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Physical and pathological assessment of cardio-hepatobiliary system with salivary gland in buffalo infected with FMD

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Abstract

This study achieved assessing clinical manifestation with salivary gland, liver, heart, gallbladder histopathological changes in buffalo infected with Foot and mouth disease. Out of 50 buffalo herd in Al-Qasim district, Babil/Iraq, the positive cases previously detected with FMD screening kit (VDRG® FMDV 3Diff/PAN Ag Rapid kit) examined clinically, compared with 10 clinically healthy animal used as control, furthermore Gross lesions in mouth dental pads, gums and hoofs and histopathological study done on salivary gland, gall bladder, liver and heart. Clinical examination revealed recumbency, rope salivation, fever, polypnea, tachycardia, depression, anorexia and lameness, tow calves with primary lactation failure died within 1-3 days. Gross pathology exhibit vesicles, erosion and ulceration in its early stages by vesicles in the plamim nasale, in the oral cavity, tongue and hoof. Histopathological examination of infected calves revealed extensive necrotic myocarditis, necrotic hepatitis with ulcerative cholecystitis although no pathological lesion in salivary glands. this study emphasise the exceeding mortality and morbidity rate of young buffalo calves infected with FMD due to sever myocardial necrosis with outlining the associated pathological lesions in non-traditional organs as salivary glands, gall bladder and human consuming organs liver along with heart.

Keywords: Gall bladder, gross lesion, heart, salivary gland

Introduction

FMD is a highly contagious, modifiable viral disease that can cross the species barrier causes annual massive economic losses to owner's stock (Al-Salihi KA, 2019) ^[6], it is an infectious disease that affects wide range of cloven-hoofed mammals of small and large ruminants and swine (Sameer and Jarullah, 2014) ^[20]. FMDV is non-enveloped RNA virus in the genus *Aphthovirus* with four structural proteins surrounded the icosahedral capsid genome of approximately 8500 bases. There are seven immunologically, distinct, serotypes (O, A, C, Asia 1, and Southern African Territories (Peng *et al.*, 2020) ^[16], with over 70 subtypes with no cross protection Antigenic variation and instability contribute to persistent nature.

The disease has expanded its geographical distribution and become endemic across extensive areas in Africa, Asia, and the Middle East, making FMD a trans boundary emerging disease of ruminants, Disease spread via movement of infected animals, contaminated animal products, and fomites; can be transmitted up to 250 km by wind (Jamal and Belsham, 2015) ^[11]. Clinical manifestation of excessive salivation and lameness due to vesicles and blisters of oral cavity, nostrils, teats, udder, lethargy, fever, decreased milk production, and blanching of coronary bands may precede vesicles, all with hyperthermia occur in infected calves while signs in small ruminant including sheep and goat were mild to nil making disease difficult in diagnosis (Knight-Jones *et al.*, 2017) ^[14]. The pharyngeal mucosa is thought to be hostile site for primary FMD virus replication, then it is transferred through lymphatics and blood vessels by viremia to oral mucosa which is secondary sites of replications (El-Karim, *et al.*, 2022) ^[7]. The disease-related morbidity rate reaches 100% while mortalities are low in adult animals as well as (Knight-Jones *et al.*, 2017) ^[14] except sudden death in young calves (Admassu *et al.*, 2015) ^[1].

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clinical signs demands huge economic damages due to loss of milk production post 2 months of observation (Arzt *et al.*, 2024) ^[4], uncontrolled open Iraqi borders, substantial economic losses due to control measures to attain disease-free status severely weakened the disease control making introduction of exotic FMDV to Iraq as well as creates a threat to the neighboring countries and worldwide, and *vice versa* (Al-Salihi KA, 2019) ^[6]. However, this study focused to figure clinical manifestations of the disease as well as assessing the salivary gland, myocardial, gall bladder and liver histopathological changes in buffalo calves.

