



## RESEARCH ARTICLE

### Suppurative Injection Site lesion In Cattle: Clinico-biochemical, Ultrasonographic and histopathological examinations

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#### Abstract

This study was carried out on a total of 40 animals (13 female cows, 14 cow bull and 13 cow calves of both sexes) with an age ranged from 1- 5 years. Animals were admitted to Zagazig Veterinary Teaching Hospital, Faculty of Veterinary Medicine, Zagazig University, Egypt with a history of injection site lesions. A history of injection at the site lesion was noted with swelling ranging from 5 to 15 cm in diameter, others were showing fluctuating swelling. Various clinical signs as abnormality in gait, painful condition on thigh, gluteal and neck muscle were recorded with respect to the site of lesion. Diagnosis was based on history, Clinico-biochemical findings, needle aspiration, ultrasound and histopathology of biopsy samples. Upon diagnosis, animals were classified into three groups; group I (Control group, n= 10), group II (Muscular abscess group, n= 17), and group III (Suppurative myositis group n= 13). Hematological findings revealed mild anemia, leucocytosis and neutrophilia in muscular abscess and suppurative myositis groups, while biochemical findings included hyperglobulinemia, elevation of serum muscular enzymes; aspartate aminotransferase (AST), creatine kinase (CK) and lactate dehydrogenase (LDH) as well as a significant increase in C-reactive protein (CRP) and fibrinogen in both diseased groups in comparison to control group. Results of ultrasonographic examination in muscular abscesses group revealed homogenous echogenic content surrounded with hyperechoic capsule which is embedded in the muscle and an increase in the echogenicity of the muscle in suppurative myositis group. Histopathological examination of the biopsy specimen from the affected muscle revealed fibrinopurulent inflammation with aggregation of dead and live neutrophils, hyaline degeneration of muscle and muscle edema. In conclusion, ultrasonography provides an accurate, non-invasive and fast technique for diagnosis injection site lesions. In addition CRP and fibrinogen values in association with total white cell count were very useful in the diagnosis and prognosis.

**Keywords:** Myositis, muscle, abscess, enzymes, CRP, cattle.

#### Introduction

Intra-muscular injection is one of the most common routes of injection performed in cattle because it is easy, fast and safe. The most common sites of intramuscular injection are thigh, gluteal muscle and neck muscle. There is wide variety of bacteria on cattle skin specially *Staphylococcus* spp. which may lead to abscess formation [1]. Faulty injection can

cause trauma to tissues resulting in an inflammatory reactions and potential injection site lesions as abscesses, suppurative myositis (diffuse muscle infection without abscess formation) and in severe cases cause necrosis of the tissue [2].

Abscesses in the injection sites "gluteal or thigh or neck muscle" can result from infection

either from unsterile needle or from animal skin associated. When myositis occurs, it is often due to direct or indirect trauma, surgery, or vascular insufficiency. Myositis appeared as localized or diffuse muscle pain, tenderness on palpation or with movement, swelling, and/or weakness [3].

Diseases of muscle as muscular abscesses and suppurative myositis need thorough clinical examination depending on palpation, needle aspiration, biopsy and ultrasonographic examination. Differential diagnosis of both diseases is difficult as both share the same clinical signs. The clinical use of ultrasound to assess the diseases of muscle is helpful as ultrasonography is considered a suitable technique to provide accurate details including the character, extent, capsulation of the examined lesion [4].

Limited numbers of laboratory tests are used to help the veterinarian to diagnose bovine intramuscular injection lesions. Leucocytosis and neutrophilia are considered the most important hematological indicators for inflammatory reactions and infection. Clinically, the changes in white blood cell count are of great importance [5]. C-reactive protein is an important element of the innate defense system which plays a role in pathogen response as in bacterial infection [6]. Serum level of Fibrinogen can express the degree of tissue damage in cattle [7]. Fibrinogen is one of plasma proteins; its normal range in adult bovine plasma is 0.3-0.7 gm/dl. Fibrinogen is used as an indicator of inflammation, bacterial infection or surgical trauma in cattle. It increases in inflammatory conditions as pericarditis, endocarditis, peritonitis, pneumonia, and nephritis [8, 9]. Skeletal muscle is known to contain isozymes of creatine kinase, lactate dehydrogenase (LDH) and aspartate aminotransferase (AST) which are released into the blood stream following muscle affection [10]. Muscle enzymes are sensitive indicators of muscle damage. The levels of aspartate aminotransferase, alanine aminotransferase and lactate dehydrogenase increased in serum in myopathic diseases. Creatine kinase (CK) level is thought to be the most sensitive laboratory test related to skeletal muscle damage [11].

Therefore, the aim of the current study is to give a highlight on the clinical, haematobiochemical and ultrasonographic examination as field techniques for diagnosis of injection site lesion in cattle.

## Materials and Methods

### *Ethical approve*

All procedures used in the present study were approved by the Committee of Animal Welfare and Research Ethics, Faculty of Veterinary Medicine, Zagazig University, Egypt.

