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Histopathology of Skin Warts in Cattle

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Abstract
Warts consider one of the almost health diseases in cattle. This study aims to study histoplathological changes in warts of clinically identified infected cattle. An overall 200 cattle of both sexes and different ages, existed at different areas in Al-Qadisiyah province were examined clinically to detect an existence of warts on different parts of the body during June-July (2023). The findings identified 13.5% of study animals were positively infected with warts. Grossly, warts were appeared as exophytic proliferation cutaneous lesions of many variable sizes in different body sites; mainly ear, mandible, muzzle, udder/teat, and perianal (genitalia). Lumpy or cauliflower multiple papillarygrey forms were existed a mostly in exposed-external areas of ear, mouth, muzzle; while, white to pink warts were seen in udder and perianal regions. Microscopically, warts showed the presence of marked erosion and necrotic lesion in exposed area of warts, eosinophilic necrotic debris and severely inflamed inverted papilliform projections extending to mass center, segments of spinosum, squamous cells epithelium, as cords and nodules enclosed centrally red keratin material surrounded by wide zone of fibrous connective tissue, high magnification of nodule enclosed centrally red keratin material (pattern of half-cross onion), surrounded by wide zone of fibrous connective tissue, inclusion bodies, many koilocytes with simple division, accentric keratosis, hyperplasia of epidermal cells and infiltration of inflammatory cells. In conclusion, gross diagnosis of warts revealed a high prevalence of infection in study areas. Also, a wide variety of irreversible skin changes were occurred due to warts. However, application of other advanced diagnostic assays such as electron microscope and immunohistochemistry could provide a moreover data about the virus and their types/strains.
Keywords: Papillomatosis, Bovine skin lesion, Gross characteristics, Light microscopy, AL-Qadisiyah province

Introduction

Wart, mainly caused by *Papillomaviruses* (PVs) that belongs to Papillomaviridae family, is a viral disease of worldwide occurrence. There were approximately more than 20 types of PVs, which described in animals causing different benign proliferative lesions in skin of several animals as well as human (de Queiroz Simões and Barth, 2017; Gallina *et al*., 2020). Mechanisms of infection are poor known; however, domestic animal may play great roles in transmission of infection through direct contact that appears having enough roles for spreading warts between animals, but indirect contacting with the contaminated fomits can

increase the percentage of infections at the field (Mazzuchelli-de-Souza et al., 2018). Indirect contact with the contaminated materials by milking machines, injections, injuries and scratches might be active route for virus transmission (Bind, 2017; Kale et al., 2019). Co-factors include immunodeficiency, long-term exposure to sunlight, hormonal imbalances and malnutrition (Ugochukwu et al., 2019). Additionally, different types of arthropod were implicated to play a role in transmission of infection suggesting their activities in reserving of infection and impacts in distribution of papillomavirus to other domesticated and wildlife animals existed at the same field (Haspeslagh et al., 2018). This leading for economically negative costs particularly among regions having wide distribution numbers of herd/flock, since bovine papillomavirus inducing a variety of chronically, benign as well as malignant tumors (Zhu et al., 2019). Bovine PV (BPV) has great importance *in vivo* models to human PV (HPV) reports more than the veterinary interesting (Cuzick et al., 2014; Tombak et al., 2019).

Significant increases in prevalence of specific antibody towards the virus proteins among the animals, indicating the commonly exposing to PVs earlier during their life stages (El-Tholoth et al., 2020). In field, the diagnosis of BPVs based on clinical signs due to the vast changes in skin which being specific for the disease due to planar progression in epidermal layers (Daudt et al., 2018). Histopathology and molecular assays have been applied usually to diagnosis of the disease and confirmation of infection (Lunardi et al., 2016).

In Iraq, widespread occurrence of warts have been observed clinically in domestic animals, but the volume of available data remain low and need to support (Hamad et al., 2017; AL-Salihi et al., 2020; Marzok et al., 2020). Therefore, this aims to studying the histopathological changes in warts of clinically identified infected cattle.

Materials and methods

Ethical approval

Scientific Committee in the College of Medicine (University of Al-Qadisiyah) was approved the work of this study.

Study animals

An overall 200 cattle of both sexes and different ages, existed at different areas in Al-Qadisiyah province, were examined clinically to detect an existence of warts on different parts of the body during June-July (2023). Then, some infected cattle were subjected to surgical collection of warts under aseptic and anesthetic conditions into plastic container contain 10% neutral buffered formalin (10% NBF).

