Iraqi Journal of Veterinary Sciences, Vol. 36, Supplement I, 2022 (151-160)
Proceedings of the 7th (1st International) Scientific Conference, College of Veterinary Medicine, University of Mosul

Iraqi Journal of Veterinary Sciences

www.vetmedmosul.com

Histopathological study of sheep lung roaming in dump zones

H.B. Al-Sabaawy[1], E.S. Mustafa[1], A.M. Rahawi[1], M.T. Jaber[2], E.K. Al-Hamdany[1] and S.M. Al-Hmdany[1]

[1]Department of Pathology and Poultry Diseases, College of Veterinary Medicine, University of Mosul, Mosul, [2]Forensic DNA Center for Research and Training, Al-Nahrain University, Baghdad, Iraq

Article information

Article history:
Received September 20, 2022
Accepted December 06, 2022
Available online December 06, 2022

Keywords:
Pneumonia
Oat cell
Nodular lesions
Necrosis
Dump zones

Correspondence:
H.B. Al-Sabaawy
hadeelbasim2006@gmail.com

Abstract

Respiratory system disease is common in sheep, which is frequently difficult to detectable in live animals until it becomes serious and complicated. This study was aimed to describe sheep lung lesions in Mosul city, which roam in dump zones and have been slaughtered at butchers' shops, fifty male and female sheep 1-5 years were used at the current work from March to December 2021; affected Samples were collected and prepared for standard histopathological analysis. The special stain was also used for carbohydrates demonstrated inside and around lung lesions. The results of the current study showed the presence of gross and histological finding changes, the gross lesion presented by inflammation of the upper right lope, congestion, and bleeding on all parts of the lope in addition to different sizes of the nodular lesion. In contrast, the pneumonic histological finding was 12.1%, circulatory disturbance 3.5%, parasitic infections 10.0%, disturbance of growth 7.1%, Necrotic area surround by swirling macrophage 2.0% in addition to the deposition of calcium salt and hemosiderin pigmentation. Organizing Pneumonia (OP) and Masson's bodies were one of the most detected lesions in the roaming sheep, it was scored by Masson Trichrome from mild, moderate, and severe fibrosis. All lesions were categorized according to the type of lesion, grade, and kind of it. From the current work, we demonstrated that pneumonic disorder represented a considerable and serious problem in the animal livestock industry and leads to economic loss in milk, wool, and sheep meat production. Additional studies are recommended to determine the etiological agent of sheep lung lesions whether it is a fungal, bacterial, or viral agent.

DOI: 10.33899/ijvs.2022.135830.2527,

Introduction

Sheep are considering the most important future livestock growth, it has the capability to convert different kinds of forages to beneficial products for population, such as milk, wool and mutton. Sheep is an important source of animal protein in all the world, it is necessary to study the pathological disease, clinical signs and symptoms that affected these animals for enhance their product of milk and meat (1). Respiratory system disease such as bronchitis, pneumonia, fibrosis and lung cancer continue to be an important problem in animals' flocks (sheep, goat, and bovine) (2), affecting all animals age and species, many factor play a role in the occurrences of these disease such as environmental condition and poor management (3,4). the effects of these diseases in small ruminants have increased in recent years, especially in developing countries (5). Pneumonia is one of the important and common diseases that affect the lower respiratory tract of sheep, especially lambs, which may be acute or chronic or even progressive type, effect on animal production, carcasses downgrading delayed growth and sudden death.so it considers a major source of

economic losses represented by treatment coast, unthriftiness (6). Due to the anatomical and histological properties of the lung as well as the ratio between the alveolar surface and metabolic weight make it more susceptible to diseases (7,8). The current work was aimed to study the more clinic pathological changes that affected sheep which roaming in dump zones including both macroscopic and microscopic changes in the lung tissue

Materials and methods

Ethical approve

The current work was approved by ethical committee for animal extermination of the forensic DNA center of Al-Nahrain University based on meeting on 5th of April 2022.

Collection of lung sample

Fifty male and female sheep 1-5 years were used in the present study during March to December 2021. The samples were collected from animals that have been slaughter at butcher's shops.

Gross and Histopathological exam

All specimens were inspected grossly in order to inspect any abnormal lesion in lung which appears by naked eye or by palpation and observation of any changes in the shape, size, color and lung texture after Specimen was collected from the infected part in a clean pack and transported to the lab in a cool pox. Histopathological examination was carried out by trimming the sample with 2 cm in size then fixed in 10%neutral buffered formalin, dehydrated, xylol cleared and blocked with paraffin wax (9,10) and finally sectioned by microtome into 5-6 thickness and stain with routine hematoxylin and eosin stain (11) as well periodic acid Schiff (PAS and Masson's Trichrome) were used for better clarification of the lesion (11).

Results

Gross pathological lesion

As shown in figure 1, 50 of the total 70 cases had gross lung involvement; the most common infection was pneumonia which was present in 17 cases and accounted for 12.1% for the total cases. Figure 2 also revealed the presence of circulatory disturbances (hemorrhage and pulmonary congestion) in 5cases which accounted 3.5% of the total cases. Infection due to parasitic were represented by hydatid cysts in 15 instances with a percentage of 10.0% (Figures 3 and 4), nodular lesion in 10 cases with 7.1% (Figure 5) and pulmonary necrosis in 3 cases with a percentage of 2.1% (Figure 6).

