



## Pathological study of the relation between the incidences of claws lesions and carpo-digital joint capsule lesions in front limbs of sheep slaughtered in Mosul

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### Abstract

For detecting a possible relation between claws lesions and the lesions of carpo-digital joint capsule, the nearest joint to the claws, with an importance in diagnosing the exact cause of lameness in sheep resulting to be brought for casualty slaughter. For that goal 128 front limb were collected from slaughtered sheep at Mosul abattoir for period from 5-25/2/2013, the gross examination was made for observing the claws lesions and the synovial capsule were harvested, preserved and prepared for histopathologic examination, then examined microscopically with recording lesions. the correlation coefficient values were estimated between claws affection and joint capsular lesions at the same limb. Results demonstrated affection of 83 limbs with claws lesions as foot rot and erosions at incidence ratio 64.84%. The histopathological examination revealed a chronic type of changes including Fibroplasia in both synovium and tunica densa at incidences 8.59% and 6.25% respectively, Foci of hyperplastic synovial lining at 4.68% of samples, adipose tissue marbleizing synovium at 7.03% of the examined sections. Mild degree of chronic synovitis was observed within 10.93% of total samples, connective tissue metaplasia was evident at 3.9% of sections at tunica densa layer, the arteriolar musculature showed vacuolation at 18.75% of samples. The correlation coefficient value was significantly positive between occurrence of foot rot disease and the incidence of chronic synovitis in carpo-digital joint capsule which can be considered as the most important conclusion in the study.

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### Introduction

The claws affection in sheep, Foot rot or so called infectious pododermatitis is one of the infectious diseases causes economic losses in small ruminants (1), the gram negative bacteria *Dichelobacter nodusus* is considered the main causative agent of it (2), other organisms as *Fusobacterium necrophorum* plays an important role in progression of pathogenesis and formation of lesions (3). The lesions include an exudative dermatitis with necrosis to the epidermal tissue in the interdigital skin, coronary bands and claw matrix and sometimes detachment of the claw from its underlying soft tissues (4), this affection negatively affect

animal production through lameness, weight loss, fertility reduction and drop in wool quality (5). Lameness is the first and most noticeable clinical sign that vary from mild to severe with redness and swelling of the interdigital skin and spread apart toes (6). The disease known to be predisposed by moist soil and muddy ground in moist and rainy weather that provide suitable environment for the causative agents to stay viable (7). A similar well known affection to the foot is Dermatophilosis or strawberry foot rot, caused by the gram positive *Dermatophilus congolensis* and characterized by dermatitis, pustules and hyperkeratosis in coronary band, it was recorded and described by (8) in Diwanya province in Iraq. Although foot affections are the most observable cause

of inducing lameness in sheep, Arthritis, the inflammation of joints appears to record high incidences in these animals through affecting one or two joints in the limb including knee joint, carpal or tarsal joints with swelling, the younger animals or lambs are the most susceptible than adults according to (9), he reported that the most common causative agent that can be isolated and cultured from the inflamed joint are *Streptococci*, *E.coli*, *Erysipelothrix rhusiopathiae* and *Fusobacterium necrophorum*, describing the inflamed joint as swollen, hot and painful. *Erysipelothrix rhusiopathiae* was mentioned by (10) as the major cause of arthritis in Australian lambs. *Mycoplasma agalactia* reported to develop arthritis in sheep were monitored in Iran with in two studies, in Qom (11) and Khusestan province (12). In Mosul city in Iraq (13) Isolated *Streptococcus dysagalactiae*, *Arkanobacterium pyogenes* and *Pseudomonas aeruginosa* from synovial fluid aspirates from knee joint of sheep showing lameness and diagnosed as suppurative arthritis. Viruses less commonly induce arthritis in sheep but it is reported as Lentiviruses (14), Most of the viral diseases inducing lameness in sheep affect claws as Foot and Mouth disease and blue tongue (15). Lameness also can appear as a complicated case containing both foot lesions and arthritis on the same affected limb. It stated by (16) that Microorganisms can lodge in joints through hematogenous dissemination or by contiguous spread from osteomyelitis or a soft tissue abscess. The joint capsule is an important component of the joint and vital for its function that mainly composed from dense fibrous tissue, it is attached to the bone and forms sleeve around joint, the capsule may exposed to an injurious agent through rheumatoid arthritis or osteoarthritis or aging leaving variable changes on it (17).

For that purpose this study targeting to reveal lesions of synovial capsule of carpo-digital joint and their relation with foot affections in front limbs in sheep slaughtered in Mosul abattoir.