### *Animals*

This study was carried on 40 animals (13 female cows, 14 cow bull, 9 female cow calves and 4 male cow calves) aged between 1-5 years, and weighing between 300-500 kg. Animals were admitted to Zagazig Veterinary Teaching Hospital, Egypt, between January 2017 and December 2018 with a history of injection site lesions. Problem was identified on the basis of history of injection with local signs; swelling, hotness and pain. Swellings ranging from 5 to 15 cm in diameter, others were showing fluctuating swelling. General signs are variable; abnormality in gait, with painful condition on thigh, gluteal and neck muscle. Diagnosis was based on the history, Clinico-biochemical findings, aspiration, ultrasound and histopathology of biopsy samples. Upon diagnosis, Animals were classified into three groups; group I (Control group, n= 10), group II (Muscular abscess, group n= 17, 10 females and 7 males), and group III (suppurative myositis, group n= 13, 7 females and 6 males).

### *Clinical examination*

All animals were subjected to thorough clinical examination with vital signs monitoring; measuring of respiration rate, heart rate and body temperature, Also, closed physical examination including inspection and palpation of the affected muscle was applied to localize the lesion as described by Dirksen *et al.* [12].

### *Blood Sampling and Biochemical Assays*

Two blood samples were collected from each animal via jugular vein puncture. First blood samples were collected in EDTA

containing tubes for haematological analysis; Total Erythrocytic Count (TEC), Haemoglobin (Hb), Packed Cell Volume (PCV), Total Leucocytic Count (TLC). Second blood samples without EDTA were collected for serum separation according to Kaneko *et al.* [13]. Serum total protein and albumin were estimated according to the method described by Henary *et al.* [14]. Muscle enzymes (AST, CK and LDH) were estimated by standard procedures using (Diagnostic Zrt. Commercial kits) which were provided by Egyptian Company for Biotechnology, Cairo, Egypt, and the reading was taken by spectrophotometer. C- reactive protein (CRP) was detected in serum by electrophoresis as described by Lee *et al.* [15]. Fibrinogen (Fb) was estimated in plasma samples using commercially available bovine kits according to Orro *et al.* [16].

#### **Ultrasonographic examination**

Ultrasonographic examination of the lesions in thigh, gluteal and neck muscles were applied using ultrasound machine (SonoScape A5V, China) connected to linear transducer with 6 MHz frequency as described by Zaidman and van Alfen [17]. Animals were examined in standing position. Examined areas were prepared by clipping and shaving of the hair, followed by application of coupling gel.

#### **Microbial culture and isolation**

In muscular abscess group pus samples were aspirated by sterile syringe using ultrasound guided technique, and in suppurative myositis sterile swabs were used. Microbial examination of pus forming bacteria was applied; pus samples or swabs were enriched on nutrient broth incubated at 37° C for 24 hours. Then cultured on nutrient agar, blood agar and Macconkey agar and incubated aerobically at 37°C for 24-48 hours. The growing colonies were picked up and subcultured. The isolates were identified according to the colony morphology, pigment production, Gram's stain and biochemical tests [18].

#### **Antibiotic susceptibility test**

The susceptibility profile of isolates to different anti-microbial agents was done by disc diffusion method by commercially

available filter paper disks, each containing a defined concentration of a specific antibiotic. The characterization of strains as sensitive, intermediate or resistant was based on the size of inhibition zones surrounding the wells [19].

#### **Surgical interference**

Cases with deep non sizeable collection which, cannot be accessed easily (suppurative myositis) were treated with appropriate antibiotic every 12 hours through intramuscular route for 48- 72 hours (Pen and strep® Norbrook company. N. Ireland (procaine penicillin 200 mg and Dihydro streptomycin 250 mg in dose of 1mL /25 kg body weight) with topical application of Ichthammol ointment 20% (Ichtyol® ALAKSA Co for chemicals. Egypt), and then drained efficiently. Cases with large superficial sizable-swellings (abscesses) were managed by efficient drainage after aseptic preparation. Then swelling wall was painted with Povidone-iodine solution 10% (Betadine®-The Nile Co for pharmaceutical and chemical industries, Cairo –A.R.E) after radical debridement to remove all dead tissues. All animals were forced for complete rest with daily dressing of the wound and injected with antibiotic (Pen and strep® Norbrook company. N. Ireland (procaine penicillin 200 mg and Dihydrostreptomycin 250 mg in dose of 1mL /25 kg body weight) for 5 successive days.

#### **Histopathological examination**

Muscle biopsy was taken from the affected area under the effect of Xylazine hydrochloride (Xylaject, Adwia Company El Obour city, Cairo, Egypt) in a dose of 0.05 mg/kg body weight intramuscular in standing position [20]. The specimens from muscles were fixed in 10% neutral buffered formalin solution, dehydrated in gradual alcohol (70-100%), cleared in xylene and embedded in paraffin. Five microns thickness paraffin sections were prepared and stained with hematoxylin and eosin (HE) dyes then examined microscopically [21].

#### **Statistical analysis**

Analysis of variance (ANOVA) with Duncan's post-hoc test was used to determine the significance of difference between the groups. Data were expressed as means ±

standard error (SE). P-value ( $p < 0.05$ ) was selected as the criterion for statistical significance.