Methodology

Animals and Samples Collection

This study examined a total of 50 buffalos (*Syncerus caffer*) calves were obtained in Al-Qasim district, Babil/Iraq at February, 2024. Both sexes aged between (≤ 6 months and 2 years), all analyzed buffalo calves have no history of FMDV vaccination. All the calves examined physically after being diagnosed with screening FMD diagnostic kit VDRG® FMDV 3Diff/PAN Ag Rapid kit by using saliva. Ten clinically healthy calves were served as the control group.

Clinical examination

General examinations performed on detectable changes in animal hair, gait and muscle (Radostits *et al.*, 2000) ^[18]. Vital signs of pulse and respiratory rate, body temperature (rectal

and axial), appetite were monitored. Examinations of the buccal cavity and claws were also performed.

Pathological studies

Histopathological Techniques

The specimens of 1cm³ taken from salivary gland heart, liver and gall bladder, specimens were processed through the routine tissue fixation, clearing, mounting and stained by (Hematoxylin & Eosin stain) procedure according to (Ahmed, 2016) ^[5].

Results and Discussion

Clinical examination and gross lesions

The infected examined calves revealed that all animals suffering from vesicles on the nasal planum, oral cavity, oral mucosa with rope salivation, anorexia depression, panting mouth breathing, fever $\geq 40.7^{\circ}\text{C}$, lameness, rough hair., tachycardia, all calves that unable to suck their dams, recumbent and died within 3 days, at 4%.of mortality rate (Figure 1) shows frequencies and rates of each clinical sign. Moreover the infected calves have indicative elevation in heart rate and respiratory rate with significant hyperthermia (Table 1) compared with control group. Gross pathology of calves infected calves dental pads exhibit vesicular lesion, multiple areas of ulceration and erosion (Figure 2a, b, c), upper surface of the gum (Figure 2d) and hoof (Figure 2e), dead calves appear emaciated (Figure 3).

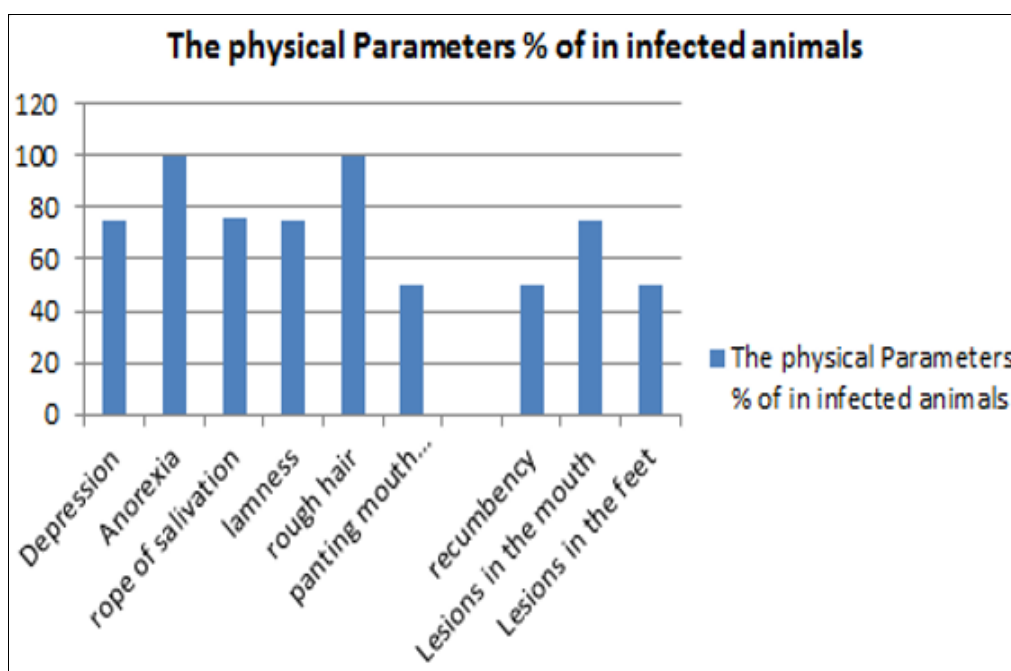


Fig 1: Frequency clinical symptoms in infected animals (%)

