Pathological and bacteriological study of lobe distribution of lesions in buffalo (Bubalus bubalis)

Sayyari, M.¹ *; Sharma, R.²
¹Department of Pathology, Faculty of Veterinary Medicine, Shiraz University, Shiraz, Iran. ²Department of Pathology, Faculty of Veterinary Medicine, Shahid Chamran University, Ahwaz, Iran.

Key Words: Pneumonia; lobe; bacteriopathological; buffalo.

Abstract

The correlation between the bacteriological agent of buffalo pneumonia and its pathologic characters were investigated. In the present study, 333 samples of buffalo lungs in Iran were studied for pneumonic lesions and evidence of bacterial and viral infection. The type of pneumonia was classified as interstitial, fibrinous or purulent bronchopneumonia and the anatomical location of lesions was also recorded. In 201 samples with interstitial pneumonia, the lungs were found to be noticeably elastic, edematous and pale. Microscopically, thickening of the alveoli walls, hyperplasia and increased numbers of monocytes was seen. Lesions were mostly found in the right and left diaphragmatic lobes.

In the 55 samples with fibrinous bronchopneumonia, the lungs were macroscopically bright and marbled and firm in texture. Thickening of the alveoli walls and large numbers of neutrophils were evident at the microscopic level, and the majority of lesions were located in the diaphragmatic lobes. Microscopically, a large number of neutrophils but few macrophages were seen.

Of the 24 samples with purulent bronchopneumonia, most lesions were found in the left diaphragmatic lobe. The main bacteria that were isolated were:

Pathological and bacteriological study of lobe distribution of lesions in buffalo (Bubalus bubalis)

Sayyari, M.¹ *; Sharma, R.²
¹Department of Pathology, Faculty of Veterinary Medicine, Shiraz University, Shiraz, Iran. ²Department of Pathology, Faculty of Veterinary Medicine, Shahid Chamran University, Ahwaz, Iran.

Key Words: Pneumonia; lobe; bacteriopathological; buffalo.

Correspondence
Sayyari, M., Department of Pathology, Faculty of Veterinary Medicine, Shiraz University, Shiraz, Iran.
Tel: +98(711)2286950
Fax +098(711)2286940
Email: Sayyari@Shirazu.ac.ir

Received: 11 January 2011, Accepted: 21 April 2011

Introduction

In Khuzestan province (south west of Iran), buffalo mortalities due to pneumonia, particularly pasteurellosis is a common event and regardless of annual vaccination, the acute form of the disease emerges every year. The objectives of this study were to describe the nature and distribution of macroscopic and microscopic lung lesions in buffalo. The adult water buffalo appears to have a high degree of resistance to diseases. But they are more susceptible to pneumonia than cattle. Husbandry weather and immunity affect morbidity. In endemic areas, from 10 to 50 percent of the buffalo population acquired soid immunity through exposure or subclinal infection. Close herding and wetness predispose to an increased morbidity (Zaman et al., 2007). In domestic animals, pneumonia can be classified into four morphologically distinct types i.e., Bronchopneumonia, Interstitial pneumonia, embolic pneumonia and granulomatous pneumonia according to the lesions’ texture, distribution, appearance and exudation. Also, based on the inflammatory lesions’ distribution in the lungs, the pneumonia can be categorized to the cranioventral (most cases of bronchopneumonia), multifocal (embolic pneumonia), diffuse (interstitial pneumonia) and local extensive (granulomatous pneumonia) pneumonia. The most common agents of suppurative bronchopneumonia, particularly in buffalo, are Pasteurella multocida, Bordetella spp., , Actinomyces pyogenes, Escherichia coli and Proteus spp. Given the importance of buffalo in milk and meat production for Khuzestan province, this study could be considered as a basis for future attempts to reduce buffalo mortality due to respiratory diseases.
throughout the entire lobe. This type of bronchopneumonia is known as lobar pneumonia and is defined as the partial or entire involvement of a lobe in which the lesions are distributed. It can occur after shipping fever has been exacerbated by transportation, congestion, stress or infections, particularly by *Pasturella spp.* (Davies et al., 2006; Prado et al., 2007), and can lead to collapse of the affected part of the lung if the main airway is obstructed (Jubb et al., 2007). In buffalo, fibrinous pneumonia is mostly manifest as the lobar type (Mandal et al., 1995; Ayse, 2000).