Histopathology

The lung tissues saved in 10% NBF were dehydrated, cleared, infiltrated, embedded, sectioned at (4-5 μm), and loaded on the slides that stained with the Hematoxylin and Eosin and examined under the light microscope (MEIJI, Japan) at 40X (Gharban et al., 2023).

Statistical analysis

The GraphPad Prism Software was served for identification of significant differences in values of serology among G1 and G2 groups at $P<0.05$ (Gharban, 2023). Values were represented as Mean ± Standard Errors (M±SE).

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Results

Among totally 200 cattle examined clinically for detection of warts, 27 (13.5%) were positively infected (Figure 1).

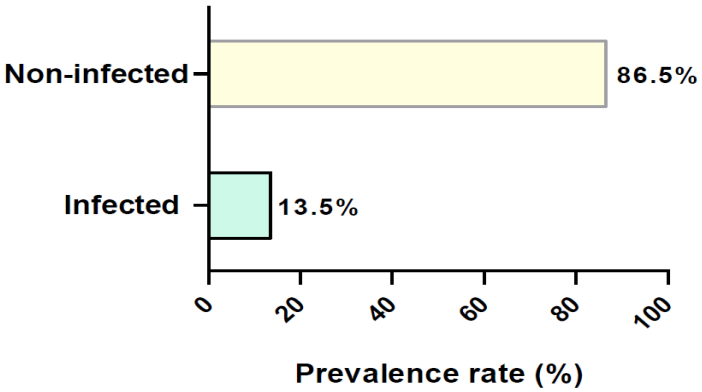

Figure (1): Total results for prevalence rate of warts in cattle (Total No: 200)

Grossly, warts were appeared as dry, horny, exophytic, proliferative cutaneous lesions that varied remarkably in their sizes and found at different sites of the body; mainly head (mandible and muzzle) and to a less extent in neck, udder, legs, and perianal /genitalia. Lumpy or multiple papillary grey form was existed a mostly in exposed-external areas of muzzle; while, solitary cauliflower warts were seen in mandible, neck, udder, legs and perianal regions (Figure 2).

Figure (2): Gross detection of warts among different bodily parts of

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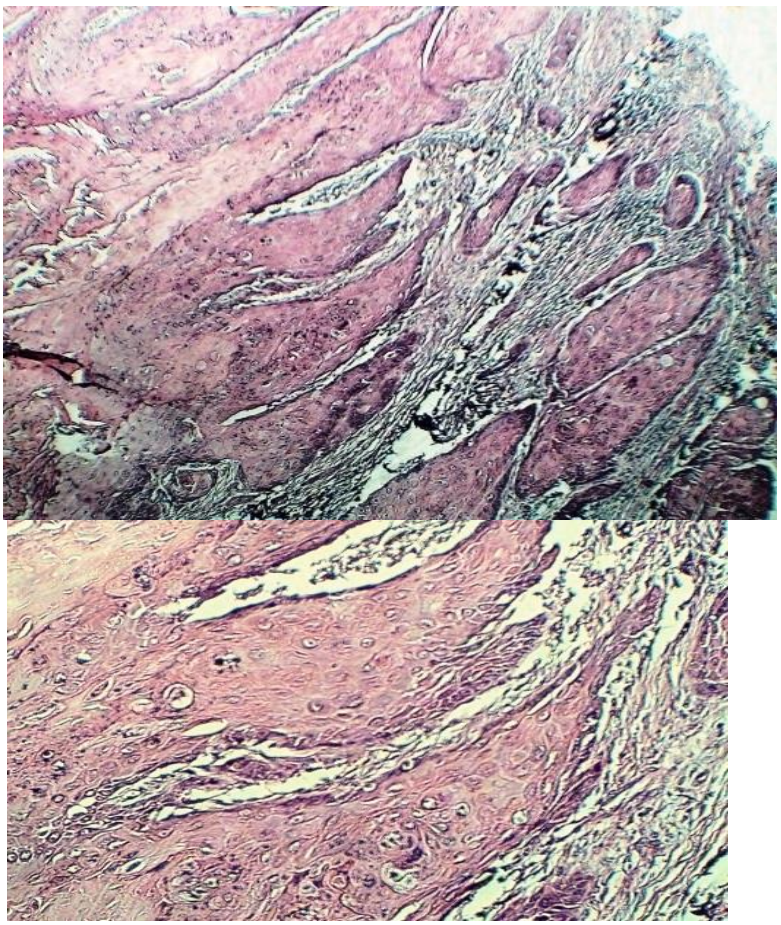
Microscopically, warts showed the presence of marked erosion and necrotic lesion in exposed area of warts, eosinophilic necrotic debris and severely inflamed inverted papilliform projections extending to mass center, segments of spinosum, squamous cells epithelium, as cords and nodules enclosed centrally red keratin material surrounded by wide zone of fibrous connective tissue, high magnification of nodule enclosed centrally red keratin material (pattern of half-cross onion), surrounded by wide zone of fibrous connective tissue, inclusion bodies, many koilocytes with simple division, accentric keratosis, hyperplasia of epidermal cells and infiltration of inflammatory cells (Figure 3).