Microscopic lesion

Table 1 showed the lung microscopic lesions which revealed the presences of different type of inflammation such as suppurative bronchopneumonia represented by infiltration of multinucleated inflammatory cell and bronchiectasis of the lung (Figures 7 and 8), also showed fibrinous bronchopneumonia which appeared as accumulation of fibrin filament inside the bronchi wall and infiltration of lymphocytes and nodule image (Figures 9 and 10), there was also oedema which was associated with vascular changes, bleeding within the alveoli and bronchi in additional to blood clots within the blood vessels image (Figures 11 and 12) Parasitic infections was evident with eggs and larval stage of the parasite inside the lung tissue in additional to presences of hydatid cysts with heavy infiltration of inflammatory cell within the lung tissue and adjacent to the hydatid cysts image (Figures 13 and 14). Disturbance of growth was seen and it was represented by demonstrated hyperplasia of the bronchi epithelial cell, atrophy of the mucous gland image (Figure 15), necrosis of the cartilage tissue and degeneration the bronchi and cell lining the alveoli was also observed image (Figure 16 and 17). There was also deposition of calcium salt with hemosiderin pigment inside lung tissue image (Figures 18 and 19).

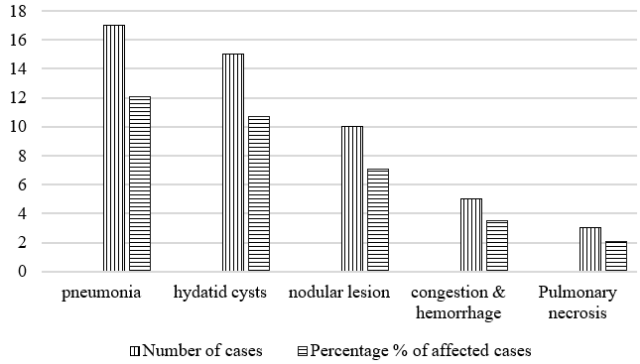

Figure 1: Showed gross finding kind, number of cases and percentage of it.

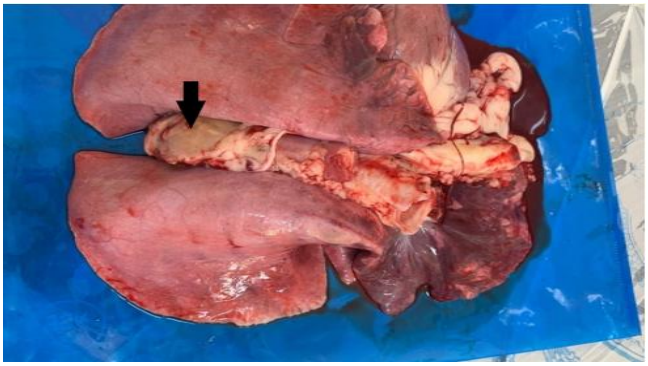

Figure 2: Macroscopic appearances of affected sheep lung showing pulmonary pneumonia with inflammation in the upper right lobe (arrow).

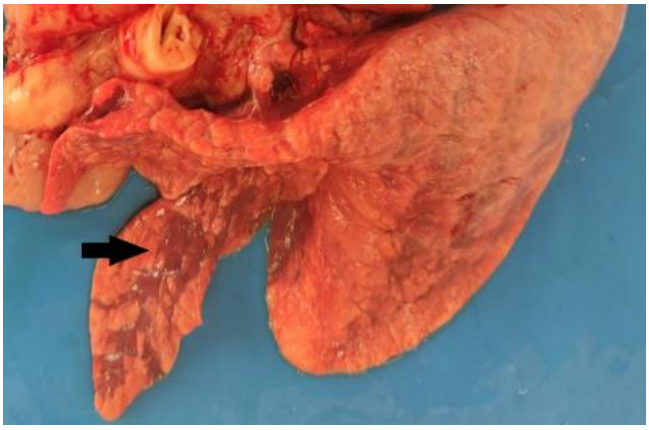

Figure 3: Macroscopic appearances of affected sheep lung showing bleeding and congestion in all part of the left lobe (arrow).

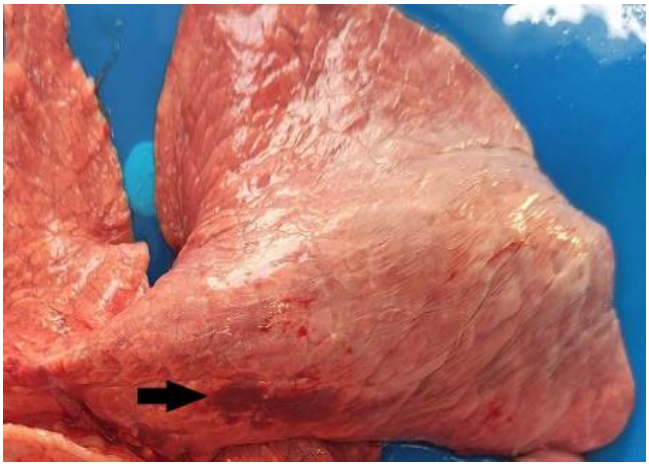

Figure 4: Macroscopic appearances of affected sheep lung showing large hydatid cyst in the lower right lobe.

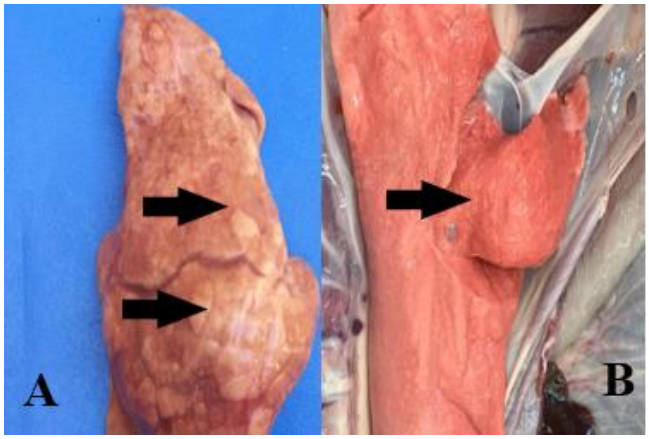

Figure 5: Macroscopic appearances of affected sheep lung (A) showing the spread of nodular lesion with various shape and size inside the pulmonary lobe (B) nodular lesion in the additional lobe of the right lung.