The pathogenesis of interstitial pneumonia is complex and involves interaction between external factors, including toxins, biological and environmental factors, with alveolar epithelium and type I and II pneumocytes. It can also arise from hematogenous injury to the alveolar capillary endothelium or alveolar basement membrane. In interstitial pneumonia, lesions cause the lung tissue to become enlarged, elastic, pale, edematous and emphysematous (Auto et al., 2007). It is characterized by formation of a hyaline membrane, epithelialization and moderate forms of adenomatosis (Thorn et al., 2000; Welsh et al., 2004). In Iran no systemic works have been performed on buffalo respiratory system. This study could be considered as a basis for future attempts to reduce the buffalo mortalities due to respiratory diseases.

**Materials and Methods**

Of the 1,622 lungs inspected at the Ahwaz slaughterhouse, 333 lungs were diagnosed with pneumonia. Selection criteria included observable lesions such as hyperemia, bleeding, edema, discoloration, solidness or hardness. After anatomical locations of lesions in various lobes were recorded, samples were transported to the bacteriology laboratory in ice bags. Both aerobic and anaerobic bacteria were identified according to procedures described by Quinn et al. (1994) were cultured using standard biochemical tests. Macintosh (Germany-hannover, lower Saxony) anaerobic glass jars with vents were used to culture obligate anaerobic bacteria in a H$_2$CO$_3$ atmosphere. Plates of Brewer's Agar inoculated with the lung swab were placed inside the anaerobic jar, which was then sealed and incubated at 37°C for 7 days. To avoid death of the obligate anaerobic bacteria on oxygen exposure, culture was carried out on reducing media. For preliminary confirmation, all anaerobes isolated on anaerobic Brewer's Agar were cultured aerobically, and organisms which failed to grow aerobically were considered true anaerobes. The cultures were then cultured using specific media containing either mannitol, indole or sorbitol. The samples were then placed into formalin (10%) to be fixed and prepared for pathological study. Finally, 3-5 mm pieces were taken for preparing sections 5-7 μm in diameter using the paraffin method, followed by hematoxylin and eosin staining. Each sample was examined twice to determine the type of pneumonia and identify the different isolated bacteria.

**Results**

Gross and microscopic lesions of the lung were categorized into three groups: interstitial pneumonia, fibrinous bronchopneumonia and supplicative bronchopneumonia (Table 1). A- Distribution of interstitial pneumonia: Out of all 201 (61%) samples, 12 (5.97%) were located in the left apical lobe, 15 (7.46%) in the right apical lobe, 14 (6.96%) in the left and accessory apical lobes (a), 69 (34.32%) in the right diaphragmatic lobe, 75 (37.31%) in the left diaphragmatic lobe, 8 (3.98%) in the accessory lobe and 8 (3.98%) in the medial lobe. The lungs affected by interstitial pneumonia are distinguishable macroscopically beholden to their pale color and rubber consistency and enlargement, thereby in some cases the rib tracks are found on the lung surface. Edema in the alveolar septa and inside the alveoli, hyperplasia of cells type II (epithelization), infiltration of lymphocytes and other mononuclear were observed microscopically, as well. From all of the samples affected by interstitial pneumonia, 201 samples were cultured, of which 33 (16.41%) and 168 (83.5%) samples were found to be positive and negative respectively. Pasturella spp from 13 samples (39.4%), *Pasturella spp* together with other bacteria like *Arcanobacterium pyogenes*, *E-coli* and *Acinetobacter*

