ISOLATION AND IDENTIFICATION SOME BACTERIAL CAUSES OF LUNG ABSCESSES IN SHEEP BY CHROMOGENIC MEDIA

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ABSTRACT

The study carried to isolation and identification some bacterial causes of lung abscesses in the sheep in Najaf province by Orientation chromagar. Forty two lung abscess were collected from died (10) and slaughtered sheep (32). The result of bacterial isolation appeared 32 bacterial isolates included 18 (56.2%) Gram positive bacteria and 14 (43.7%) Gram negative bacteria according morphology and color of colony and confirmed by classical biochemical test. The Staphylococcus aureus were predominant bacteria 15 (46.8%), Escherichia coli 9 (28.1%), Klebsiella pneumonia 5 (15.6%) and Staphylococcus saprophyticus 3 (9.3%).

INTRODUCTION

Lung, the major organs of respiratory system, is susceptible for several infectious and non-infectious agents cause pathological change in farm animals, among these agents, pneumonia (acute and chronic) causes weakness and fate animal leading to high economic loss (1,2).

The respiratory system diseases such as pneumonia and unspecific toxemias were the most common causes of deaths in sheep (3). The respiratory diseases in sheep complex causes such bacteria virus, the bacterial causes of pneumonia is most frequent and lead to high mortality and economic loss (3).

Abscess is a necrotizing lesion characterized by a pus-filled cavity (4). The most common causes of lung abscesses due to aspiration are some bacteria such as Staphylococcus aureus and Klebsiella Mycobacterium tuberculosis. (5). Orientation chromagar is one of a number of new chromogenic agars, it used for identification of some Gram negative bacteria and some Gram positive bacteria (6) the basis of
differentiation of bacteria according to colony colors produced by reactions of (genus or species) specific enzymes of bacteria with a proprietary chromogenic substrate(7, 8, 9, 10).

**MATERIALS AND METHODS**

Forty two specimens (pus) were taken from dead (10) and emergency slaughtered sheep(32) suffering from respiratory signs in some farm of Al Najaf province between the April 2014-April 2015.

Samples were taken by sterile syringes with 17G needles, while thick pus samples taken by sterile surgery blade. The samples put in tubes containing a transport medium (3 ml of tryptone soy broth). Sample transported by a cooled box to the laboratory of veterinary medicine college.

Bacterial examination: All pus samples were cultured onto blood agar (10% defibrinated sheep blood) were incubated at 37°C for 24 hr. The positive growth were sub cultured on chromagar, incubated at 37°C for 24 hr. The identification were done according to the color and morphology of the colony, which described in the product catalog of the company (CHROMagar, France). Confirmed diagnosis by Gram stain and biochemical tests such as catalase, oxadase, coagulase, indole, methyle red, Voges- Proskauer, lactose fermentation and citrate test.

Statistical analysis. The data were analyzed by SPSS version 11.5, (11).

**RESULTS**

A total of 42 samples of the lung abscess (Fig.1) of sheep (10 dead and 32 slaughtered animals) were cultured on blood agar and all samples showed positive growth. The bacterial isolates were sub cultured by streaking on chromagar. 32 out of 42 bacterial isolates, were obtained by chromagar media (8 isolates from dead and 24 from slaughtered animals), table (1).
Figure (1) : lung abscesses of sheep

Table (1): bacterial isolation and identification by Chromagar and biochemical test.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Blood agar</th>
<th>%</th>
<th>Chromagar</th>
<th>%</th>
<th>Classical biochemical test</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deadsheep</td>
<td>10</td>
<td>10</td>
<td>100</td>
<td>8</td>
<td>80</td>
<td>8</td>
</tr>
<tr>
<td>Sulghter</td>
<td>32</td>
<td>32</td>
<td>100</td>
<td>24</td>
<td>75</td>
<td>24</td>
</tr>
<tr>
<td>Total</td>
<td>42</td>
<td>42</td>
<td>100</td>
<td>32</td>
<td>76</td>
<td>32</td>
</tr>
</tbody>
</table>

Thirty two bacterial isolates were obtained in this study. The isolates included 18 (56.2%) Gram positive bacteria and 14 (43.8%) Gram negative bacteria. The Gram positive bacteria were **S. aureus**, **S. saprophyticus**, **S. aureus** was dominant bacteria was 15 (83.3%), 6 from dead and 9 from slaughtered animals. **S. saprophyticus** was 3 (16.6%), 0 from dead and 9 from slaughtered animals, the Gram negative bacteria were **K. pneumoniae** (35.7%), 1 from dead and 4 from slaughtered animals and **E. coli** 9 (64.2%), 2 from dead and 7 from slaughtered animals as in table (2).
Table (2) : percentage of bacterial species isolated from pus of dead slaughter sheep.