## Results

### Clinical findings

All the clinical findings of the examined animals were summarized in Table (1). Weakness is common sign in all affected animals. Lameness and ataxia when the thigh or gluteal muscles are affected and discomfort while sitting and standing. General signs

included slight rise in body temperature ( $38.87 \pm 0.05$  in group II and  $38.91 \pm 0.05$  in group III) compared to group I ( $38.34 \pm 0.04$ ). A significant rise in heart and respiratory rates was recorded. Heart rates were  $64.30 \pm 2.03$ ,  $81.30 \pm 1.80$  and  $81.90 \pm 0.90$  in groups I, II and III, correspondingly. However the respective respiratory rates were  $15.40 \pm 0.7$ ,  $27.40 \pm 0.91$  and  $28.90 \pm 0.57$ . In groups II and III, anorexia was recorded 13 animals compared to normal appetite in 17 animals.

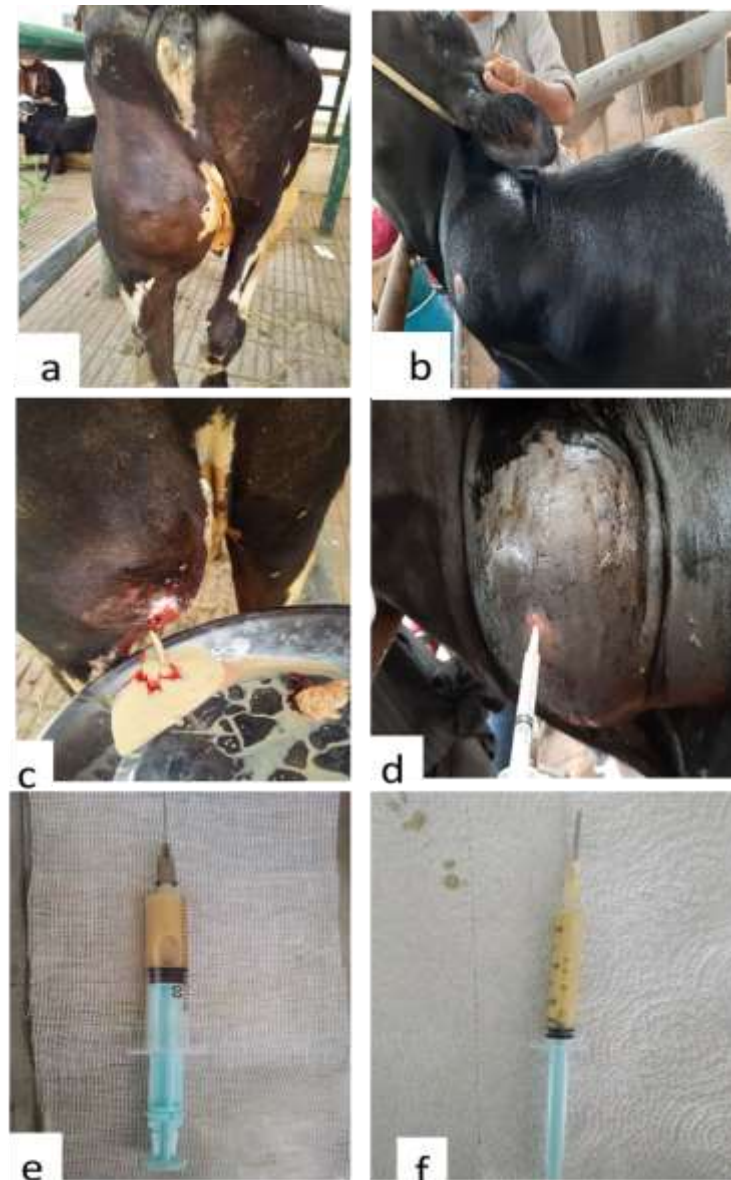
**Table 1: Clinical findings in muscular abscess and suppurative myositis in affected cattle groups in comparison to control animals**

Clinical Findings	"Control group"	Diseased groups	
	Group I (n=10)	Group II "muscular abscesses" (n=17)	Group III "suppurative myositis" (n=13)
<b>Vital signs*</b>			
Heart rate/min.	$64.30 \pm 2.03^b$	$81.30 \pm 1.80^a$	$81.90 \pm 0.90^a$
Respiration rate/min.	$15.40 \pm 0.76^b$	$27.40 \pm 0.91^a$	$28.90 \pm 0.57^a$
Rectal temp. (C°)	$38.34 \pm 0.04b$	$38.87 \pm 0.05a$	$38.91 \pm 0.05a$
<b>General signs</b>			
Alertness	Alert	Alert (15), depressed (2)	Alert (12), depressed (1)
Appetite	Good	Good (13), inappetence (4)	Good (9), inappetence(4)
Abnormal posture and gait	Absent	Present (13)	Present (10)
<b>Site of lesions</b>			
Neck muscle	----	5 of 17	6 of 13
Gluteal muscle	----	3 of 17	2 of 13
Thigh muscle	----	9 of 17	5 of 13
<b>Local signs</b>			
Swelling of the region	Absent	Circumscribed swelling varied from 5-15cm in diameter	diffuse swellings
Tenderness	Absent	Painful (17)	Painful (13)
Local temperature	Normal	Hotness (17)	Hotness (13)

All data having different letters are differ significantly at  $p < 0.01$

Localized and main clinical signs in early stage were swelling of affected muscle, redness, hotness and tenderness. Hair coat covering affected area was dry, rough and erected and the swelling had enlarged gradually. On clinical examination, a fluctuating mass "presence of pus" was palpated in muscular abscess group, while only swelling was detected in suppurative myositis group. These swellings ranged from few centimeters to large and huge ones (Figure1, a and b). Abscesses may rupture and drain to the surface, or become encapsulated

by fibrous connective tissue and drained by sterile syringe (Figure 1, c, and d).. While in suppurative myositis the exudate extends along the fascial planes and small amount of pus was drained. In advanced stage, signs are more severe with high fever and sometimes toxemia and inflamed skin especially when multiple sites were involved. The course is more progressive and can lead to more complications and less response to treatment. All the treated animals were cured effectively without recurrence.