Table 1: Rates of heart, respiratory, fever of infected calves

Parameters	Infected animals	Control
Body temperature/ $^{\circ}\text{C}$	40.7	38.5
Heart rate / min	116-119	91-101
Respiratory rate / min	41-45	29-33



Fig 2: Gross lesions of calves infected with FMD showing vesiculo-ulcerative lesions in dental pads, gums and hoof respectively



Fig 3: Dead calves with emaciation

Foot-and-mouth disease is an extremely important disease of artiodactyls worldwide including Iraq which is endemic with disease appearing periodically and causing major economic losses in Iraq. 50 foot and mouth clinically affected buffalos were examined in February 2024 at Al-Qasim district, Babil. This research performed for assessing: Clinical sings, histopathological changes in investigated animals. This current study revealed that mortality rate was about 4% in young calves, a result that is compatible with the Salim *et al.*, (2019) ^[19]; Aktas *et al.*, (2015) ^[3] findings, whom displaced that the mortality rate was 9.90% and 11.34% respectively in the acute myocardial form of the FMD-diseased calves, although this study showed that the diseased calves of clinical form were have, depression, feverish, accompanying in appetite, vesicles that may coalesce to form bullae which rupture leaving raw ulcers. Fluid-filled vesicles are present in the oral cavity, lips, rostral plate, and tongue accompanied with ptyalism. The same signs indicated by (Sarry *et al.*, 2022) ^[17]. Sudden death conceder conspicuous sign of 1-2 months old calves. These findings totally agreed with (Ali *et al.*, 2016) ^[5] who registered in young calves (2) months of age just before death they have tachypnea, arrhythmia, fever, with tachycardia; while with aged calves, peculiar signs are mouth and feet vaculation lesions. Elhaig and Elsheery (2014) ^[9]

recorded cardiac arrhythmia besides labored breathing just before death. Meantime, less than 1-month old buffalo calves were found dead with indistinguishable clinically signs.

This study showed presence of vesicular lesions dispend on dental pad, gum and hoof erosion and ulceration with inflamed borders frequently appear at the site of raptured vesicles, the same lesions mentioned by (Jubb *et al.*, 2016) ^[12].

Histopathological examination of internal organs:

Histopathological examination of the mandibular and sublingual salivary glands of calves died from infection with FMD showed no pathological lesions (Figure 4a, b) respectively, while heart histopathological examination of heart shows focal coagulative necrosis (zinker necrosis) of muscle fibers (Figure 5 a, b, c and d) with endomyocardium edema between myocardium muscles (Figure 5b and c). While histopathological examination of infected calf liver died from FMD infection revealed congestion of central and portal vein(Figure 6 a, b and d) with necrosis of hepatocytes and hemorrhage in sinusoids between hepatocytes (Figure 6a, b and c). However histopathological examination of gall bladder shows acute cholecystitis with mucosal erosions (Figure 7).