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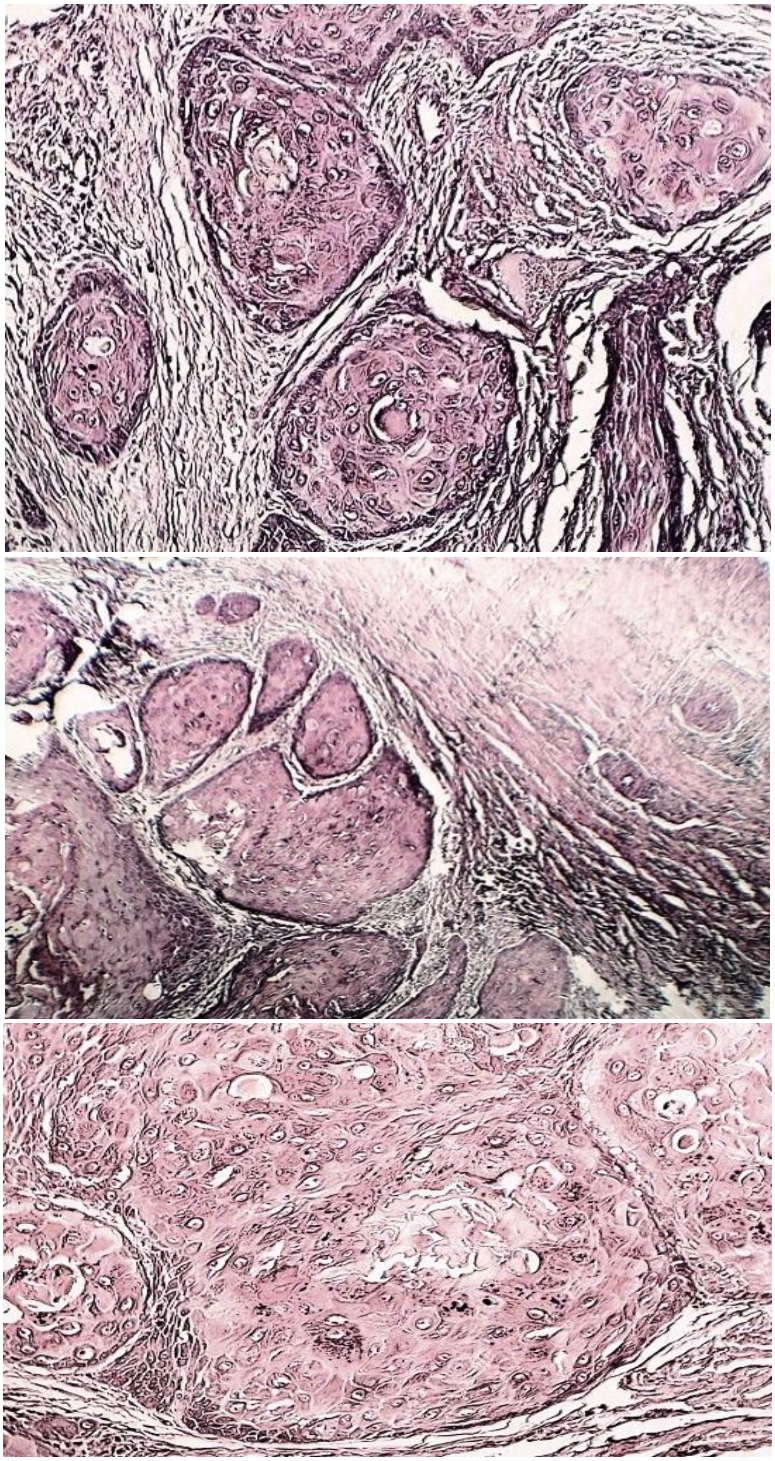