## Materials and methods

### Sample collection

The collection of 128 front limbs of slaughtered sheep were made at Mosul abattoir in the period from 5-25/2/2013, they directly packed, numbered and transformed to the laboratory of pathology were examined for gross foot-rot lesions, then the skin dissected over carpo-digital joint and the capsule were excised gently and preserved in 10% neutral buffered formalin.

### Histological preparations and examination

Samples were trimmed, dehydrated, Cleared, impregnated and blocked in paraffin wax, then sectioned to 5 micrometer sections and stained with hematoxylin and eosin, dehydrated and mounted with DPX (18-22). The histopathological examination were done under light microscope with photographing using AMSCOPE digital camera.

## Statistical analysis

Lesions detected by histopathological examinations, percentage of incidences were estimated, the 0 and 1 grading system were used to express lesion occurrence in each synovial capsule sample (19), also to express occurrence of foot-rot lesion or not. Data were analyzed using SPSS version 19 to determine the correlation coefficient values by 2-tailed Pearson correlation under significance at  $P \leq 0.01$  (23).

## Results

Gross examination revealed foot rot lesions at 83 limbs from total 128 limb examined at incidence ratio 64.84% (Figure 1). The histopathological examination of the carpo-digital joint capsule sections explored a pathological changes recognized from normal histological features of the joint capsule (Figure 2/A). Those changes were represented by mild chronic type of changes, these were included Fibroplasia in both synovium and tunica densa at incidences 8.59% and 6.25% respectively. (Figure 2/ B and C). Foci of hyperplastic synovial lining were observed at 4.68% of samples (Figure 2/D). Fat or adipose tissue marbleizing synovium was noticeable at 7.03% of the examined sections (Figure 3/A). Mild degree of chronic synovitis observed at 14 sections with an incidence rate 10.93% of total samples (Figure 3/B, C and D). Fibrocartilaginous connective tissue metaplasia were evident at 3.9% of capsule sections at tunica densa layer (Figure 4/A). The capsular arteriolar musculature showed vacuolation at 18.75% of samples to be the most noticeable change observed (Figure 4/B) (Table 1). The correlation coefficient value was significantly positive ( $r=0.235$ ) between occurrence of foot rot disease and incidence of chronic synovitis (Table 1).

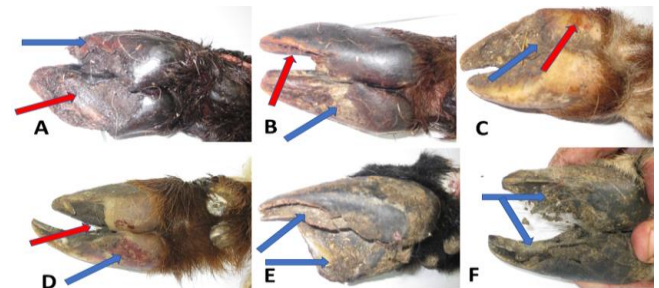


Figure 1: The gross appearance of ovine claws affected with foot-rot showing: A- planter (Red arrow) and anterior (Blue arrow) erosive lesions. B- posterior planter (Blue arrow) and anterior medial (Red arrow) erosive lesions. C- planter erosions (Blue arrow) and erosive lesions on the heel beneath the coronary band (Red arrow). D-Planter erosions on the heel (Blue arrow) and anterior bilateral medial erosions (Red arrow). E-left planter erosion and right anterior sloughing (Blue arrows). F-Anterior bilateral deep planter erosion with dry suppurative exudate (Blue arrows).

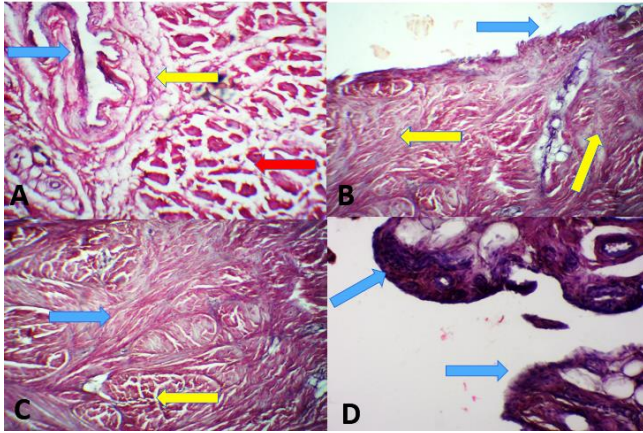


Figure 2: Sections in carpo-digital joint capsule showing: A- Normal morphology of synovial lining (Blue arrow). Synovial layer (Yellow arrow) and collagen bundles in tunica densa (Red arrow).40X. B- Focal hyperplasia of synovial lining (Blue arrow) and Fibroplasia of synovium and tunica densa (Yellow arrow).40X. C- Fibroplasia of the fibrous septa (Blue arrow) between collagen bundles (Yellow arrow) of tunica densa.100X D- Focal hyperplasia of synovial lining cells (Blue arrow).100X. Staining H&E for all images.

