and leukocyte infiltration (arrow with 2head) HE, bare20. B: the alveoli filled with eosinophilic substances (serous exudates) (arrow) and leukocytes infiltration (arrowhead) HE, bare100. C: thickening of interlobular septa by fibrin thread (arrow) some alveoli filled with alveolar macrophage (arrow with 2head) HE, bare 100. D: thickening in the pleura by fibrin thread and leuko-cytes infiltrations (arrow) and presences of thrombotic mass inside pulmonary blood vessels (arrowhead), HE, bare 100.

Discussion

Goat industry in Egypt is considered one of the main important sources for meat and milk production and consequently important for human life maintenance (17). Respiratory diseases in goat continues to be a major problem and generally resulting from exposure of animals to environmental and managerial stressors (18).

Particularly in poorer nations, pneumonia poses a serious danger to the production of small ruminants (19). Physical examination, laboratory results, and other imaging methods should all be used to make an accurate clinical diagnosis of pneumonia (20).

The clinical symptoms of pneumonia in goats included pyrexia, sneezing, and coughing, followed by mucous membrane congestion, bilateral nasal discharge, tachypnoea, dyspnea, tachycardia, ocular discharge, anorexia, and mucopurulent nasal discharge and crackles sound on lung auscultation. The results concurred with those of Chung et al. (5).

The hematological examination of pneumonic goats revealed anemia, which may be caused by

an iron deficiency that might be attributed to a decrease in protein and energy intake as a result of anorexia associated with pneumonia, or the sequestration of iron in bone marrow macrophages and hepatocytes during infection, making it unavailable for utilization in hemoglobin synthesis and inhibiting erythropoiesis. It could also be resulted from destruction of red blood cells by micro-organisms secretions (21). The significant increase in both WBCs and neutrophils may be related to inflammatory lesions and presence of bacterial infection in pneumonic goats. However, the significant reduction in lymphocytes could be explained by the adrenal gland's activation under stress in response to the tissue being infiltrated by toxins (22).

Hyperpyrexia during the acute stage of the disease and metastatic infection of the liver and kidneys leading to hepatic and renal dysfunction may be responsible for the hypochloremia with hyperkalemia (23). Hyperkalemia might be occurred in respiratory disorders, especially if acidosis is present. This might be attributed to the exchange of potassium from intracellular to extracellular fluid with H⁺ ions (24).

Higher SAA may be attributed to its function in moving, binding, and scavenging lipoproteins from hepatic cells during the inflammatory phase (25). SAA is also necessary for the APR through the activation of phagocytic cells (macrophage and neutrophil) or the eradication of coliform germs (8, 26).

Following inflammation and/or infection, tissue injury (pleuropneumonia in this study) may induce higher serum Hp levels (27). According to a recent publication, Hp has strong bacteriostatic potentials through the binding of free hemoglobin (Hb), which leads to the creation of Hp-Hb complexes that are then eliminated by the reticuloendothelial system (28).

Pneumonic goats had significantly higher MDA values than control values, whereas significantly lower TAC values were seen in the diseased group compared to the control group (29). As is well known, inflammatory diseases are associated to stimulated oxidative responses and reduced antioxidant defenses. This might be the result of excessive lipid peroxidation in plasma and cells due to many factors or diseases that cause excessive NADPH production, which in turn enhance lipid peroxidation in the presence of the cytochrome P450 system (30).

The changes in pulmonary function tests, which included considerably lower pH and pO_2 and significantly higher pCO_2 , tCO_2 , and HCO_3^- , were similar to those observed by Nagy et al. (31) in pneumonia-affected calves. These results could be explained by respiratory acidosis, which causes pneumonia-related hypoxic conditions and pulmonary hypoventilation. The findings agreed with those of Ghanem et al. (16).

Pulmonary air content makes it more difficult to evaluate the lung parenchyma using ultrasound in a healthy animal. Where the lungs are filled with air, total reflection causes the intercostal transmission of ultrasound to only reach the visceral pleura and cease at the air-filled alveoli (32). When the pulmonary air content is decreased and the lung resembles a liver, an accurate ultrasonographic examination of the lung tissue can be achieved. Visceral pleural surface irregularity may be a precursor to consolidation (33).

The consolidations in the current investigation were consistently hypoechoic and uniform, which may have been caused by the accumulation of exudate, blood, and mucus. Similar results have already been published (34). In this investigation,

the right lung's cranioventral region exhibited the majority of the consolidation. The findings were in contrast to those of Tharwat and Al-Sobayil (32), who noted that consolidation in the right lung was predominantly caudo-dorsally.

For confirming pneumonia in goats, radiography is regarded as one of the most reliable tests (35). According to Masseur et al. (36), radiography research had a 94% sensitivity for identifying cattle lower respiratory tract infections. According to the results of the radiographic examination, the severity of pneumonia in goats was determined and scored as 1, 2, and 3 by looking at the parts of the lung that were affected and the infiltration density in relation to healthy goats. The findings were supported by Falcon et al. (37). Radiographic examination of the lungs that reveals severe lung alterations may be used to determine the condition's severity and prognosis (14).

The present study's findings are supported by the observation that pneumonic lungs with dark red patches and consolidation of several lobes with firm consistency are the most prevalent gross lesions detected in the lungs of most cases (38).

Different investigated goats showed variable degrees of bronchopneumonia, according to the histopathological examination. These results might be explained by the variable susceptibilities of the infected goats. The toxic proteins leukotoxin, lipopolysaccharide, and polysaccharide (39), as well as the inflammatory factors produced by neutrophils and other inflammatory cells (40), may be the cause of these histopathological alterations.

Conclusion

Diagnosis of caprine pneumonia could be confirmed by hematological, biochemical mainly SAA, Hp, TAC and MDA, blood gas analysis, ultrasonographic, and radiographic examination, supported with postmortem and histopathological examination.

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All authors contributed in the study design and in the writing of the manuscript. The samples were collected by SMA, who also monitored the animals. The data was analyzed by SMA, and the results were reviewed by NEA. The final manuscript was read and approved by all authors.

The authors declare that they have no competing interests.

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