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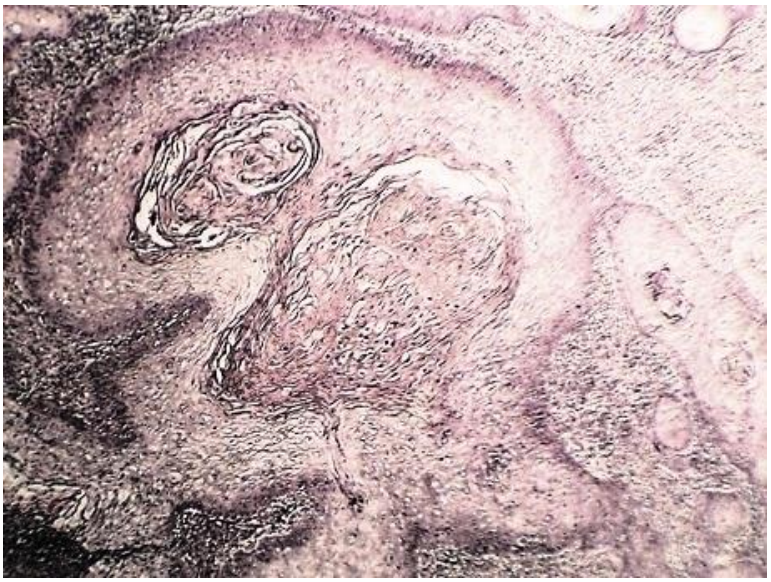

