Incidence of Contagious Bovine Pleuropneumonia in Livestock of Pothohar Plateau Punjab, Pakistan

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Abstract  
Mycoplasma mycoides variant mycoides causes contagious bovine pleuropneumonia (CBPP), an important infectious disease of cattle's respiratory system. To determine the prevalence of CBPP, a cross-sectional study was done in the Pothohar plateau (districts Rawalpindi, Attock and Jhelum) Punjab. Using a sandwich enzyme-linked immunosorbent assay, 850 serum samples were tested for specific antibodies against M. mycoides subspecies mycoides. Age, gender, and breed were all examined as potential risk factors. The overall prevalence in this study was 17.65%. CBPP prevalence was highest in animals over 7 years of age (27.78%), followed by 4-6 years of age (15.65%), and lowest in animals under 4 years of age (13.33%). This study establishes the prevalence of CBPP in the state, which could pose a threat to the state's cattle export market and necessitates the development of control measures to reduce the disease's economic losses.

Keywords: Contagious bovine pleuropneumonia; ELISA; Mycoplasma; Pothohar plateau.

1. Introduction  
Livestock is essential and predominant part of agriculture system. Productivity and income growth in livestock has robust income multiplier and poverty alleviating outcomes [1]. Livestock sector works as a cushion in moderating the ebb and flow in crop yield on food availability for consumption by stabilizing the food Supply [2]. Mycoplasma mycoides subspecies mycoides tiny colony biotype causes contagious bovine pleuropneumonia (CBPP) [3], an infectious disease of the respiratory system of ruminants [4]. CBPP is currently endemic in Sub-Saharan African countries, with the majority of outbreaks recorded from 20 different countries [5], with Ethiopia, Cameroon, Angola, and Nigeria reporting the highest number of cases [6]. According to the Office International des Epizooties, the CBPP has documented annual economic losses of more than $2 billion owing to mortality [7], production losses, and higher spending in disease control [8]. The disease can be passed from one animal to the next at the herd level via respiratory aerosols, saliva, urine, foetal membranes, and uterine secretions [9]. Anorexia, fever, dyspnea, polypnea, cough, and nasal secretions are all symptoms of CBPP [10]. The majority of infections are localized to the respiratory tract [11], while arthritis in calves less than 6 months of age is common [12, 13]. Early indications of CBPP can be subtle or non-existent, making it difficult to identify it from any other severe pneumonia with pleuritis [14]. Marbling, thickening of interlobular septa, sequestra development, and consolidation of the lungs of CBPP-infected mice were discovered during necropsy [15]. Lesions having a cranio-ventral pattern are frequently unilateral. In some cases, a straw-colored fluid can be visible in the thoracic cavity [16]. A variety of studies, including typical clinical abnormalities, isolation of the etiologic agent from pneumonic lungs [17], and serological
examinations, are used to confirm the diagnosis of CBPP [18]. Serological methods, such as the complement fixation test (CFT) and the enzyme linked immunosorbent assay (ELISA) [19], are employed at the herd level to screen for CBPP positive animals [20], but due to their nature as presumptive testing, none of the two tests can be deemed gold standard for confirmation [21]. There was no published report on the prevalence of CBPP in Pothohar plateau Punjab, Pakistan, to our knowledge. This prevalence data will provide a first indication of the presence of CBPP in targeted animals grown in this geographic area.

2. Materials and Methods

2.1. The study’s Area
The research was carried out in Pothohar plateau Punjab’s districts Rawalpindi, Attock and Jhelum from September 2020 to July 2021. Rawalpindi is in the northernmost part of the Punjab province, located 275 km to the north-west of Lahore Latitude: 33° 36’ 2.52” N Longitude: 73° 04’ 4.44” E, Attock Latitude: 33.7731 Longitude: 72.3741. The latitude of Jhelum, is 32.940548, and the longitude is 73.727631.

2.2. Collection and processing of samples
A total of n= 850 blood samples were obtained aseptically in clot activator vacutainers from individual animals presenting respiratory symptoms before slaughtering in abattoirs. Vacutainers were labelled and delivered to the SB Lab Rawalpindi, for molecular pathology testing. Serum was separated from each sample as described by [22] and analyzed using a qualitative bovine contagious pleuropneumonia sandwich enzyme-linked immunosorbent assay kit (MyBioSource, California, United States) to detect antibodies specific to coated antigens of CBPP using a Bio rad PR 4100 Microplate Reader to detect antibodies specific to coated antigens of CBPP (Hercules, California, United States). Positives and negatives were interpreted as follows: [20].

2.3. Risk Factors
Data on the animals’ age, breed, and gender were also obtained. Animals were divided into three age groups: under 4 years, 4-6 years, and more than 7 years. Cattle were divided into local breeds and cross breeds based on their breed [23].