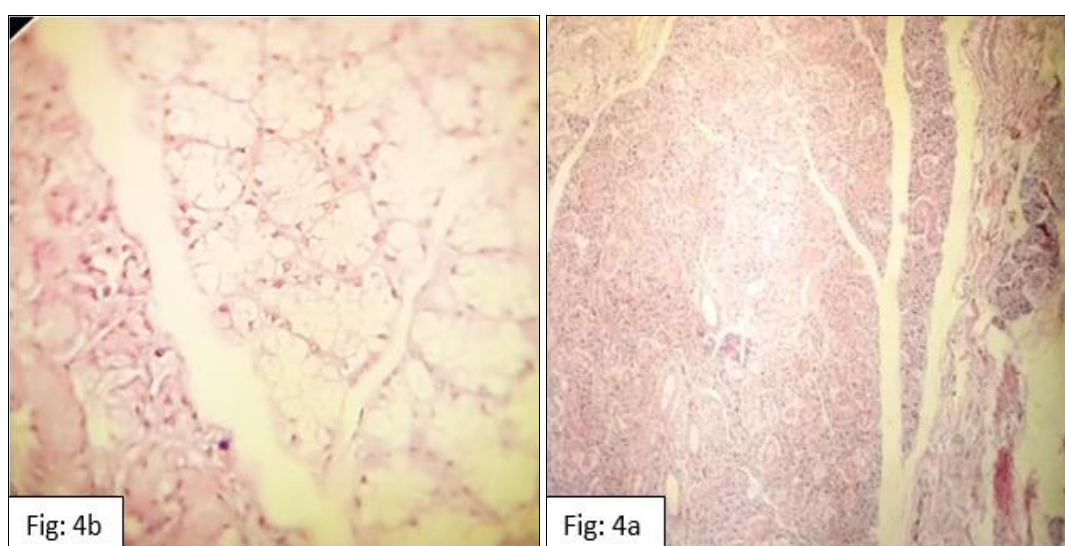


Fig 4: Histopathological section of infected calf with salivary gland, showing no pathological lesions, (H&E, 400x)