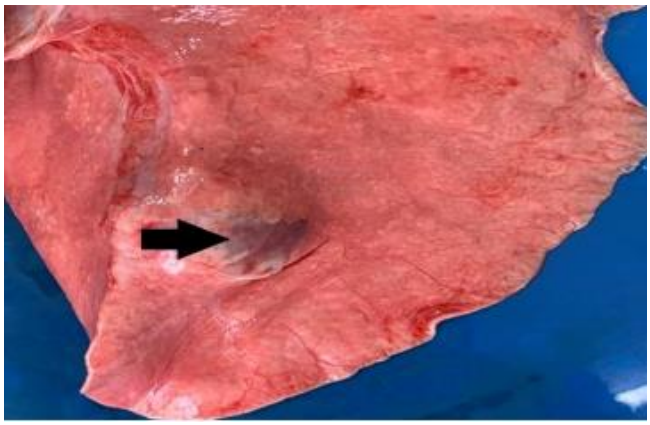

Figure 6: Macroscopic appearances of affected sheep lung showed necrotic granulomatous inflammatory foci in the posterior lobe of the lung (arrow).

Table 1: Categories of lung lesion including description, grade of it and type of lesion

Categories	Lesion description	Grade	Type of lesion
Inflammation	Inflammation with infiltration of lymphocytes inside bronchi wall, bronchiectasis in additional to multiple nodules inside lung tissue	Mild	Progressive
Disturbance of circulation	Blood clots within the blood vessels &bleeding within the alveoli and bronchi	Mild	Progressive
Parasitic infections	Larva stage, eggs & hydatid cyst inside lung tissue	Moderate	Progressive
Disturbance of growth	Hyperplasia of the epithelial cells lining the bronchi as well as there is atrophy of the mucous gland	Moderate and sever	Non-progressive
Necrosis	Necrosis in the cartilage surrounding the bronchi &degeneration inside the cell lining the alveoli	Sever	Non-progressive
Calcification	Deposition of calcium salt in the tissue and presence of hemosiderin pigmentation inside the lung	Sever	Non-progressive

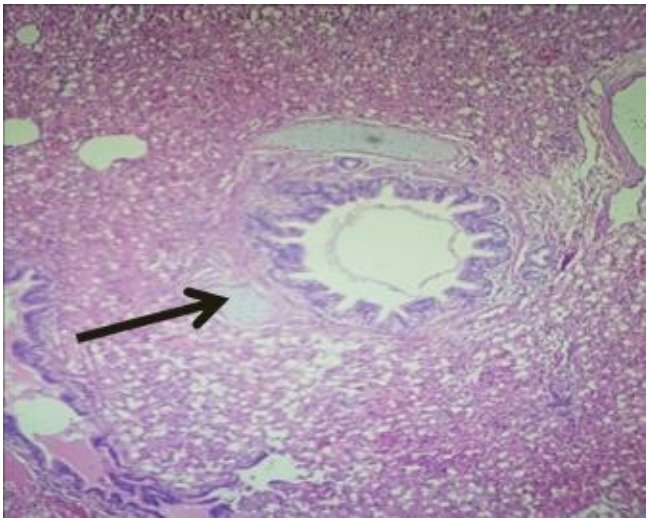

Figure 7: Micrograph of sheep lung showed infiltration of inflammatory cell with inflammatory exudate. H&E, 100x.

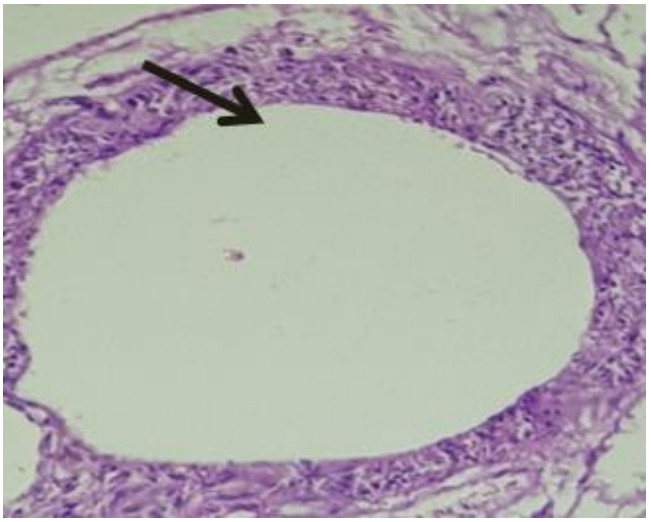

Figure 8: Micrograph of sheep lung showed bronchiectasis of the lung. H&E, 100x.