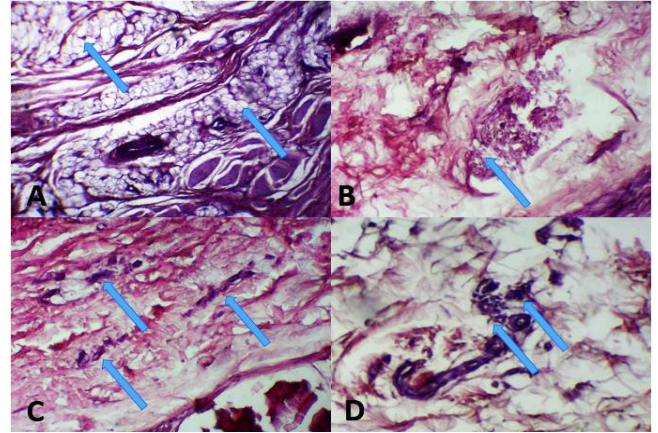


Figure 3: Sections in carpo-digital joint capsule showing A- Fat tissue marbling synovium (Blue arrows).40X. B- Fibrotic focus of chronic inflammation with lymphocyte, fibroblast and collagen under synovial lining (Blue arrows).100X. C- infiltrations of lymphocyte and plasma cells with in synovium (Blue arrows). 100X. D- Mild chronic inflammatory reaction with infiltration of lymphocyte and plasma cells from capillaries of synovium (Blue arrows).100X. Staining H&E for all images.

Table 1: Lesions of carpo-digital joint capsule, number of cases, Incidences and correlation coefficient values between lesions of capsule and foot-rot occurrence in the front limb

Microscopic lesion	No. samples affected	% lesions incidence	CV with foot rot
Fibroplasia in synovium	11	8.59%	0.048
Fibroplasia of tunica densa	8	6.25%	0.095
Focal hyperplasia of synovial lining	6	4.68%	-0.002
Fat tissue marblesation in synovium	9	7.03%	-0.107
Chronic synovitis	14	10.93%	0.235**
Connective tissue metaplasia	5	3.90%	0.141
Vacuolation of Arteriolar musculature	24	18.75%	0.037

Total number of samples=128, samples affected with Foot-rot=83, significant correlation (\*) between lesion of capsule and Foot-rot at  $P \leq 0.01$ .

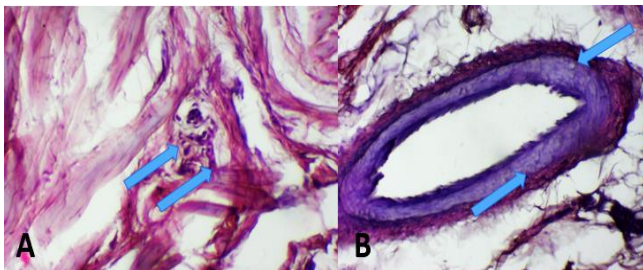


Figure 4: Sections in carpo-digital joint capsule showing A- Islet of metaplastic changes showing chondroblasts and cartilage matrix within tunica densa layer (Blue arrows). 100X. B- Vacuolation in the musculature of arteriole in the synovium (Blue arrows).100X. Staining H&E for all images.

## Discussion

As a general analytical view to the results, a few types of lesions have been found in the wall of synovial capsule. The presence of fibroplasias as foci in both synovium and tunica densa layers mostly reflects previous traumatic lesions and previous inflammatory affections (24), also the abnormal motion of the limb may be related to lameness stretches the fibrous joint capsule and laxity of the joint resulting in developing fibroplasias in the joint capsule and formation of osteophytes, the fibroplasia of joint capsule also mentioned in the pathogenesis of frozen shoulder which confirmed by histological staining of collagen I and II and the immunohistochemical detection of vimentin and conclusion were made that an incidence of fibroplasia at the anterior and