**Figure 1: Photography of;** large swelling at the thigh region in 4 years old cow (a) and the side of the neck in one year old heifer (b) , note yellowish to greenish creamy aspirated content (c and e) and yellowish creamy aspirated content from the previous swelling (d and f).

The occurrence and frequency distribution of muscular abscesses and suppurative myositis in animals were illustrated in Table. 1

Muscular abscesses and suppurative myositis were more common in thigh muscle; 9 (52.9%) out of 17 cases and 5 (38.5%) out of 13 cases, respectively. While the lowest incidence of both lesions "muscular abscesses and suppurative myositis" was in gluteal muscle, (3 cases from 17, 17.6%) and (2 cases from 13, 15.4%), respectively. In neck muscle, muscular abscess and suppurative myositis were detected in 5/17 cases (29.4%) and 6/13 cases (46.2%), correspondingly.

#### ***Hematological and biochemical findings***

The hemogram values for all groups are provided in Table 2. Significant decreases ( $p < 0.01$ ) in red blood cell counts, packed cell volume and hemoglobin concentration were noted on both diseased group, meanwhile significant increases in white blood cell counts ( $p < 0.05$ ) and neutrophil ( $p < 0.001$ ) were noted on both diseased groups when compared to control group. Alterations in red blood cell counts, packed cell volume and hemoglobin concentration were not significantly different between both diseased groups.

**Table 2: Hemato-biochemical parameters in muscular abscess and suppurative myositis cattle affected groups in comparison to control animals**

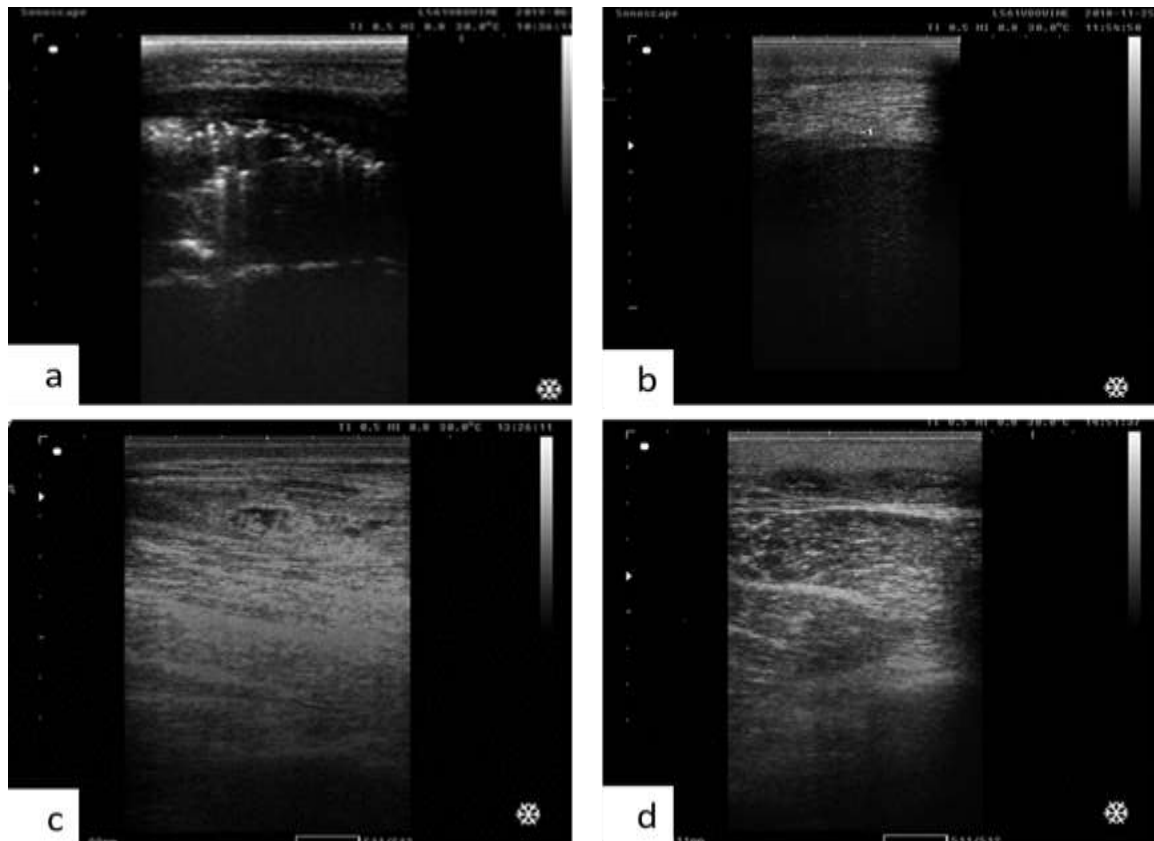
Items	Group I (n=10)	Group II (n=17)	Group III (n=13)	P-value
	Control	Muscular Abscess	Suppurative Myositis	
RBCs ( $10^6/\mu\text{l}$ )	$6.812 \pm 0.267^a$	$5.550 \pm 0.306^b$	$5.084 \pm 0.317^b$	< 0.01
PCV (%)	$33.80 \pm 0.583^a$	$27.80 \pm 1.021^b$	$26.44 \pm 1.103^b$	< 0.01
Hb (g/dl)	$10.40 \pm 0.286^a$	$8.838 \pm 0.280^b$	$8.400 \pm 0.240^b$	< 0.01
WBCs ( $10^3/\mu\text{l}$ )	$9.058 \pm 0.299^b$	$10.98 \pm 0.519^a$	$11.17 \pm 0.697^a$	< 0.05
Neutrophil ( $10^3/\mu\text{l}$ )	$3.160 \pm 0.236^b$	$8.060 \pm 0.507^a$	$8.500 \pm 0.689^a$	< 0.001
Total protein (g/dl)	$7.160 \pm 0.129$	$6.620 \pm 0.442$	$6.540 \pm 0.225$	0.308
Albumin (g/dl)	$3.940 \pm 0.163^a$	$2.820 \pm 0.296^b$	$2.560 \pm 0.117^b$	< 0.01
Globulin (g/dl)	$3.120 \pm 0.162^b$	$3.800 \pm 0.182^a$	$3.720 \pm 0.218^a$	< 0.05
AST (u/l)	$45.85 \pm 3.820^b$	$90.06 \pm 10.50^a$	$92.80 \pm 8.267^a$	< 0.01
LDH (u/l)	$144.0 \pm 18.39^b$	$285.4 \pm 32.52^a$	$322.2 \pm 41.17^a$	< 0.01
CPK (u/l)	$79.20 \pm 4.852^c$	$108.60 \pm 13.12^b$	$202.4 \pm 14.43^a$	< 0.01
CRP (ng/ml)	$0.786 \pm 0.156^b$	$11.06 \pm 4.220^b$	$24.49 \pm 6.260^a$	< 0.01
Fibrinogen (gm/dl)	$0.496 \pm 0.059^b$	$1.250 \pm 0.198^a$	$0.890 \pm 0.062^a$	< 0.01