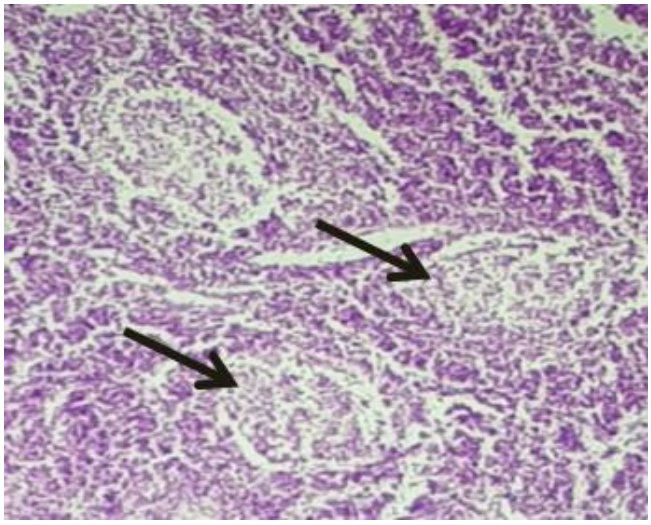

Figure 9: Micrograph of sheep lung showed multiple nodules inside the tissue. H&E, 100x.

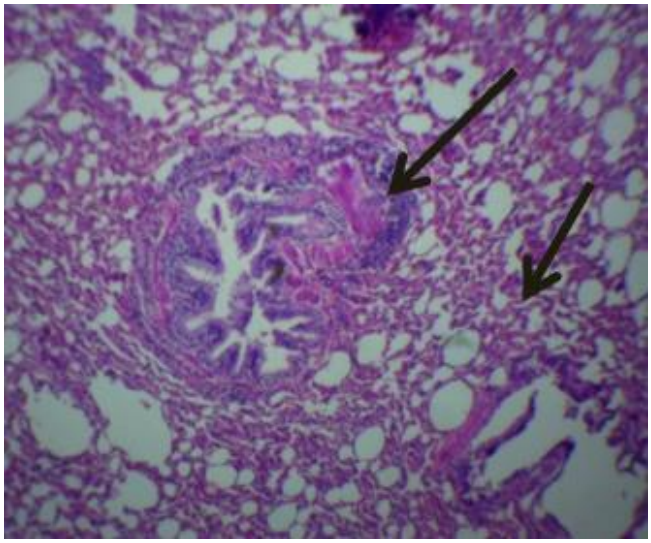

Figure 10: Micrograph of sheep lung showed infiltration of lymphocytes with accumulation of fibrin filaments inside bronchi wall and emphysema (arrow). H&E, 100x.

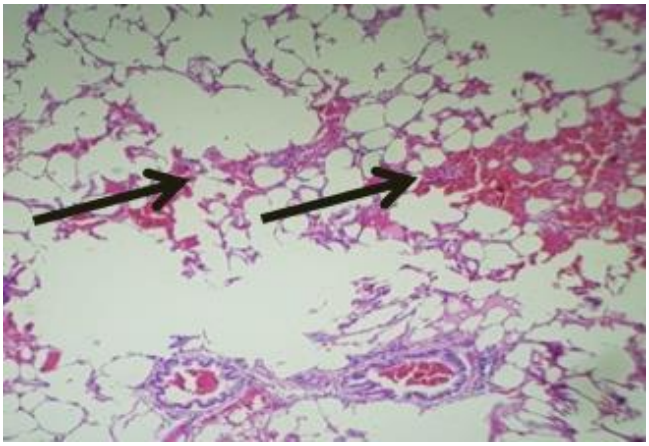

Figure 11: Micrograph of sheep lung showed thrombus with infiltration of inflammatory cell (arrow). H&E, 100x.

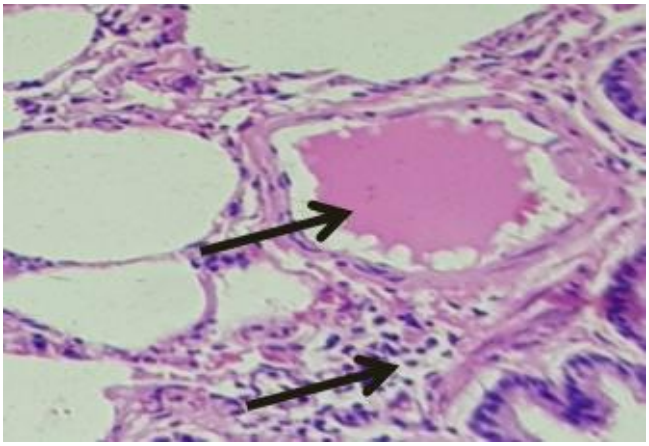

Figure 12: Micrograph of sheep lung showed thrombus with infiltration of inflammatory cell (arrow). H&E, 400x.

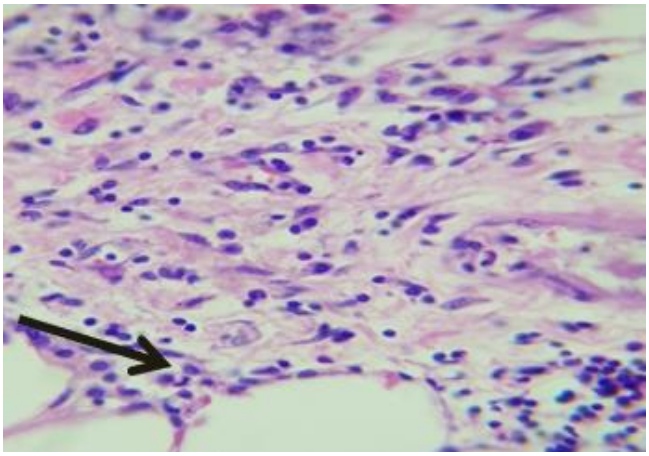

Figure 13: Micrograph of sheep lung with parasitic pneumonia showed egg and variable stage of parasites. H&E, 400x.

