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Bacteriological and Histopathological Studies on pulmonary lesions of Camels (*Camelus dromedarius*) in Sudan

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ABSTRACT

Respiratory diseases of camel especially pneumonia continue to be a major problem commonly encountered in camel. In this study, 45 pneumonic lungs from one humped camels with different sex and ages ranging from six months to 15 years and originated from different states of the Sudan including Kassala, Al Gedarif, Kordofan and Darfur these were subjected to isolation, identification of bacteria and determination of the pathological changes. The isolates were then fully confirmed by biochemical identification using conventional and automated techniques including API kits and automated system Vitek2 Compact. 80 bacterial isolates were recovered, they were: 15 (18.75%) *S. aureus*, 7 (8.75%) S. *epidermedius*, 5 (6.25%) S. *warrner*, 1 (1.25%) S. heamolyticus, 6(7.5%) Str. *pneumonia*, 5(6.25%) Str. *pyogenes*, 1(1.25%) Str. *suis* 11(13.75) *E. coli*, 3(3.75%) *Coryneulcerans*, 1(1.25%) C. *amycolatum*, 1(1.25%) *Actinomyces naeslandii*, *Actinomyces pyogenes*, 9(11.25%) *K. pneumonia*, 7(8.75%) *Ps. aeruginosa*, 2(2.5%) *Aeromonas salmonicida*, 1(1.25%) *Burkhorder iacepacia*, 2(2.5%) Bacillus spp and 1(1, 25%) *Flacklamia homonis*. The histopathological changes were emphysema with an incidence of 25 (55.6%), atelectasis in 24 (53.3%), haemorrhge in 20 (44.4%), oedema in 19 (42.2%) , inflammatory changes represented by fibrinous pneumonia in 25 (55.6%), purulent bronchopneumonia in 13 (28.9%), Aspiration pneumonia in 10 (22.2%), interstitial pneumonia 9 (20%) and abscesses in 4 (8.9%) and Tumors in 3 (6.7%). Pneumonia is known in camel as complex multifactorial disease in which bacterial, viral, mycoplasma and fungal infections combine with other predisposing factors such as rearing systems, stress factors, climatic changes, and unhygienic conditions. Identification of the pneumonic pathogens in the present work cleared that *Staph. aureus*, was the most pneumonic bacteria isolated from lung tissue at rate of 18.75%

Keywords: Camel, Sudan, Bacteria, pathological lesions, lungs, pneumonia

INTRODUCTION

Camesl (*Camelus dromedarius*) is an important multipurpose livestock species uniquely adapted to harsh arid and semi-arid areas (Agab, 2006). They play a vital economic role that can support the survival of millions of people .One-humped camel used mainly as a source of milk, meat and weber. In addition, camel is used as a mean of transportation and tourism (Wernery and Kaaden 2002). Respiratory diseases of camel especially pneumonia continue to be a major problem commonly encountered in camel (Wernery and Kaaden, 2002). The disease is multifactorial in origin often involve a combination of infectious agents as well as environmental and management factors. Bad sanitation, stressful conditions, and an immunosuppressed state are often related to camel respiratory infections (Abass, and Omer, 2005, Taha *et al.,* 2007). Outbreaks occur in camels in different countries, causing economic losses represented in decrease animal productivity, cost of treatment, condemnated parts and sudden death (Bekele, 2008, Abu-Bakr, *et al.*, 2010). Only few studies were found in the available literature on pathological and bacteriological affections of pneumonic lungs in camels (Al-Tarazi, 2001) Therefore, the objective of this work was to study the bacterial aetiology as well as histopathological findings of pneumonia in camels in Sudan.

MATERIALS AND METHODS

Forty five samples of pneumonic lung tissues were collected from camel (*Camelus dromedaries*) from different areas, ages and sex. Bacteriological and pathological studies were done according to standard methods.