Concerning the biochemical parameters, the mean values of serum total proteins recorded non-significant decrease ( $P=0.308$ ) in muscular abscesses ( $6.620 \pm 0.442\text{g/dl}$ ) and suppurative myositis group ( $6.540 \pm 0.225\text{g/dl}$ ) in comparison to the control group ( $7.160 \pm 0.129\text{g/dl}$ ), meanwhile serum albumin were significantly lower ( $p<0.01$ ) in both diseased groups than control one. A significant increase ( $p<0.05$ ) in serum globulin levels were recorded in both diseased group. Additionally, a significant increase ( $P < 0.01$ ) in serum muscle enzymes; AST, LDH and CK by two or more folds in both muscular abscesses and suppurative myositis with higher enzymatic activity in suppurative myositis group than muscular abscesses group. Significant increase ( $P < 0.01$ ) in serum CRP about (14 fold,  $11.06 \pm 4.220\text{ng/mL}$ ) in muscular abscess group and (34 fold,  $24.49 \pm 6.260 \text{ng/mL}$ ) in suppurative myositis group compared to control group ( $0.786 \pm 0.156 \text{ng/mL}$ ). The level of fibrinogen was significantly higher ( $P < 0.01$ ) in both diseased groups, (2.5 fold,  $1.250 \pm 0.198 \text{gm/dl}$ ) in muscular abscesses group and (1.7 fold,  $0.890 \pm 0.062 \text{gm/dl}$ ) in suppurative myositis group

in comparison to control group ( $0.496 \pm 0.059 \text{gm/dl}$ ).

#### **Ultrasonographic findings**

Ultrasonographic examination of group II "muscular abscesses" showed hypoechoic to hyperechoic contents with swirling movement surrounded by a thin white hyperechoic capsule embedded inside the muscular tissue. Echogenicity of abscesses contents varied according to its duration and identified as acute and chronic abscesses. Acute abscesses (in 7 of 17 cases 4 females and 3 males) appeared as a homogenous hypoechoic to hyperechoic structure (Figure 2, a and b). Chronic abscesses in 10 (6 females and 4 males) out of 17 cases showed more hypoechoic to anechoic with white hyperechoic septa. Ultrasonography of the group III (Supportive myositis) revealed diffuse inflammation of the muscular tissue with increased echogenicity of the muscle fibers in different grades. The muscle fibers were interspersed with hypoechoic to hyperechoic contents with loose of the normal muscle structure (Figure 2, c and d).



**Figure 2:** Ultrasonographic imaging of; muscular abscess in the thigh region of cattle showing circumscribed hyper echoic capsule with hyoechoic to hyperechogenic content deeply seated between muscle fibers, the small gas particles inside the abscess represented by hyperechoic foci with comet tail arifacts (a and b), and suppurative myositis in the neck region showing different grade of echogenicity and looseness of normal muscular structure (c and d).