**Table 1:** Distribution of lesions in different lobes of lung.

<table>
<thead>
<tr>
<th>No.</th>
<th>Lobe</th>
<th>Interstitial pneumonia</th>
<th>Fibrinous pneumonia</th>
<th>Suppurative pneumonia</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total cases</td>
<td>Percentage</td>
<td>Total cases</td>
<td>Percentage</td>
</tr>
<tr>
<td>1</td>
<td>Left apical</td>
<td>12</td>
<td>5.9</td>
<td>-</td>
</tr>
<tr>
<td>2</td>
<td>Right apical</td>
<td>15</td>
<td>7.5</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>Left and accessory apical</td>
<td>14</td>
<td>6.9</td>
<td>-</td>
</tr>
<tr>
<td>4</td>
<td>Right diaphragmatic</td>
<td>68</td>
<td>34</td>
<td>19</td>
</tr>
<tr>
<td>5</td>
<td>Left diaphragmatic</td>
<td>75</td>
<td>37</td>
<td>18</td>
</tr>
<tr>
<td>6</td>
<td>Accessory</td>
<td>8</td>
<td>3.9</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>Middle</td>
<td>8</td>
<td>3.9</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>Total</td>
<td>201</td>
<td>100%</td>
<td>55</td>
</tr>
</tbody>
</table>
spp from 6 samples (18.18%), other bacteria such as pseudomonas cuosis from 2 samples (6.06%) in the left apical lobe, staphylococcus epidermidis from 1 sample (3.03%) in the right diaphragmatic lobe and from 2 samples (6.06%) in the left diaphragmatic lobe, Staphylococcus non-haemolytic and Arcanobacterium pyogenes from 9 samples (27.27%) were isolated.

Distribution of fibrinous broncho pneumonia: Out of the entire 55 (61.51%) samples affected by this type of pneumonia, 19 (34.54%) were located in the right diaphragmatic lobe, 6 (10.9%) in the auxiliary apical lobe (a), 7 (12.72%) in the right apical lobe, 18 (32.72%) in the right diaphragmatic lobe, 2 (3.63%) in the medial lobe and 3 (5.45%) in the auxiliary lobe. Macroscopically, the lungs affected by fibrinous pneumonia were found congested, heavy, and edematous with marbled lobules. At the microscopic level the dilatation of the alveoli walls due to edema and the abundance of pink liquid fibrin and the accumulation of neutrophils in alveoli spaces were seen. Out of the 55 (16.51%) samples , cultures were prepared from 24 samples, of which 16 samples were found positive as follows: in the right and left diaphragmatic lobes, 6 (37.5%) pasteurella spp, in the left diaphragmatic, 4 (25%) pasteurella spp and Arcanobacterium pyogenes and E.coli, in the right diaphragmatic lobe alone, 1 (6.25%) Pasteurella spp and E.coli, in the right and left apical lobe 3 (18.75%), pasteurella spp and staphylococcus spp and in the medial lobe 2(12.5%) pasteurella Spp.

Suppurative bronchopneumonia: Out of a total of 24 (7.20%) lesions, 11(45.82%) in the left diaphragmatic lobe, 9 (37.5%) in the right diaphragmatic lobe, 2 (8.23%) in the right apical lobe, 1 (4.16%) in the medial lobe and 1 (4.16%) in the auxiliary lobe were seen. Macroscopically the suppurative lungs were found red to grey, solid and pale showing dark and suppurative exudate in the bronchi and bronchioles and debris and secretions in the bronchus and bronchioles in which inflammatory cells of neutrophils and macrophages are predominant. Out of 22 (7.2%) suppurative pneumonia, cultures were prepared from16 samples and 13 samples (81.25%) were found positive as follows: 3 (23.07%) Ecoli, 4 (30.76%) Arcanobacterium pyogenes and 3 (23.07%) Staphylococcus aureus 3 (23.07%) proteus spp.