***Bacteriological Studies*:** A piece of lung tissue from each samples were cultivated under aseptic condition into blood agar, Brain heart infusion agar, MacConkey's agar, mannitol salt agar, EMB and nutrient agar, then incubated aerobically at 37°C for 24 hours .This was followed by morphological identification of the suspected colonies onto the fermented media according to their staining reaction, shape, size and arrangement. The isolates were then fully confirmed by full biochemical identification using conventional and automated techniques which were API kits and full automated system Vitek2 compact.

***Histopathological Studies*:** For histological sections, the collected lung samples were rapidly fixed in 10% formalin solution, paraffin embedded and sections were prepared and stained with hematoxylin and eosin according to Bancroft and Gamble (2002).

RESULTS AND DISCUSSION

Table (1) shows the different species of bacteria that are isolated from different cases, *Staphylococcus* spp was the most isolated

Table (2) shows the various lesions found in the affected lungs, emphysema, atelectasis, hemorrhage and fibrous pneumonia were the most observed lesions

Table (3) shows the bacteria isolated from affected lungs cases; mixed bacterial infections were isolated from the different lesions.

Table 1: The bacterial species that isolated from affected lungs of camel

Bacterial isolaes	Number of isolate	The percentage
Staphylococcus spp	28	35%
S. aureus	15	18.75%
S.epidermedius	7	8.75%
S. warrner	5	6.25%
S. heamolyticus	1	1.25%
Str. Pneumonia	6	7.5%
Str. Pyogenes	5	6.25%
Str. Suis	1	1.25%
E. coli	11	13.75%
Coryneulcerans spp	3	3.75%
C. amycolatum	1	1.25%
Actinomycesnaeslandii	1	1.25%
Actinomycespyogenes	2	2.5%
K. pneumonia	9	11.25%
Ps. Aeruginosa	7	8.75%
Aeromonassalmonicida	2	2.5%
Burkhorderi acepacia	1	1.25%
Bacillus spp	2	2.5%
Flacklamia homonis	1	1.25%
Total	80	100%

Table 2: The different pathological lesions found in the affected lungs of camels

Lung lesions	The number of affected lungs
1- Emphysema	25(55.6%)
2- Atalactasis	24 (53.3%)
3- Heamorrhge	20(44.4%)
4- Odema	19 (42.2%)
5- Pneumonia	
5-a Fibrious pneumonia	25(55.6%)
5-b Purulent bronchopneumonia	13(28.9%)
5-c spirasion pneumonia	10 (22.2%)
5- dinterstial pneumonia	9 (20%)
6- Abccesses	4 (8.9)
7- Tumors	3 (6.7%)
8-adhision	1(2.2%)

Table 3: The type of bacteria isolated from the different lesions in the lungs of camel

Lung lesions	Bacteria isolated
Fibrious pneumonia	*E. coli, Ps. aeruginosa, K. pneumonae, Staph spp, Aeromonassalmonicida*
Purulent bronchopneumonia	*Staph spp, Strep spp, E. coli, Corynespp, Burkhorderiacepacia, Aeromonassalmonicida*
interstial pneumonia	*E. coli, sp. Aeruginosa, K. pneumonae,*
Abccesses	*Staph spp, Strep spp,Actinomycesspp, Corynespp, Facklamiahominis, Ps. Aeruginosa*
Tumors	*S. suis, staph spp, Bacillus spp*

Histopathological examination of lungs revealed thickening of the alveolar wall and interlobular septa, infiltration of inflammatory cells and exudation of edematous fluid into alveolar lumen (Figure 1), emphysema, necrosis and sloughing of bronchiolar lining epithelium (Figure 2), purulent core surrounded by connective tissues (Figure 3), hyperplasia of smooth muscle of bronchioles and blood vessels, blood vessels dilatation and hyperplasia of the bronchiolar lining epithelium (Figure 4). Per bronchiolar fibrosis and atelectasis (Figure 5) and fibrosis of lung parenchyma (Figure 6).