<table>
<thead>
<tr>
<th>Bact. Isolates</th>
<th>No.(%)</th>
<th>Bacteria spp.</th>
<th>No.(%)</th>
<th>Dead No.(%)</th>
<th>Emergency slaughter No(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>G+ B</td>
<td>18(56.2)</td>
<td>S.aureus</td>
<td>15(83.3)</td>
<td>6 (40)</td>
<td>9 (60)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>S.saprophyticus</td>
<td>3 (16.6)</td>
<td>0 (0)</td>
<td>3 (100)</td>
</tr>
<tr>
<td>G- B</td>
<td>14(43.8)</td>
<td>E.coli</td>
<td>9 (64.2)</td>
<td>2 (22.2)</td>
<td>7 (77.7)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>K.pneumonia</td>
<td>5 (35.7)</td>
<td>1(20)</td>
<td>4 (80)</td>
</tr>
<tr>
<td>Total</td>
<td>32</td>
<td></td>
<td>32 (100)</td>
<td>9(28.1)</td>
<td>23 (71.8)</td>
</tr>
</tbody>
</table>

Identification of the bacterial species were done according to color and morphology of colony were described by Catalog of manufacturer of the product (CHROMagar, France) and used classical biochemical test for confirmation diagnosis, table (3) figure (2,3,4,5)

Table (3): Color and morphology of colony on chromagar media

<table>
<thead>
<tr>
<th>Bacteria spp.</th>
<th>Color and morphology of colony</th>
</tr>
</thead>
<tbody>
<tr>
<td>E.coli</td>
<td>dark pink to reddish</td>
</tr>
<tr>
<td>S.aureus</td>
<td>golden, opaque, small</td>
</tr>
<tr>
<td>S.saprophyticus</td>
<td>pink, opaque, small</td>
</tr>
<tr>
<td>K.pneumonia</td>
<td>metallic blue</td>
</tr>
</tbody>
</table>
Figure (1) growth *S. aureus* on chromagar and which have golden, opaque, small colony

Figure (2) growth *S. saprophyticus* on chromagar and which have pink, opaque, small colony
Figure (3): growth *E.coli* on chromagar and which have dark pink to reddish colony.

Figure (4): growth *K.pneumonia* on chromagar and which have metallic blue.
DISCUSSION

The respiratory diseases in the sheep is one of the most important problems are cause the death and reduction in productivity In many countries (12). The lack data and studies about lung abscesses of the sheep, but there are many studies isolated similar the current bacteria from pneumonia, pathologic lesions of the lung as well as the normal flora of the respiratory system in sheep (13, 14, 15).

The current study showed the Gram positive bacteria was higher proportion than Gram negative bacteria. In earlier study (16) found Gram positive bacterial infection most causes of lung abscesses, these results were agreed with our results. Another report showed the percentage of Gram positive bacteria (55.2 %) and Gram negative bacteria was (44.7%), that similar percent of our bacterial isolates (17). Classical Culture and biochemical method identified 32 bacterial isolates from lung abscesses of the sheep, included S. aureus (46.8%), E. coli (28.1 %), K. pneumonia (15.6 %), S. saprophyticus (9.3%), which similar bacteria species obtained by (18) and (19) from nasal swabs apparently healthy and from diseased sheep, while disagreement with results whom reported by (20, 21). Also study in Iran mention the bacterial causes of lung lesion and abscesses in sheep carcasses were Pasteurella multocida, Staphylococcus aureus, Klebsiella pneumoniae, Corynebacterium pseudotuberculosis and Actinomyces pyogenes (24.53%), (20.75%), (15.09%) (7.55%), (1.89%) (22) respectively.

This variation due to geographical area and some factors like stress, immune system status and transportation. (23).

The Staphylococcus aureus was dominant bacteria isolated from lung abscesses, this result in agreement with finding by (24) who isolated S. aureus from subcutaneous abscesses in sheep, but (25) found Fusobacterium necrophorum dominant bacteria isolated from bovine abscesses. The difference of bacterial isolation might be due to sample size and potential pathogens appears when repression of the normal bacteria and lead to a variety of pathologies (26).

The results were reported a low percentage of S. saprophyticus isolated from abscesses this like recorded by (27). Ahmed and his group (28) in a previous study observed low K. pneumonia percent from nasopharyngeal of diseased and apparently

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healthy sheep 8.5% .This result is less than recorded by our study .The result of E.coli isolation was (28.1 %) which similar (29) record E.coli(24.7 %), while Klebsiella pneumonia 5% were less than what we obtained in our study. The variable of Klebsiella isolates percent due to the source of Klebesiella spp. It isolated from soil and also from nasal cavity for several healthy animals species (30).

**CONCLUSION**

The results showed the percentage of Gram positive bacteria were higher than Gram negative bacteria and *S.aureus* was dominant bacteria isolated from the lung abscesses of the sheep.

**REFERENCE**


27. KANOEK M, NOUK A and TODA M. (1984). Isolation of obligate anaerobic bacteria from bovine abscesses in sites other than the liver.