### **Gross and microbial findings**

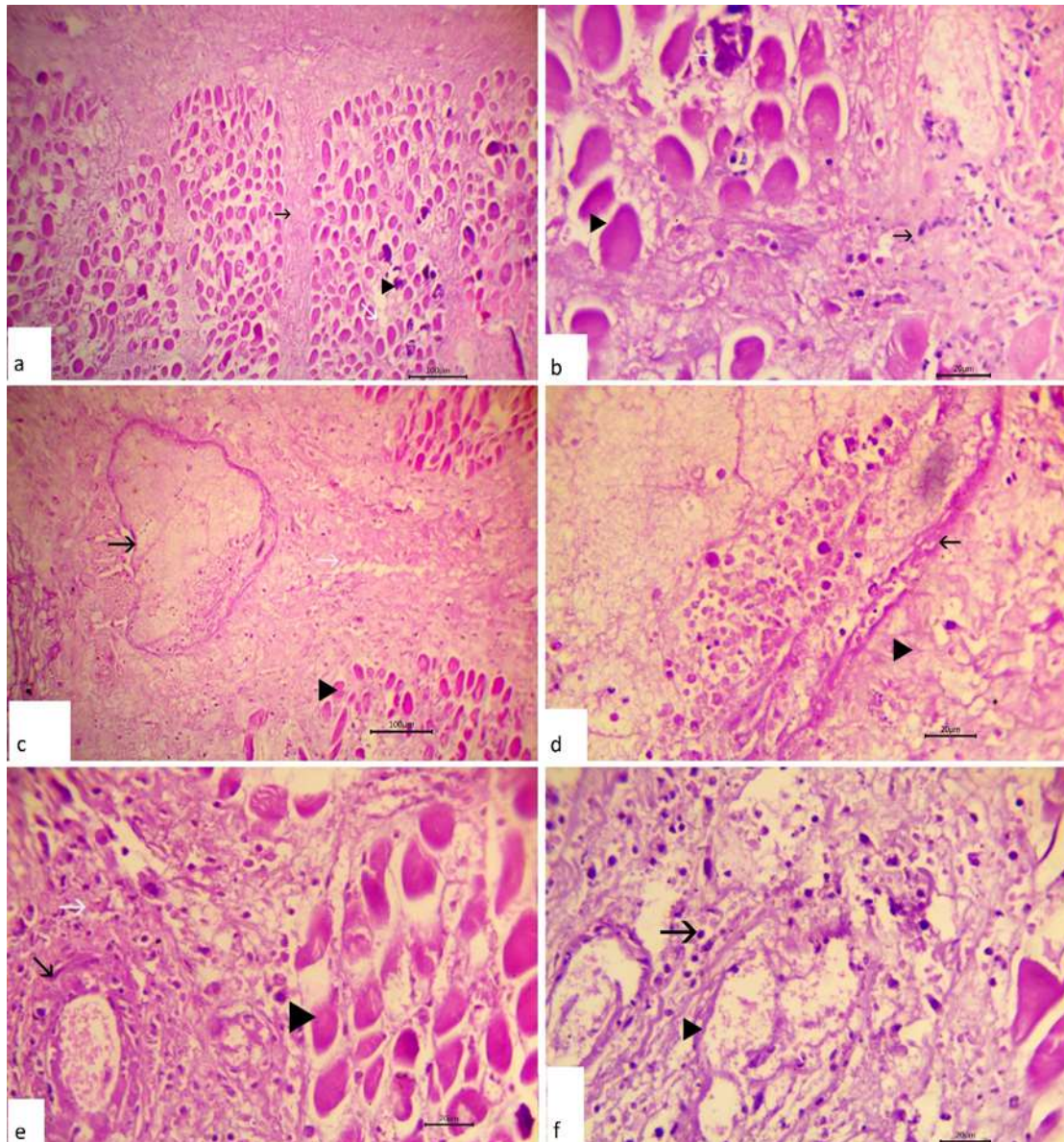
Yellowish to greenish creamy contents were aspirated from the most suppurative lesions (75%) (Figure 1, c and d) and yellowish creamy contents (Figure 1, e & f) from 25% of cases. Microbial culture and isolation of the causative microorganism in muscular abscesses and suppurative myositis revealed combination of *Staphylococcus aureus* and *Streptococcus* spp. in most cases (95%) and *Corynebacterium* spp. in 5% of cases (untabulated data).

### **Antibiotic susceptibility test**

The isolated microorganisms were highly sensitive to Penicillin (++++), Amoxicillin (++++), Gentamycin (++++), Ampiclox (+++) and Cloxacillin (++) (untabulated data).

### **Histopathological results**

The muscle showed fibrinopurulent inflammation represented by faint eosinophilic fibrin thread with aggregation of dead and live neutrophils and hyaline degeneration of muscle with bluish  $ca^{++}$  crystals deposit on it and intermuscular edema represented by empty space (Figure 3, a and b). Thrombus formation inside blood vessel attached to the wall of blood vessels consist of faint eosinophilic fibrin thread, RBCs and mononuclear cells with fibrin thread around blood vessels and dead and life neutrophils aggregations ( Figure 3, c and d). Later the muscle showed fibrinoid degeneration in the wall of congested blood vessels and around blood vessels with neutrophils and mononuclear cells infiltrations (Figure 3e) then formation of neovascularization and aggregations of mononuclear cells (Figure 3f).