2.4. Statistics Analysis
The Pearson chi-square test was performed to examine the variations in measured prevalence frequencies. P<0.05 was used to determine whether there were significant differences in frequency.

3. Discussion and Results
The presence of n = 150 positive CBPP 17.65% was discovered in a study of n = 850 cattle serum samples. Significantly, the highest prevalence was found in Jhelum (20.32%), followed by Attock 17.50%, and Rawalpindi 14.62% (Table 1).

<table>
<thead>
<tr>
<th>Districts</th>
<th>Animals Examined</th>
<th>Positive Sample</th>
<th>Positive (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rawalpindi</td>
<td>260</td>
<td>38</td>
<td>14.62</td>
</tr>
<tr>
<td>Attock</td>
<td>280</td>
<td>49</td>
<td>17.50</td>
</tr>
<tr>
<td>Jhelum</td>
<td>310</td>
<td>63</td>
<td>20.32</td>
</tr>
<tr>
<td>Total</td>
<td>850</td>
<td>150</td>
<td>17.65</td>
</tr>
</tbody>
</table>

The findings of this investigation matched those of a serological survey conducted in a commercial cattle market in Nigeria by [24], which revealed a prevalence of 10.65 % utilizing the ELISA serological method. In contrast to our findings, [25] identified significant prevalence 32% in an abattoir survey for CBPP surveillance in Nigeria. This can be related to animal health, sample processing, vaccination programme, and farm and herd management [26, 27]. The highest CBPP frequency was found in animals aged > 7 years 27.78%, followed by animals aged 4-6 years 15.65% and animals aged 4 years 13.33% (Table 2).

<table>
<thead>
<tr>
<th>Age</th>
<th>Animals Examined</th>
<th>Positive Sample</th>
<th>Positive (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 4 Years</td>
<td>210</td>
<td>28</td>
<td>13.33</td>
</tr>
<tr>
<td>4-6 Years</td>
<td>460</td>
<td>72</td>
<td>15.65</td>
</tr>
<tr>
<td>7 and Above</td>
<td>180</td>
<td>50</td>
<td>27.78</td>
</tr>
<tr>
<td>Total</td>
<td>850</td>
<td>150</td>
<td>17.65</td>
</tr>
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These findings revealed a significant association between the age of the animals surveyed and the prevalence of CBPP (P<0.05). Female animals 21.32% had a higher prevalence of CBPP than male animals 11.56% (P<0.05) (Table 3).
In addition, as shown in Table 4, cross breed animals had a greater positivity 23.40% than local breed animals 10.53%, with a P<0.05 difference.

### Table 4. Breed wise Incidence of Contagious Bovine Pleuropneumonia in livestock

<table>
<thead>
<tr>
<th>Breed</th>
<th>Animals Examined</th>
<th>Positive Sample</th>
<th>Positive (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Local</td>
<td>380</td>
<td>40</td>
<td>10.53</td>
</tr>
<tr>
<td>Cross</td>
<td>470</td>
<td>110</td>
<td>23.40</td>
</tr>
<tr>
<td>Total</td>
<td>850</td>
<td>150</td>
<td>17.65</td>
</tr>
</tbody>
</table>

Significantly (7 years and above, followed by the age group of 4-6 years, and the lowest frequency was found in the age group of 4 years) across the three age groups (Table 2). The findings were consistent with those of [28], who found that CBPP is significantly frequent in animals above the age of seven. CBPP is also found in older animals, according to Andrews and indis [29]. This could be owing to the fact that older animals are more likely to be exposed to infections for longer periods of time, have lower levels of immunity, and have more sequestrum than younger animals, who may benefit from maternal immunity to some extent [30, 31]. In contrast to our findings, [32] found no significant difference in the prevalence of CBPP in cattle based on their age (P>0.05). It's worth noting that female animals 21.56% had higher positivity than male animals 11.56%, indicating that female animals were more likely to be infected with CBPP than male animals [33]. Furthermore, cross breed animals had the highest prevalence of CBPP 23.40% compared to local breed animals 10.53%, indicating that cross breed animals were more likely to be infected with CBPP than local breed animals [34]. The prevalence of CBPP was likewise higher in non-descriptive animals [35], according to the current study's findings, which were consistent with [30] previously published data. This could be explained by the fact that male animals were typically sold at a younger age for profit, but females were typically kept for breeding.

### 4. Conclusion

In conclusion, while the initial results of the study imply that CBPP is present in the specific animals studied, this research has limitations. The failure of the ELISA to detect low antibody levels in animals at the early stages of CBPP is the study's main weakness. More confirmative procedures, such as clinical findings, national serological surveillance, post-mortem inspection at the slaughterhouse, and cultures of the etiological factor of CBPP, followed by molecular characterization, should be included in future investigations.

### Conflict of Interest

The authors declared there is no conflict of interest of this article.

### References