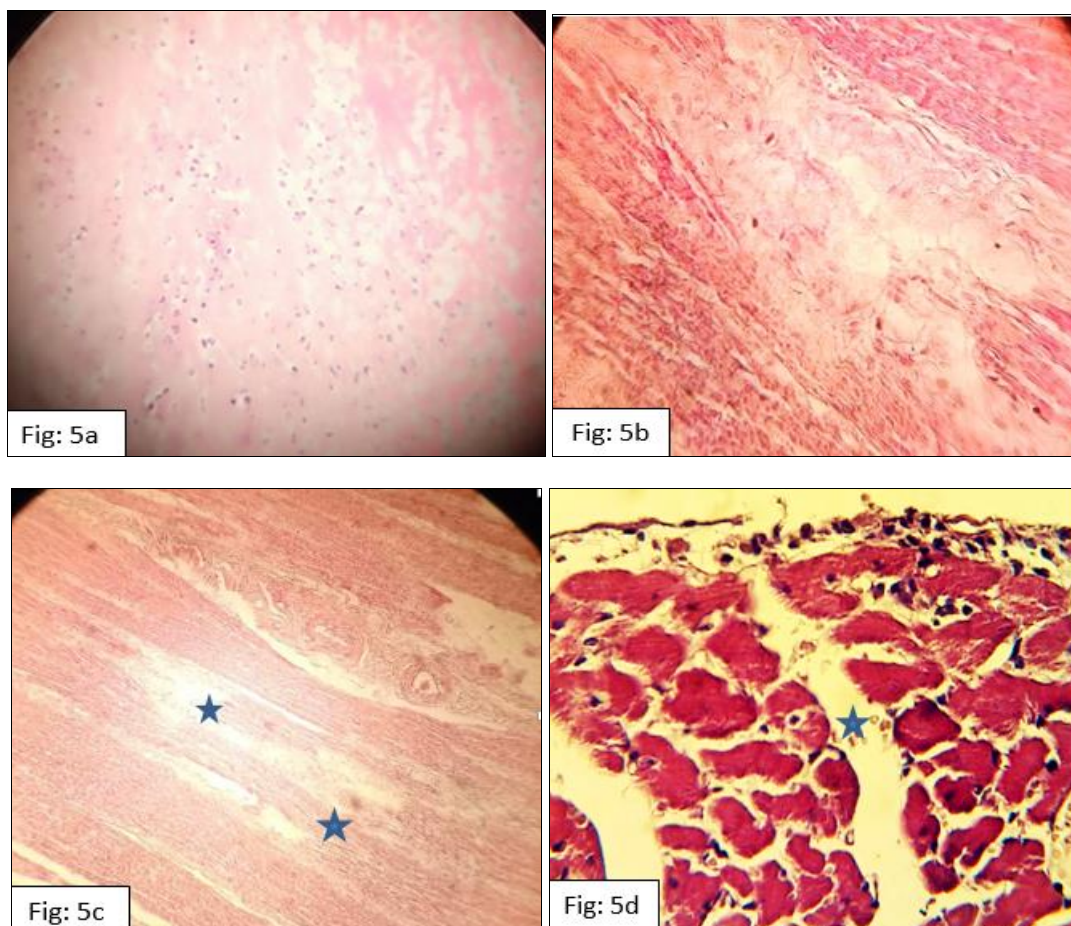


Fig 5: Histopathological section of FMD infected calf heart, showing endomyocardial focal coagulative necrosis with edema (star), (H&E, 400x)