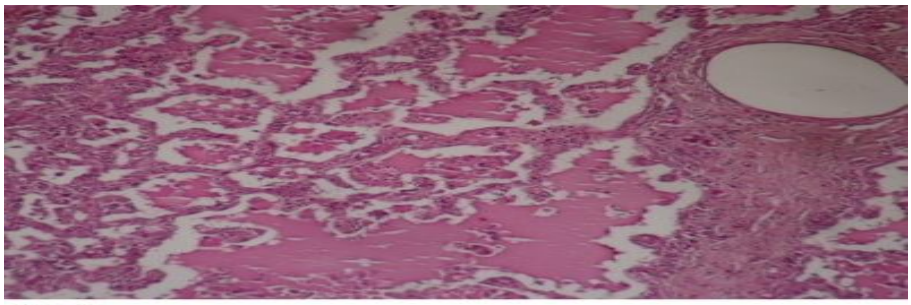

Fig 1: lung section showing thickening of the alveolar septa(interstitial pneumonia), edema, infiltration of inflammotary cells, blood vessels dilatation and thickening of the interlobular septa.

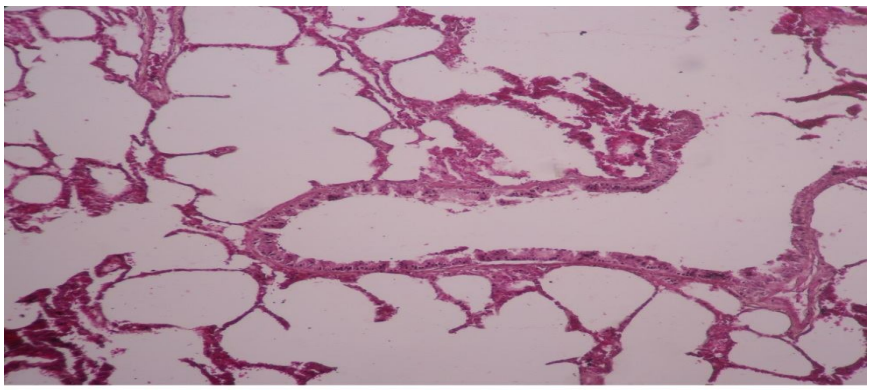

Fig 2: lung section showing enlargment of airspace with evidence of loss or destruction of their walls (emphysema), necrosis and sloughing of the bronchiolar lining epithelium.

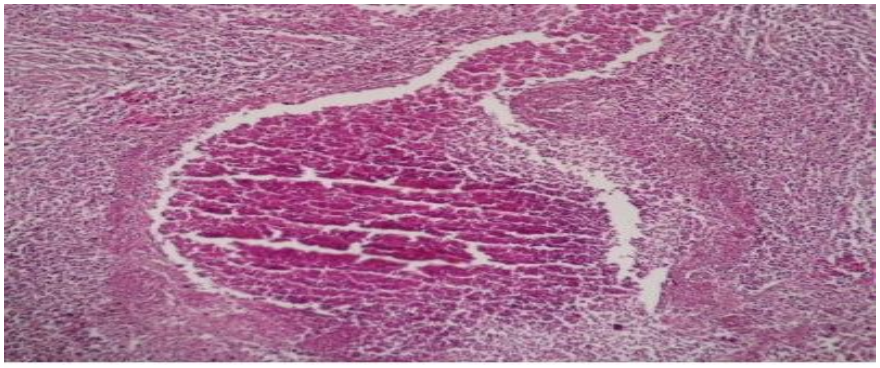

Fig 3: lung section showing abscessation of pulmonary parenchyma.