posterior structures of the capsule occur or in entire joint capsule (25,26), such lameness can be provided by foot rot lesions as we noticed in results at a weak correlation link 0.048. The fibrosis of the joint capsule reduces the capsular volume and may result from trauma, fractures and tendon injuries (27). Focal hyperplasia of the synovial lining cells was observed at 4.68% of samples, although there was no significant correlation link with foot rot incidence but the presence of hyperplastic foci may be associated with the release of growth factors or mediators locally through occurrence of synovitis, this particular change was described accompanying chronic and rheumatoid arthritis were synovial lining thickened to three or five layers of cells (28). The normal synovial membrane mostly appeared as acellular structure composed of one or two cell thickness intimal layer and synovial sublining layer containing the blood and lymphatic vessels, fat cells, fibroblasts with few lymphocytes and macrophages (29). Those membranes anatomically classified according to the composition of the subintimal layer and content of fibrous, areolar and adipose tissues, this may be variable according to the position of the joint in the body, the subintima may contain hyaline or fibrocartilage tissues and even osteoblasts (30). Based to those information's and microscopic examination the marbleisation with fat tissue with in subintimal synovium at 7.03% of samples reflects an intense presence of adipocytes that may convey general obesity of the animal or related to the age progression that younger animals contain much more fat in their synovium. The same explanation manages observation of metaplastic changes to chondroblasts with in subintimal synovium at 3.09% of samples that increases with senility or age progression. The significant correlation link 0.235 between presence of chronic inflammatory foci insynovium and incidence of foot rot lesions in claws can be managed under two explanations, Either extension of the infection from the claw to the adjacent joints through venous drainage and lymphatics as mentioned by (13) who isolated multiple pathogenic microorganisms from synovial fluid of knee joints of lamed sheep, or the most probable examination that the abnormal motion of the affected limb may induce the physical damage and stretching the synovium of the joint (25), causing inflammatory response that involve cellular infiltrates, cytokine production, enzymes and relevant proteins secretion, increase synovial lining layer thickness, vascular and lymphatic changes and the production of metalloproteinases that stimulates osteoclast formation (30).

## Conclusion

The study demonstrated presence of multiple mild to moderate changes in the capsule of the carpo-digital joint with a positive correlation link between the occurrence of foot rot and the development of chronic synovitis in the carpo-digital joint capsule in sheep.

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## Conflict of interest

No conflict.

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## دراسة مرضية للعلاقة بين حدوث آفات الظلف وآفات المفصل المشطي السلامي في القوائم الأمامية للأغنام الواردة إلى مجزرة الموصل

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### الخلاصة

للتحري عن احتمال وجود ارتباط بين آفات الظلف وآفات المفصل المشطي السلامي، المفصل الأقرب للظلف، في الأغنام، أجريت هذه الدراسة لما لها من أهمية في تشخيص أسباب حدوث العرج في هذه الحيوانات مؤدياً إلى جلبها للجزر اضطرارياً، من أجل هذا الهدف تم جمع ١٢٨ قائمة أمامية من الأغنام المجزرة في مجزرة الموصل للفترة من ٢٠١٣/٢/٢٥-٥ واجري عليها الفحص العياني لملاحظة آفات الظلف و تم استئصال المحفظة الزلالية للمفصل المشطي السلامي وأجريت عليها التحضيرات النسيجية والفحص المرضي النسيجي وسجلت الآفات المرضية وتم إيجاد معامل الارتباط بين إصابة الظلف وآفات المحفظة المفصالية في نفس القائمة. أظهرت النتائج إصابة ٨٣ قائمة بآفات الظلف متمثلة بتآكل وتعفن الظلف بنسبة إصابة ٦٤,٨٤% في حين أظهر الفحص المرضي النسيجي للمحفظة المفصالية وجود تغيرات مزمنة تمثلت بفرط التنسج الليفي في الطبقتين المصلية والليفية للمحفظة بالنسبتين ٨,٥٩% و ٦,٢٥% على التوالي، يؤر من فرط التنسج لخلايا البطانة المصلية في ٤,٦٨% من العينات، تخلل الأنسجة الدهنية في الطبقة المصلية في ٧,٠٣% من العينات، التهاب المحفظة المصلية المزمن الطفيف لوحظ عند ١٠,٩٣% من إجمالي العينات فضلاً عن الحؤول للنسيج الضام بنسبة ٣,٩% و تفجي خلايا الطبقة العضلية في شريينات المحفظة عند ١٨,٧٥% من إجمالي العينات المفحوصة. سجلت النتائج وجود علاقة ارتباط إيجابية بين حدوث آفات تعفن الظلف في القدم وظهور التهاب المحفظة المصلية المزمن الطفيف في المفصل المشطي السلامي والذي يمكن اعتباره الاستنتاج الأهم في هذه الدراسة.