Discussion

The results of histopathological and bacteriological examinations in the present survey showed that 74% lung of buffaloes had interstitial pneumonia. It is believed that viruses and bacteria play primary and secondary roles in interstitial pneumonia, respectively. In a previous study, parainfluenza type 3 virus was found in 73% of buffalo lungs affected by interstitial pneumonia. The affected lungs were pale and elastic, and most of the lesions were confined to the diaphragmatic lobes. Microscopically, lungs were found to have thicker alveolar walls, a hyaline membrane in the alveoli, an increase in alveolar epithelial cells, no polymorphs in the alveoli and a predominance of mononuclear cells (Ribble et al., 1995; Tegtmeier et al., 1999). In a retrospective study on respiratory diseases conducted in 121 farms hosting 21.5 million cattle, interstitial pneumonia was found to be the most prevalent form of pneumonia, and hyaline membrane formation was the most significant microscopic lesion (Loneragan et al., 2002). Other lesions included thickening of the alveolar septa, which was seen in 46% of cattle, and accumulation of inflammatory mononuclear cells, which was seen in all lungs of calf that suffered from interstitial pneumonia. Results from our study are similar to findings of other researchers. (Mandal et al., 1995; Ayse, 2000). In fibrinous bronchopneumonia, the major findings were the accumulation of exudate and the thickening of alveolar walls due to the increase of fibrin and neutrophils. The accumulated fibrin that formed thick yellow plaques over the pleural surface is characteristic of this disease. Buffalo are very sensitive to pasteurlosis, and mortalities have been reported worldwide (Welsh et al., 2004). In Khuzestan province, the disease occurs even in vaccinated cattle in the form of a peracute respiratory disease showing fibrinous exudates. A fibrinous bronchopneumonia mortality rate of between 10 and 75% has been reported among calves that had been transported, and the main factor that exacerbated the disease was collection of the animals from different areas (Ribble et al., 1995). Out of 8222 macroscopically examined lungs that had been taken from the abattoir, pneumonia was detected in 500 (6.1%) samples. Also the Bacteria including Pasteurella multocida (13.6%), Staphylococcus aureus (7.2%), Corynebacterium spp. (6%), Streptococcus spp. (3.8%), E.coli (2.8%), Bacillus spp. (2.2%), Pseudomonas spp. (2%) and Pasteurella and mixed bacteria (25%) were isolated and identified from 311(82.2%) lungs (Ayse, 2000). In a study conducted on 40 cattle, Pasteurella multica and Respiratory syncytial virus( RSV) were isolated from the respiratory tract of 34% and 21% of animals respectively.it agreed that fibrinous pneumonia was caused by Pasteurella spp. (Thorn et al., 2000). The simultaneous Isolation of the virus, and Pasteurella spp. and other secondary agents such as Corynebacterium and Staphylococcus were reported too (Dyer et al., 2004; Stepe et al., 2005). In acute cases, cultures obtained from lung lesions or tracheal swabs or washes will be diagnostic. In chronic cases, bacterial cultures may be less rewarding; although Pasteurella spp. or Mannheimia spp. may have caused the initial infection, cultures taken later may reveal.
Arcanobacterium pyogenes, a common cause of lung abscesses (Storz et al., 2000). In the present study, purulent pneumonia was observed in the diaphragmatic, medial and apical lobes. Bacterial agents such as Arcanobacterium pyogenes, E. coli, Pasteurella spp. and Staphylococcus aureus were isolated. The high percentage of lesions due to interstitial and fibrinous bronchopneumonia observed in the buffalo warrant further study to elucidate the causative factors. Factors such as insufficient feed intake, hot and humid weather conditions and gas emissions from oil rigs might be influential.

Acknowledgements

I would like to express my appreciation to Dr. Jamshidian, Mr. Ghalaympour and Mr. Hasani for their technical assistances. This study was supported by the Faculty of Veterinary Medicine, Shahid Chamran University, Ahwaz, Iran.

References