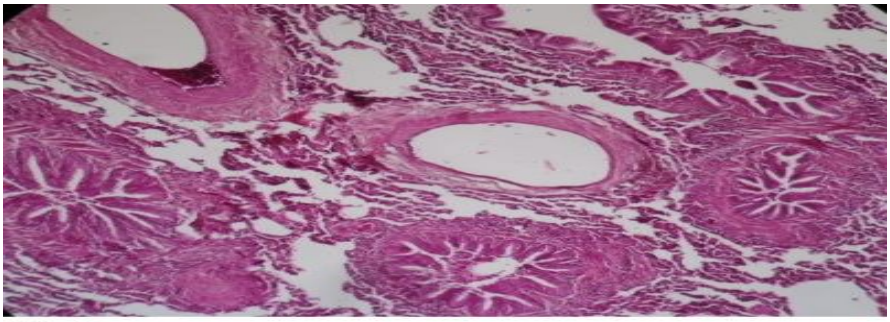

Fig 4:
lung section showing dilatation of the blood vessels, hyperplasia of smooth muscle of bronchioles and blood vessels and hyperplasia of the bronchiolar lining epithelium.

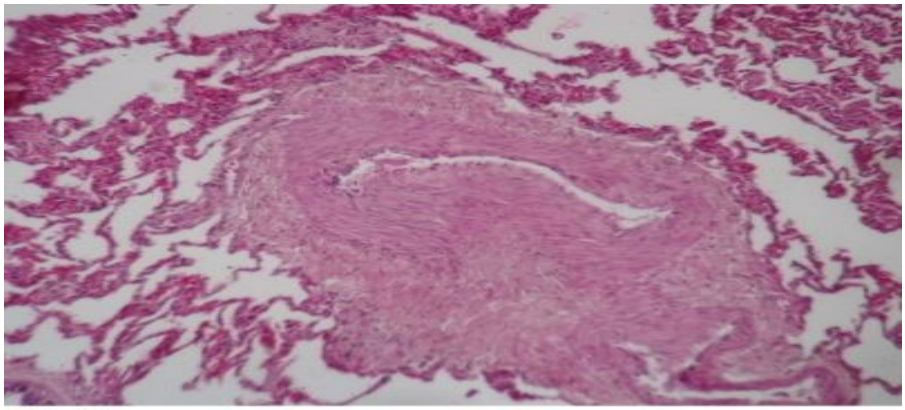

Fig 5:
lung section showing narrowing of bronchiolar lumen by peribronchiolar fibrosis and alveolar collapse (atalactasis).