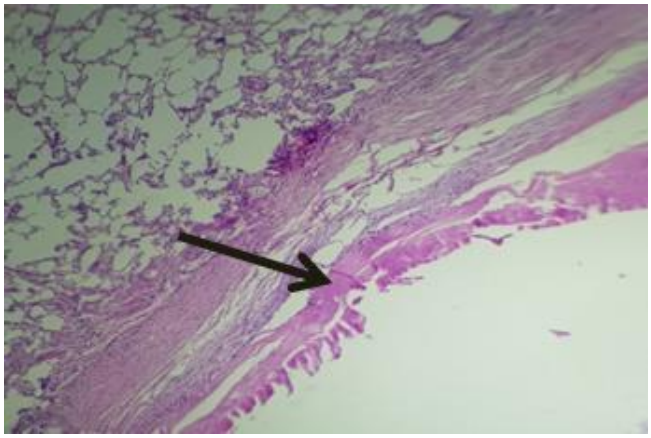

Figure 14: Micrograph of sheep lung showed the presence of hydatid cyst inside lung tissue. H&E, 100x.

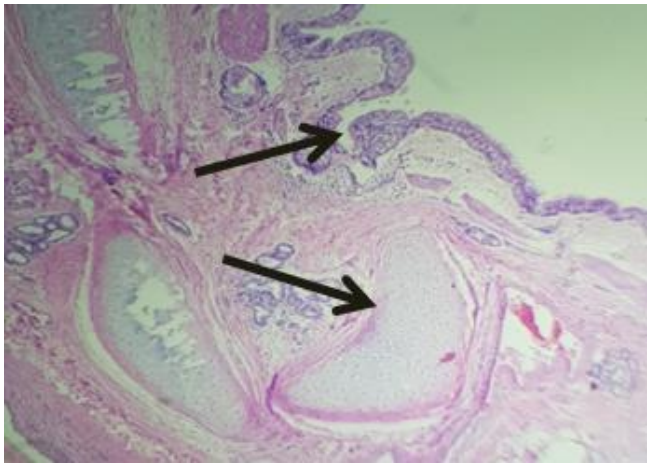

Figure 15: Micrograph of sheep lung showed hyperplasia of the epithelial cell lining the bronchi with atrophy of the mucous gland. H&E, 100x.

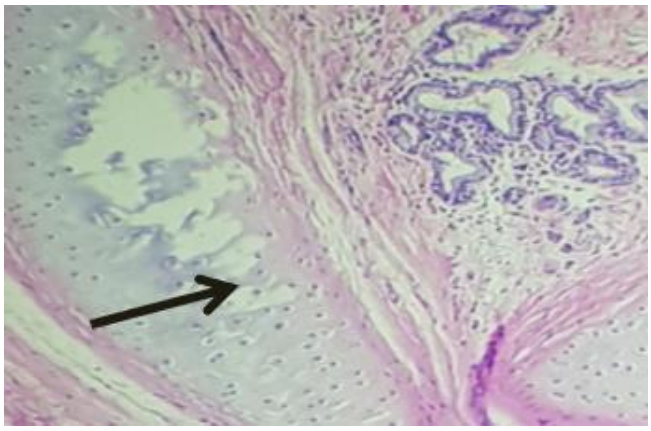

Figure 16: Micrograph of sheep lung showed necrosis inside the bronchi cartilage. H&E, 100x.

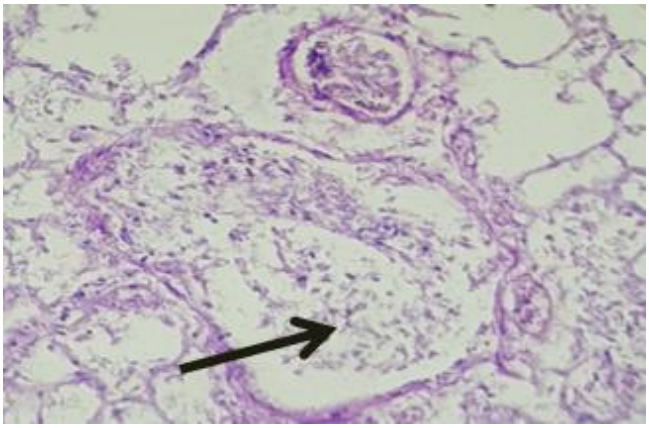

Figure 17: Micrograph of sheep lung showed degeneration and necrotic lesion inside the bronchi and cell lining the alveoli image. H&E, 100x.

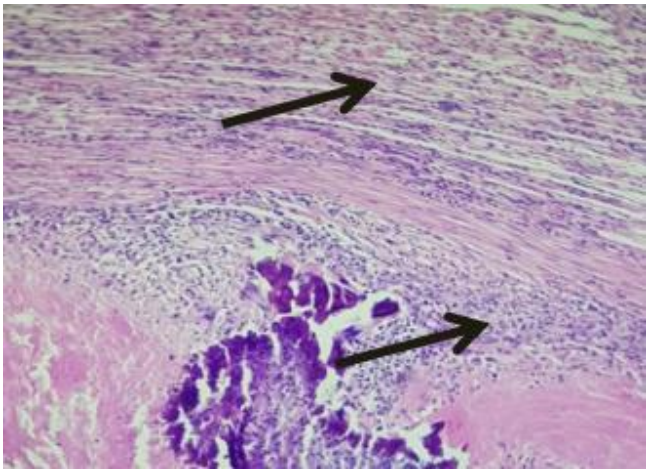

Figure 18: Micrograph of sheep lung showed deposition of calcium salt with coagulate necrosis and infiltration of inflammatory cell. H&E, 100x.