**Figure 3:** photomicrograph of H&E stained muscle tissue of Suppurative Injection Site lesion In Cattle (a)Fibrinopurulent inflammation with aggregation of neutrophils (black arrow) and bluish  $Ca^{++}$  crystals (arrow head) with intermuscular edema (white arrow), bar 100. (b) High power of previous (a) to show fibrin thread with aggregation of neutrophils (black arrow) and hyaline degeneration (arrow head) with calcification (white arrow) bar 20. (c) Thrombus formation (black arrow) and fibrinopurulent inflammation (white arrow) with hyaline degeneration (arrow head) 100 bar (d) High power of previous (c) to show thrombus (arrow) with fibrin thread and neutrophils aggregations (arrow head) bar 20. (e)Fibrinoid degeneration (black arrow) with neutrophils and mononuclear cells infiltrations (white arrow) and hyaline degeneration (arrow head) bar 20. (f) neovascularization (arrow head) and aggregations of mononuclear cells (arrow) bar 20.

## Discussion

Faulty injections can cause various lesions varies from only trauma to tissues resulting in an inflammatory reactions and potential injection site lesions as abscesses and in severe cases cause necrosis to the tissue. Additionally, incorrect intramuscular

injections of the gluteal or biceps femoris muscles may cause sciatic nerve paralysis which is most commonly seen in young animals [22]. Also usage of broken or blunt needles increases the chance of causing tissue trauma, causing a greater inflammatory reaction [23]. Clinical findings of the examined animals were similar to that



described by Radostits *et al.* [11] and Smith [24].

Regarding systemic reaction, there was an increase in body temperature, heart rate and respiration in groups II and III than control group. This may be due to the infectious agent causing different lesions; our results were in agreement with Radostits *et al.* [11].

Local swellings were due to accumulation of pus content either collected into capsule or interspersed between the muscle fibers. Tenderness and hotness were attributed to the inflammatory reaction.

In our study, the effect of infection on hematological parameters was evident. Moderate anemia was recorded in the hemogram of both diseased groups, this alteration in hemogram might be a feature of anemia of inflammation that include suppressive effects of proinflammatory cytokines on erythropoiesis, and elevated inflammatory markers, such as C-reactive protein [25].

Leucocytosis was higher in muscular abscesses group than suppurative myositis group. The degree of leucocytosis may give indication about severity of the diseases and progress of the case during treatment. White blood cells are the basic cells of the immune system, which determine normal body function. Leucocytosis and neutrophilia were indicative of inflammatory responses together with infection. It is worthy to mention the return of the leukogram within normal limits together with clinical improvement can be interpreted as a favorable sign, whereas a rapid fall in leukocytes without clinical improvement is regarded as a grave prognostic sign. Meanwhile serum albumin were significantly lower ( $p < 0.01$ ) in both diseased groups than control one and this may be attributed to reduced appetite. Globulin concentrations are of more value in these cases. Laboratory abnormalities included hypoproteinemia, neutrophilia, and hyperfibrinogenemia were in harmony with that of Pellegrini-Masini *et al.* [26]

In the present study, there was a significant increase in serum muscular enzymes in both diseased groups than the control group; the elevation in suppurative

myositis group was higher than in muscular abscess group. Increase of serum muscular enzymes was due to the release of the enzymes from altered muscle cell membrane. Muscle enzymes are sensitive indicators of muscle cell damage. Although serum elevations of serum AST, ALT and LDH can be seen in many myopathic and other diseases, elevation of creatine kinase(CK) level is thought to be the most sensitive laboratory test result related to skeletal muscle damage [23,27]

Fibrinogen level was significantly higher in both diseased groups, but it is highly elevated in suppurative myositis group. Fibrinogen is commonly measured in bovine species as it is the best indicator of inflammatory reactions because fibrinogen concentrations often increase prior to leucocytes alterations [28, 29].

Higher fibrinogen levels in both diseased groups, as compared to the control group might be due to its enhanced hepatic synthesis as the result of a severe inflammatory process [30]. The important function of fibrinogen is the formation of fibrin that needed for tissue repair as it providing a matrix for the migration of inflammatory related cells and migrated phagocytes, enhancement of phagocytosis, antibody-dependent cellular toxicity and apoptosis delay. Fibrin matrix also enables the movement of fibroblasts and other cells and stimulates their production during the healing of damaged tissue [31, 32, 33].

A significant increase of serum CRP in both diseased groups represents a good indicator for inflammatory conditions. Fibrinogen and CRP are constituents of acute phase protein; some studies have shown that infections increased serum CRP levels in cattle [34, 35]

Ultrasonographic scanning is useful for evaluation of normal soft tissues structure and their lesions including abscesses, myositis and tendon injuries [34]. It provides information about the size and margin of the lesion and its extension into the adjacent muscle tissue or other surrounding structures. Abscesses commonly consist of a well defined echogenic rim "capsule" with an irregular homogenous hypoechoic internal content characterized by swirling movement and an echogenic debris

"pus cell" or echogenic strands "fibrous tissue" producing compartmentalization of the lesion. Sometimes the central area become anechoic as the debris settles out or echogenic as the pus inspissates. The echogenic inflammatory cells interspersed the muscle fiber in suppurative myositis making the muscle structure more echogenic. Ultrasonographic images of diseased animals under study were similar to that obtained by Abouelnasr *et al.* [37] and Braun *et al.* [38].