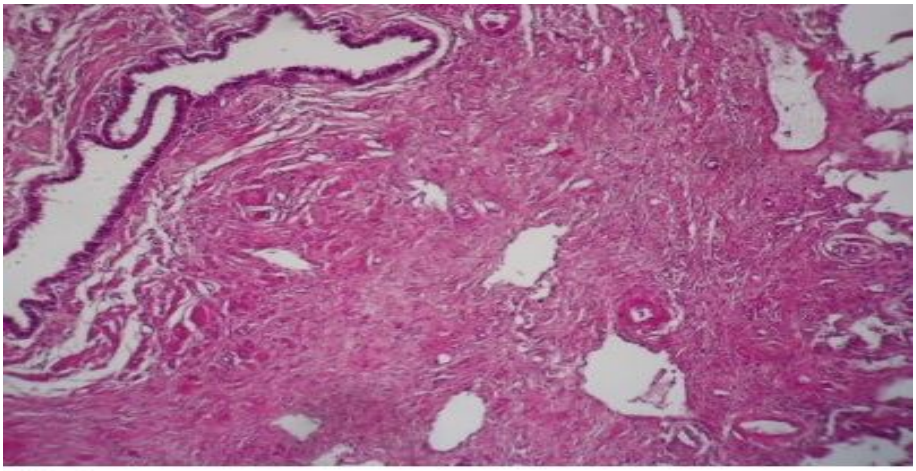

Fig 6:
lung section showing fibrosis of lung paranchyma

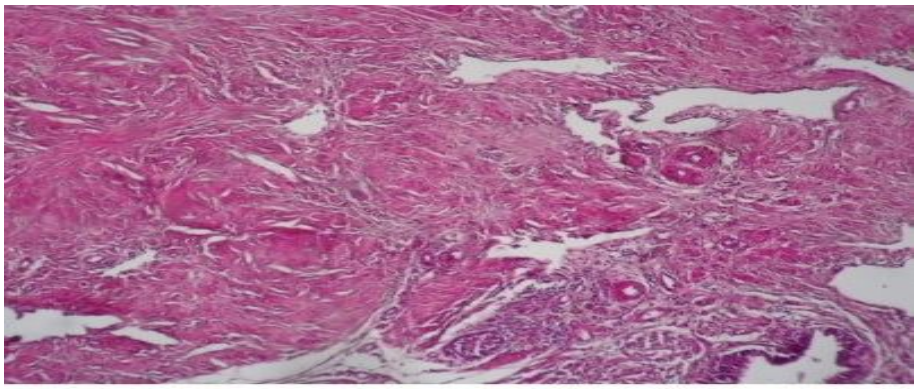

Fig 7: lung section showing narrow veins lumen due to fibrosis (sclerosis).

Respiratory disorders are still serious problem facing camel rearing. The importance of respiratory diseases of camel comes from their prevalence, effect on productivity and for some extent their international spread (Taha, Shlaby *et al.,* 2007; Abass and Omer, 2005).

The respiratory infection is a complex multifactorial disease in which bacterial, viral, mycoplasma and fungal infections combine with other predisposing factors such as rearing systems, stress factors, climatic changes, and unhygienic conditions to produce the disease (Alagaili, *et al* 2014).

A total of 80 bacterial isolates were collected from the 45pneumonic lungs cultured. More than one bacterial species were isolated from 40 pneumonic lungs and no bacterial growth was obtained from 5 lungs. Failure of bacterial isolation in 5 lungs tissues with observable pneumonia may be due to suggestive of mycobacterial infection and in addition probable viral implication or other anaerobic bacteria.

Identification of the pneumonic pathogens in the present work cleared that *staph. aureus,* was the most pneumonic bacteria isolated from lung tissue at rate of 18.75% . This result is higher than that recorded by Al-Tarazi (Al-Tarazi, 2001) as 4%, and is lower than (Awol, *et al*, 2011) as 21.1%.*S. aureus* may predispose the animals to infection by pathogens these results are in agreement with (Taha *et al.,* 2007), (Sayed and Zaitoun, 2009).

In this study different types of pneumonia were noticed these varied from mild to severe fibrinous pneumonia, interstitial pneumonia, and suppurative inflammation. These were observed in most of the examined lungs. These results are consistent with the previous observations of (Al-Tarazi, 2001; and Awol *et al.,* 2011) who demonstrated that acute and chronic suppurative, fibrinous, and interstitial bronchopneumonia were the predominant lesions in dromedary lungs.

In this investigation, the histopathological examination of the lungs revealed that, the recorded pulmonary lesions were classified into the following, lesions associated with changes in air contents such as emphysema and atelectasis, or inflammatory lesions as pneumonia (fibrinous pneumonia, interstitial pneumonia and spiration pneumonia), bronchitis or non-inflammatory lesions represented by pulmonary fibrosis, hemorrhages, edema and tumors.

In the present study, pathological examination of lungs of 45 slaughtered camels revealed different pulmonary lesions. The incidences of lung lesions

are nearly similar to those recorded by (Al-Tarazi, 2001) who recorded the incidence of lung lesions among slaughtered camels in Northern Jordan and almahdi in Saudi Arabia Camels (El-mahdy, *et al,* 2013).

Regarding the alveolar emphysema, 25 cases were recorded in the present work and represented by 55.6% from the total lung lesions. This incidence is higher than that recorded by (El-mahdy *et al.*, 2013) who found that, alveolar emphysema represented 20 % from total lungs lesions of slaughtered camels.