Special stain

In table 2 the histopathological finding of lung showed the presences of positive neutral mucopolysaccharides (mild - moderated) which demonstrated by periodic acid Schiff stain (Figures 20-22), collagen fiber was prominent as fibrotic foci in addition to mason bodies which distinguish as a proliferation of fibroblast (deposition of collagen) by Masson Trichrome (Figures 23-27).

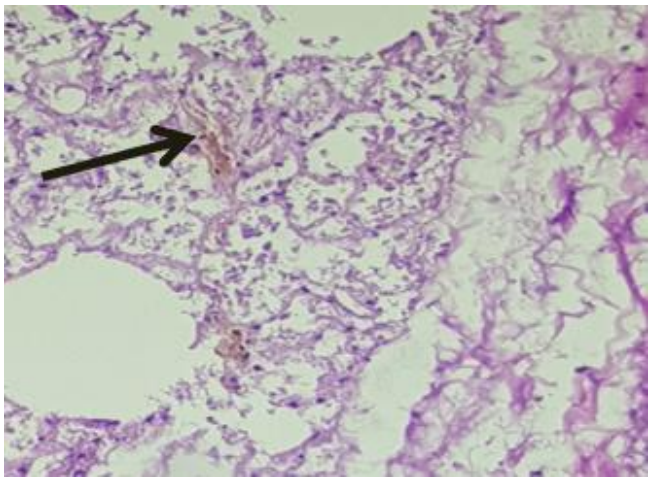

Figure 19: Micrograph of sheep lung showed hemosiderin inside lung tissue. H&E, 100x.

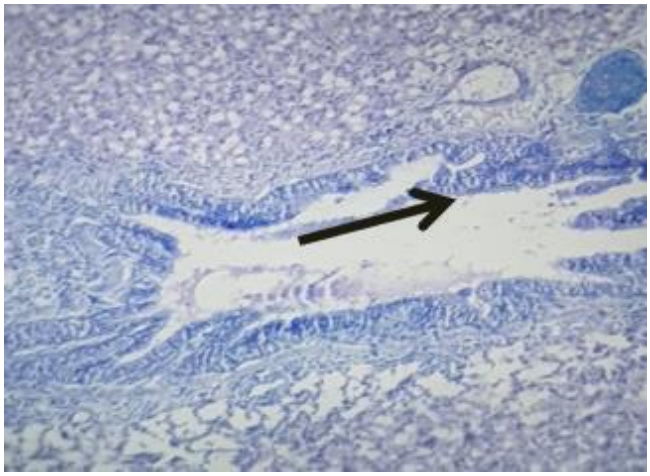

Figure 20: Micrograph of sheep lung showed hyperplasia of gland with mild mucopolysaccharides. PAS stain, 100x.

Table 2: Categories of lung change according to figure number, type of stain, degree and scoring

Figure number	Type of stain	Scoring	Degree of lesion
18	PAS	+	Mild
19	PAS	++	Moderate and sever
20	PAS	+++	Sever
21	PAS	+++	Sever
22	Masson trichrome	+++	Sever
23	Masson trichrome	+	Mild
24	Masson trichrome	++	Moderate
25	Masson trichrome	+++	Sever

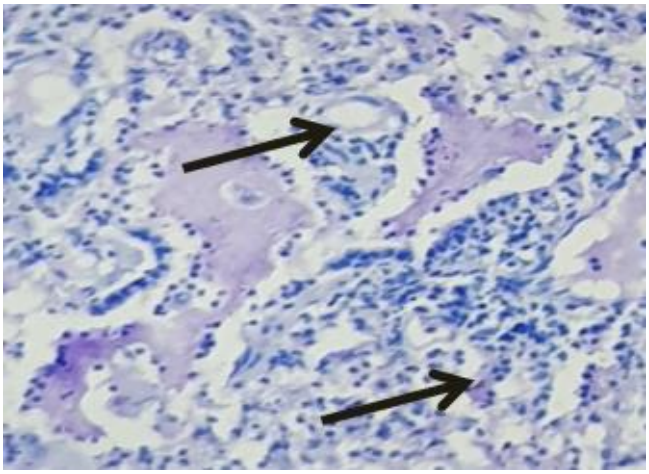

Figure 21: Micrograph of sheep lung showed hyperplasia of gland with moderate and sever mucopolysaccharides. PAS stain, 400x.

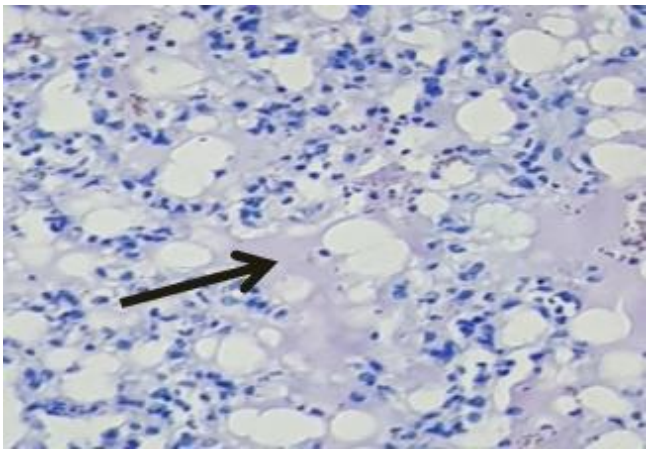

Figure 22: Micrograph of sheep lung showed necrosis and emphysema inside alveoli wall. PAS stain, 400x.