Bacteriological examination of the aspirated fluid revealed that *Staphylococcus aureus*, *Streptococcus* spp. Are the main causes of the lesions and in some cases, *Corynebacterium* spp. Those result agreed with Brook [39]. Animal skin contains a wide variety of bacteria as *Bacillus* spp. *Staphylococcus* spp. which may lead to abscesses formation [1]. Streptococcal infection of skeletal muscle can take in the form of phlegmon, an abscess and in severe cases necrotizing myositis. Involvement of skeletal muscles may occur as a manifestation of septicemia or by invasion of muscle following wound contamination. So, multiple sites may be involved. Repeated usage of needles either by not changing needles or not applying aseptic techniques increases the chance of transferring pathogens between animals and increases the risk of formation tissue lesions [40].

Diagnosis of suppurative lesions after injection is typically confirmed by a muscle biopsy. The histological character of lesions was typical of severe inflammatory reactions mainly in form of fibrinopurulent inflammation and neutrophils aggregations our results were in agreement with of O'Toole *et al.* [41].

Although the IM injection procedure appears minor, but it needs proper assessment of the animal and appropriate techniques to prevent the rare but dangerous complications which may occurs. Early diagnosis, use of appropriate antibiotics and prompt surgical intervention are required for better outcomes in these cases. Furthermore, leucocytes count and CRP value is an important tool in prognosis, treatment, and follow- up of these cases.

## Conclusion

Injection site lesions are common in cattle. Injection technique is critical for treatment of many diseases, sterile needles must be used. Inadequate cleaning of syringes or unsterile is often the cause of localized infection at the injection site. The volume of drug in each injection should not increase than 10-15 mL in young cattle in the same site. Ultrasonographic examination offers an excellent method for differentiation between swellings, measurement of the abscesses, detection of capsule and the relationship with the surrounding tissues, which help the surgeon in detection of the preferred treatment technique. Detection of leucocytes and serum CRP level is essential for diagnosis, prognosis and follow up of muscular diseases in cattle. Results necessitates aseptic preparation of skin before an intra-muscular or sub-cutaneous injections in cattle, as well as following the manufacturer's recommendations for dosage, route of administration, and expiration date.

## Conflict of interest

The authors declare no conflict of interest.

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## المخلص العربي

الإصابات القيحية لموضع الحقن في الأبقار: الفحوصات الاكلينيكية والبيوكيميائية والموجات فوق الصوتية

## والهستوباثولوجي

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أجريت هذه الدراسة على ٤٠ حيواناً ١٣ بقرة ، ١٤ ثور بقري و ١٣ عجل بقري من كلا الجنسين ، تراوحت أعمارهم بين ١-٥ سنوات. وقد جاءت الحيوانات إلي مستشفى الزقازيق البيطري التعليمي ، كلية الطب البيطري ، جامعة الزقازيق ، مصر بتاريخ مرضى لاصابة موضع الحقن العضلي . وتم تحديد المشكلة على أساس التاريخ المرضي مع وجود تورم يتراوح قطره من ٥ إلى ١٥ سم ، وكذلك أعراض مختلفة وفقاً لموضع الاصابة ، مثل خلل في المشي ، ألم في عضلات الفخذ و الألية و الرقبة. وقد استند التشخيص إلى التاريخ المرضي والنتائج الاكلينيكية والبيوكيميائية والبذل بالإبرة و الموجات فوق الصوتية والتحليل الهستوباثولوجي لنسيج العضلات. وبناء على التشخيص ، تم تقسيم الحيوانات إلي ثلاث مجموعات؛ المجموعة الأولى (المجموعة الضابطة ، ن = ١٠) ، المجموعة الثانية (مجموعة الخراج العضلي ، ن = ١٧) ، والمجموعة الثالثة (مجموعة التهاب النسيج العضلي القيحي ن = ١٣). وكشفت نتائج الهيماتولوجي عن فقر دم خفيف وزيادة عدد كرات الدم البيضاء وخلايا النيتروفيل في مجموعة الخراج العضلي ومجموعات التهاب النسيج العضلي القيحي، في حين أوضحت نتائج التحاليل البيوكيميائية عن نقص ألبومين الدم ، وزيادة الجلوبيولين ، وارتفاع إنزيمات العضلات. زيادة في بروتين سي التفاعلي (CRP) والفيبرينوجين بشكل كبير في كلا المجموعتين المريضة بمقارنتهما بالمجموعة الضابطة بينما كشفت نتائج الفحص بالموجات فوق الصوتية في مجموعة الخراج العضلي عن وجود محتوى صديدي متجانس محاط بغلاف hyperechoic وعميق في العضلات ، بينما زادت echogenicity في مجموعة التهاب النسيج العضلي القيحي. بينما كشفت نتائج الهستوباثولوجي لنسيج العضلات المصابه عن التهاب ليفي صديدي مع تجمع خلايا النيتروفيل الميتة والحية ، وتنكس الهياكل في العضلات وورم العضلات. الخلاصه اثبتت هذه الدراسه أن الموجات فوق الصوتية تعتبر تقنية دقيقة لتشخيص حالات إصابة موضع الحقن. وكذلك فان تقييم (CRP) والفيبرينوجين مع عدد الخلايا البيضاء مفيد للغاية في التشخيص والتنبؤ بالحاله.