Concerning atelectasis which was detected in 24 (53.3%) and accompanied with pulmonary fibrosis and alveolar emphysema, histopathologically, the alveoli were collapsed, partially or completely compressed, free from air and slit like in appearance.

The interalveolar blood capillaries were dilated and engorged with blood. These results were in accordance with those reported by (Bekele, 1999; El-mahdy *et al.,* 2013; El-Tigani, *et al*, 2004).

20 cases of pulmonary hemorrhages (44.4%) were detected in this study. Macroscopically, multiple reddish patches of variable sizes were seen on pulmonary surfaces. In one case, large irregular sharply demarcated dark red area was noticed. Microscopically, severe congestion of pulmonary blood vessels and inter alveolar capillaries together with the presence of multiple areas of hemorrhages. These results are in agreement with the result of Tigani (El-Tigani *et al.,* 2004).

In the present work, 19 (42.2%) pulmonary edema were recorded. The gross examination of the lungs showed pale edematous areas. Microscopically, accumulations of edematous fluid in the alveoli with congestion of the interalveolar capillaries were seen (Figure 1).

Four (8.9) lungs showed multiple abscesses (1-2 cm in diameter), from which *S. aureus*, hemolytic streptococci spp and *A. pyogenes* were isolated. Such types of abscesses in camel lungs were reported in Iraq, where five camels showed small localized abscesses in the lungs and *A. ovis* was isolated (AL-ANI 1990). These results agree with that was mentioned in the literature about causative agents of abscesses (Noble, 1998), (Ruoff, 1998). In this work, fibrinous pneumonia was found in 25 (55.6%), it was characterized grossly by presence of dark reddish, bluish and purple hepatized areas with emphysematous pale areas gave the lungs mosaic appearance. Microscopically, congestion and thrombosis of some blood vessels of the interlobular septa with areas of red hepatization evidenced by large number of extravasated erythrocytes mixed with few lymphocytes and fibrin threads and areas of gray hepatization represented by presence of fibrin threads mixed with inflammatory cells mostly lymphocytes with few macrophages and neutrophils in the alveoli were noticed, in some areas the alveoli contained plugs of condensed fibrin infiltrated by macrophages and few fibroblast cells as seen in (Figures 5, 6) In addition, thickening of the interlobular septa and subpleural zone due to fibrinous deposits mixed with inflammatory cells particularly lymphocytes with focal areas of compensatory alveolar emphysema were noticed. These microscopic findings are in accordance with that recorded by Bhardwaj *et al.,* (2006).

In cases of interstitial pneumonia represented 9(20%), the lungs were grossly firm in consistency and pale pink in color with thickened

interlobular septa. Microscopically; the alveolar septa were thickened by congested alveolar blood vessels and accumulation of edematous exudates containing leukocytes. In some areas the pneumocytes were proliferated leading to marked thickening of the alveolar septa (Figure 1.). Prominent thickening of the alveolar septa due to presence of homogenous, eosinophilic materials with fibrinous exudate, and few inflammatory cells mainly mononuclear cells and fibroblast cells giving the honeycomb appearance, in addition the alveoli contained mononuclear cells and few detached cells (Figure 1).

In this study, purulent cases of bronchitis and bronchiolitis were diagnosed Grossly 13 (28.9%), thickening of the walls of bronchi and bronchioles with occlusion of their lumens with thick whitish mucous were seen. Microscopically, congestion and thrombosis of the peribronchial blood vessels, hyperplasia and desquamation of the lining epithelium of the bronchi and bronchioles with accumulation of desquamated cells mixed with inflammatory cells in their lumens were prevalent. Focal mononuclear inflammatory cells aggregation beside some bronchioles was noticed (Figure 4). These microscopic pictures were also described by Al-Tarazi Zubair and (Al-Tarazi, 2001; Zubair, Khan *et al.*, 2004).

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