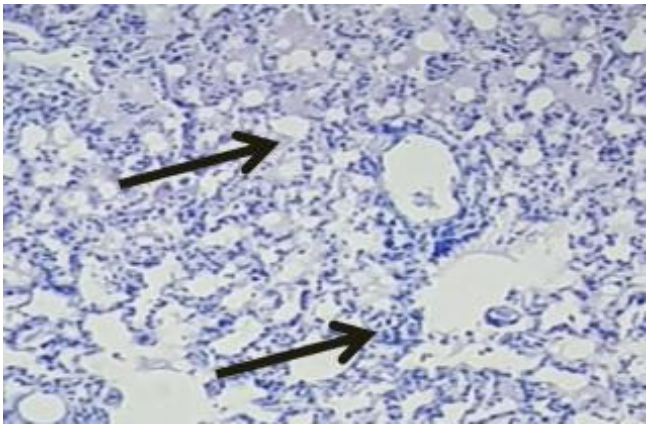

Figure 23: Micrograph of sheep lung showed swirling macrophage (oat cell). PAS stain, 100x.

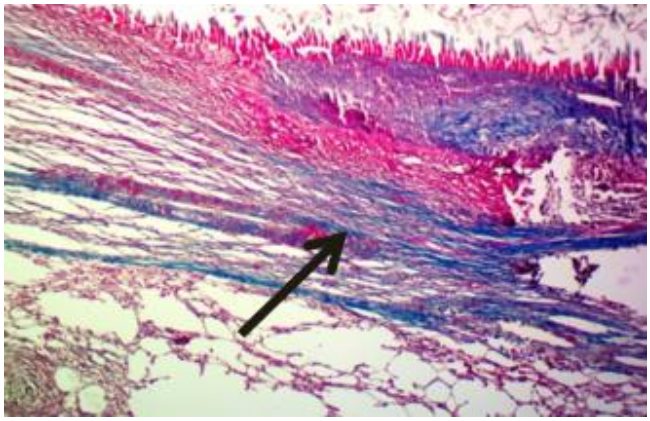

Figure 24: Micrograph of sheep lung showed multi fibrotic foci. Masson's Trichrome, 100x.

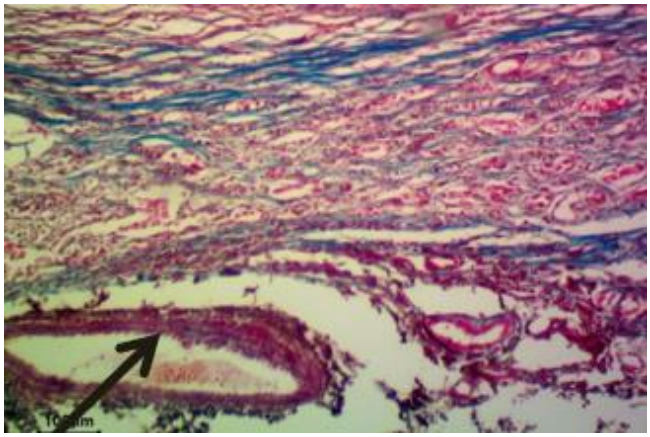

Figure 25: Micrograph of sheep lung showed thinking of alveolar wall and involve bronchiolar lumen. Masson Trichrome. 100x.

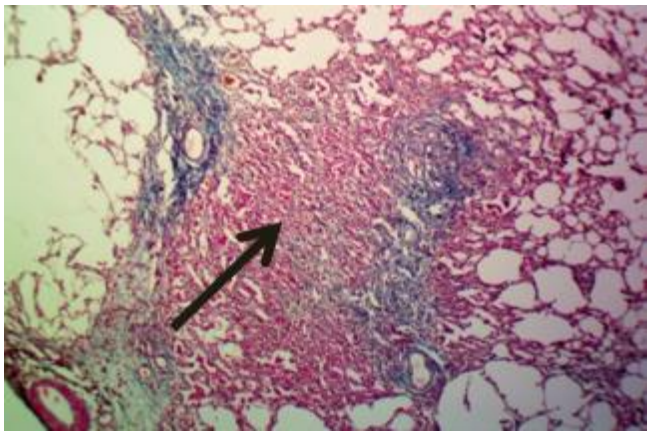

Figure 26: Micrograph of sheep lung showed masons bodies are distinguish as proliferation of fibroblast embedded in collagen and has coalescing bundle as papillary like lesion. Masson's Trichrome, 100x.

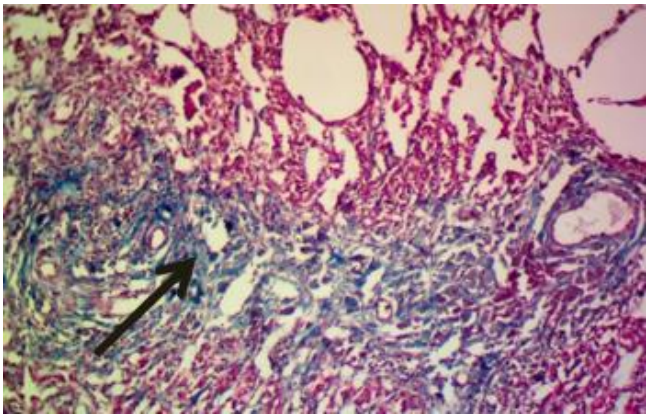

Figure 27: Micrograph of sheep lung showed proliferation of Masson's bodies as embedded in collagen and has coalescing bundle as papillary like lesion. Masson's Trichrome, 100x.