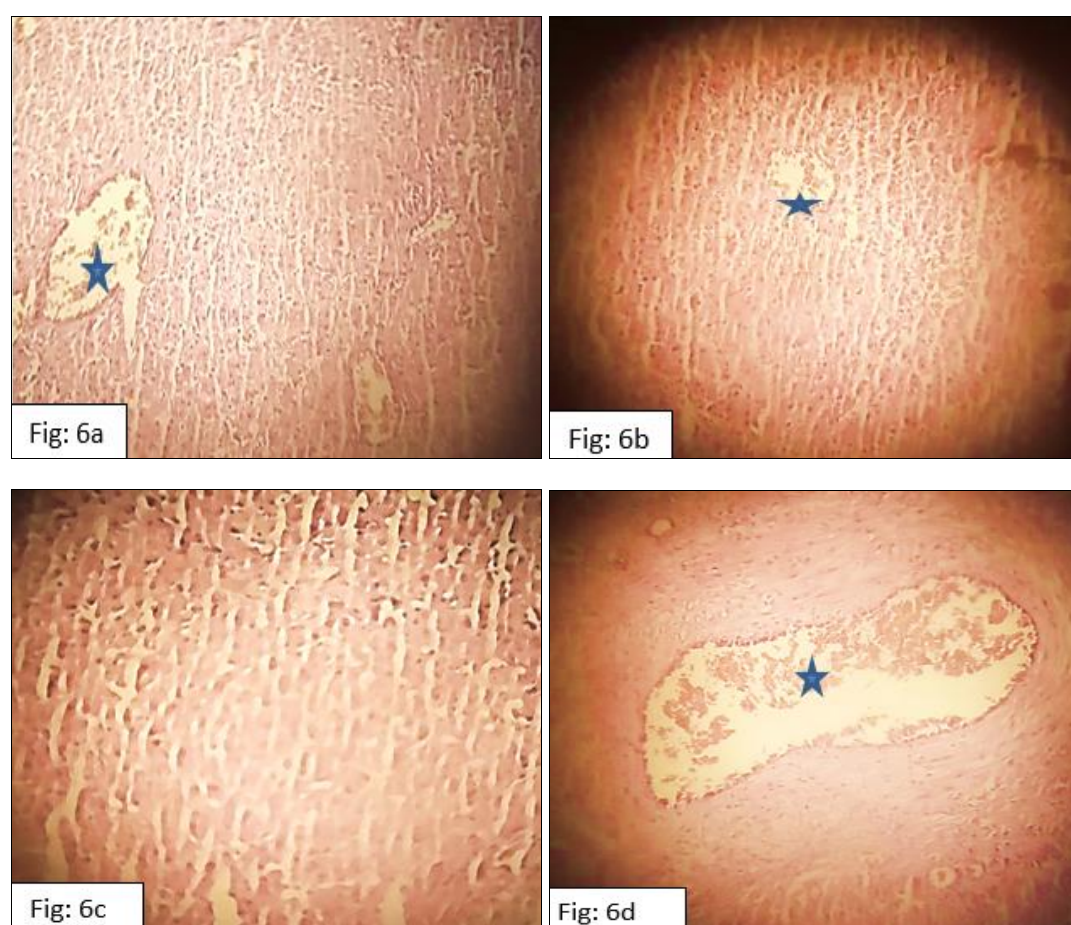


Fig 6: Histopathological section of liver of infected calf with FMD Shows congestion of central and portal vein (star) with necrosis of hepatocytes and hemorrhage in sinusoids between hepatocytes, (H&E, 100X)

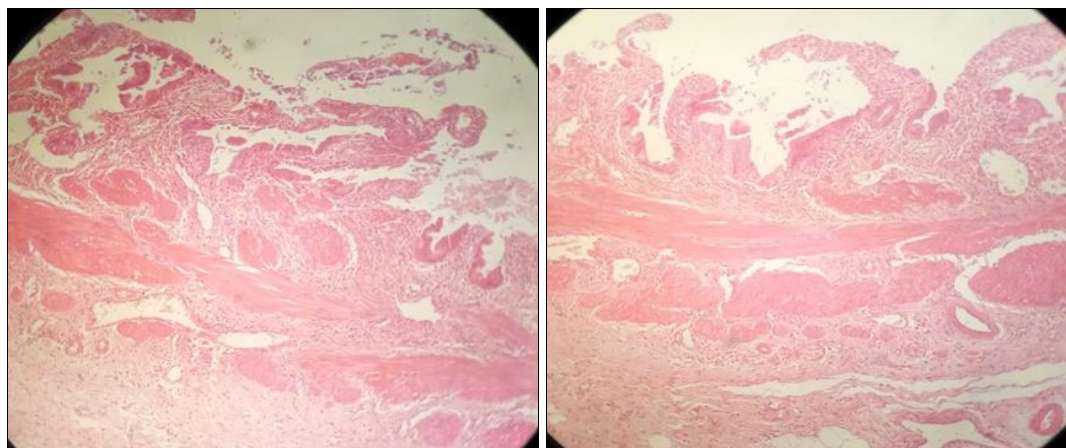


Fig 7: Histopathological section of gall bladder of infected calf with FMD shows acute cholecystitis with mucosal erosions, (H&E, 200X)

The histological changes in the myocardium of infected calves revealed myocardial muscle cells coagulative necrosis with myocarditis associated with interstitial edema, which are compatible with (Karapinar *et al.*, 2010) [13]. In the acute form, of FMD, degeneration and necrosis of myocardial fibers occur due to viral replication (Stalker, 2007) [15], laterally focal myocardial ischemic infarction developed, ending in complete heart failure due to stoppage of electrical signal transport to other parts of the cardiac muscle (Islam *et al.*, 2017) [10]. In this study, liver lesions were represented by congestion of central and portal vein with necrosis of hepatocytes; a result totally agreed with (Ali *et al.*, 2016) [6] that assisted hepatic cells degeneration as well as thickening hepatic capsule in addition to infiltration of lymphocytes and macrophages in fibrotic portal areas; these findings were in a partial agreement with (El-Amir *et al.*, 2014) [8], that conducted presence multi-focal hepatocellular necrobiosis and necrosis areas due to secondary clostridial infection within the necrotic area.

Conclusion

FMD in Iraq still requires the application of active measures that aid in its prevention and control. This work emphasizes the importance of FMD as a viral disease causing significant morbidity and mortality in buffalo, and this work denotes pathological lesions with full description of non-traditional organs as salivary glands, gall bladder and liver along with heart. Necrotic myocardiopathy assumes the etiology of death in young buffalo calves.

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Declaration of Conflict of Interest

The author declares that there is no conflict of interest.

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