Discussion

Disorder of the respiratory system are still considering a big problem facing animals' livestock (12,13), the significant important of sheep respiratory system diseases depended on prevalence, animal value and impact on animal production (14,15). Respiratory disorder are caused by a complicated, multifactorial illness in which viral, fungal, bacterial and mycoplasma infection work in conjunction with stress factor associated with transit, commingling and weaning (16,17), at the current study lung inflammation showed high incidences 33% and divided into three section according to the gross and histopathological examination (purulent bronchopneumonia, fibrous bronchopneumonia and nodular inflammation) this attributed to the lack of weeds and pasturing in hot dry summer which predisposes the animal to nutritional stress, theses result agreement with Dar *et al.* (18), Abdalla (19) and Naccache *et al.* (20). The anatomical structures of lung, shortness of the respiratory passage in sheep and their direct branching to the front cranial lobe may play a major role in increase the incidence of lung infection (21,22).

Infection due to parasitic represented by hydatid cysts with 30% of percentage were also demonstrate at the present study and these contributed to importation of animals from different regions (23,24), as well city environmental pollution plays a major role in occurrences of parasitic infection. large percentage of loose doge that indirect contact with other animals (sheep, cow and goat) without any restriction, play role in transmission of larva, cysts and egg of tape worm such as Echinococcus spp which spreads the infection and polluted the soil (25-28)

The histopatholgical features of bronchi showed the present of epithelial cell hyperplasia and disturbances of growth inside the epithelial cell lining the mucus gland 12% this could be due to the continuous irritation of the epithelial cell with inflammatory factor (injury, irritation and infection) (29,30).

Vascular changes represented by congestion, newly formed thrombus and bleeding of the blood vessels within the alveoli and bronchi was found in 10% of the affected cases and these may contribute to trauma-induced contusions, neoplasia specially hemangiosarcoma and bacterial infection (31,32). Furthermore, degeneration and necrosis of the epithelial cells lining the alveoli and the cartilage was observed in 6% and this might be due to the ischemia which occurs due to vascular damage (33). The necrotic area often surrounded by edge of elongated spindle cell known as (swirling macrophages or oat cell) (34-36), and the formation of oat cell in bronchopneumonia has been contributed to the release of toxin from Gram negative bacterial infection which demonstrated in most cases of fibrins bronchopneumonia (37,38). In addition to these finding there was also deposition of calcium salt due to low pH that resulted from chronic infection and parasitic disease (11,39), hemosiderin pigment, represented by the presence of copper-colored granules inside the tissue due to present of hemorrhage inside lung tissue (35,39). The presence of neutral mucopolysaccharides inside lung may be contributed to the bronchi and alveoli inflammation or due to prolonged action of irritative insults (40).

Conclusion

From our observation we concluded that lung lesion can consider s an important ovine disease affected on activity, function of the respiratory system and leading to economic losses. Prevention is important such as animal monitoring, medical treatment and periodicals exam for all herds.

Acknowledgments

The researchers are grateful to the University of Mosul, Collage of Veterinary Medicine for their support to achieve this work

Conflict of interest

The author declines that there is no conflict of interest

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الآفات المرضية النسجية لرئة الأغنام التي ترعى على مخلفات المناطق

هديل باسم ذنون[1]، إيناس شيت مصطفى[1]، أصيل محمد رحاوي[1]، محمد طارق جبر[2]، انتصار خزعل الحمداني[1] و سؤدد محمد الحمداني[1]

[1]فرع الأمراض وأمراض الدواجن، كلية الطب البيطري، جامعة الموصل، الموصل، [2]مركز الدنا العدلي للبحث والتدريب، جامعة النهرين، بغداد، العراق

الخلاصة

تعتبر اضطرابات الجهاز التنفسي شائعة الحدوث في الأغنام، ويصعب في كثير من الأحيان اكتشافها بسهولة في الحيوانات الحية الى أن تتطور وتمثل مشكلة خطيرة، ولغرض تحديد الآفات الرئوية للأغنام في مدينة الموصل والتي ترعى على مخلفات المناطق والمجزورة في محلات القصابة، تم فحص ٥٠ عينة لذكور وإناث رئات الأغنام والتي تراوحت أعمارها بين ١-٥ سنوات وللفترة من شهر أذار الى شهر كانون الأول خلال سنة ٢٠٢١ حيث تم جمع العينات المصابة لأجل إجراء الفحص النسيجي لها، أظهرت نتائج الدراسة وجود آفات عيانيه ومجهرية عديدة حيث تمثلت الآفات العيانية بوجود الالتهابات الرئوية في الفص الأيمن واحتقانات ونزف بالإضافة الى أحجام مختلفة من العقيدات أما الآفات النسجية فتمثلت بوجود الالتهابات الرئوية القيحية وبنسبة ١٢,١٪، فيما كانت نسبة اضطرابات الدوران ٣,٥٪، أما الإصابات الطفيلية فكانت نسبتها ١٠٪ كما أظهرت عينات أخرى وجود اضطرابات في النمو وبنسبة ٧,١٪، أما نسبة النخر والتنكس الرئوي فكانت ٢,٠٪ محاطة بخلايا دفاعية تسمى خلايا الشوفان، بالإضافة الى ترسب أملاح الكالسيوم وصبغة الهيموسدرين ويعتبر الالتهاب الرئوي وأجسام ماسون من اكثر الآفات التي تم الكشف عنها في الأغنام التي رعت على مخلفات المناطق حيث تراوحت شدتها من الخفيفة الى المتوسطة والشديدة الضراوة. نستنتج من الدراسة الحالية أن الآفات الرئوية تمثل مشكلة كبيرة وخطيرة في صناعة المواشي الحيوانية وتقود الى حدوث الخسائر الاقتصادية في إنتاج الحليب والصوف واللحوم للحيوانات ونوصي بإجراء المزيد من الدراسات لتحديد نوع العامل المسبب للآفات الرئوية في الأغنام سواء أكانت عوامل فطرية أم بكتريه أم فايروسية.