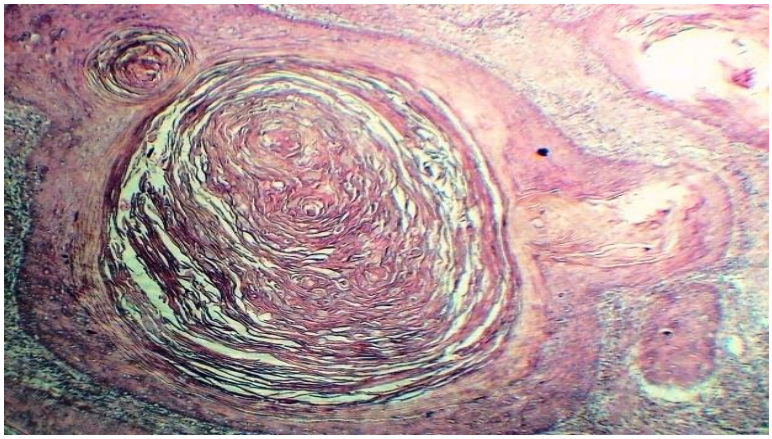

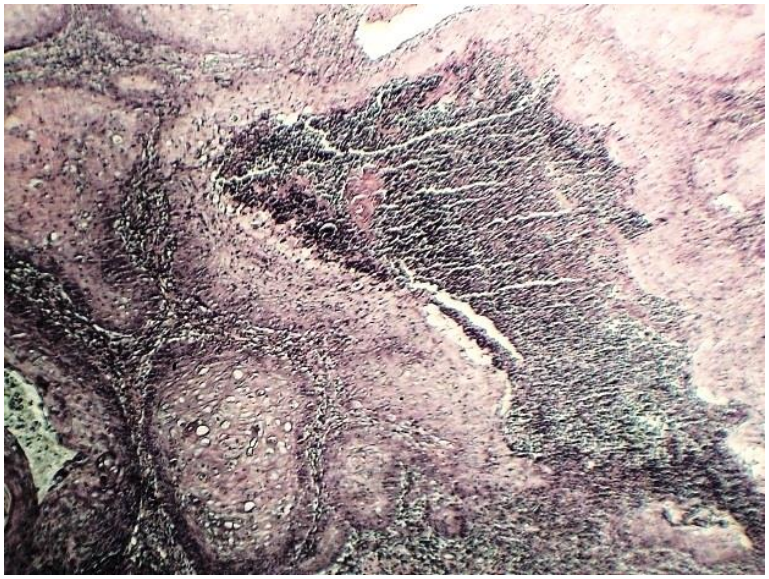

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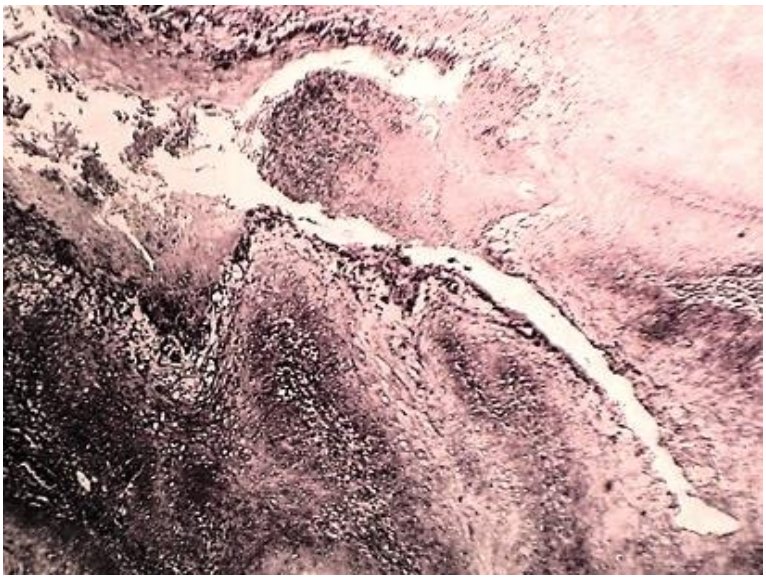

Discussion

Based on gross characteristics, the prevalence of warts in study cattle was 13.5%, which higher than detected in Egypt (Salib and Farghali, 2011). Our gross observations were identical to findings of other studies which described the warts as dry, horny cauliflower-like appearance (Hamad et al., 2018; Mansour et al., 2019; Al-Salihi et al., 2020). In present study, warts were existed among different bodily areas but almost predominantly in head. Among different studies, the most commonly affected sites by warts were udder of lactating cows, eye, mouth, ears, neck and genitalia (Salib and Farghali, 2011), teats and necks (Hamad et al., 2018); shoulders, neck and head (Mansour et al., 2019); as well as genitalia, head, shoulders and perianal (Al-Salihi et al., 2020). However, McMurray et al. (2001) showed that cattle warts were differred in their tissue specificity based upon the virus's genotype and the related diseases. Ahmed and Hassanein (2012) recorded that the white and colored head and/or body cattle having a high sensitivity to subsequent occurrence of warts than others. Arslan et al. (2018) reported that cattle warts can identify anywhere on the body; but frequently, on skin surface of heads, necks, teats, udders, genitalia.

Our findings demonstrated that different changes in wart lesions were seen microscopically as reported by several authors (Araldi et al., 2015; dos Anjos et al., 2019; Timurkan and Alsigir 2017). Different researchers reported that changes appear in the affected layers of the skin (epidermis, dermis) might include hyperkeratosis, thickening and hyperplasia of the epidermis, vascular congestion, extensive infiltration of inflammatory cells mainly macrophages and fibroblasts, inflammation of the stratum spinosum, vacuolization and infiltration of inflammatory cells into the skin and hyperplasia of the stratum basale are also observed. These results have been confirmed by various studies conducted (Movassaghi and Mohammadi, 2009; Mt et al., 2012; Araldi et al., 2015a, b; Al-Sabaawy and Al-Sadi, 2021). Ballooning degeneration of epidermal cells and eosinophilic endocytoplasmic inclusions can also be seen (Atasevan et al. (2016). In addition, histopathological changes in epidermal layer were with that mentioned by De Sanjosé et al. (2013) as the occurrence of swelling of the

stratum layer, ballooning and vaculotion. Concerning different types of PVs, BPV-1 and -2 were demonstrated in skin samples of domestic and wild animals (Scagliarini et al., 2013; Savini et al., 2016; Garcês et al., 2020). Bovine delta-PVs have the ability for infecting not only epidermal cells but dermal cells also causing bovine fibropapilloma (de Villiers and Gunst, 2009) and equine sarcoma (Brandt et al., 2008). Other investigators have shown that BPV-2 and -13 cause fibrinosis, while BPV-1 can cause papillomas and skin lesions without the involvement of fibroblasts (Araldi *et al*., 2017; Roperto *et al*., 2018). Similar behavior was indicated in infected animals with BPV-2 in Brazil (Mazzuchelli-de-Souza et al., 2018), where histopathological lesions confirmed as papillomas were located on the hind limbs. Other studies identified BPV-1 as the only strain existed in lesions, demonstrating an important pathogenic role in cutaneous papillomas (Ukpo et al., 2011; Augustin et al., 2018). Furthermore, the fact that the BPV-1 was the cause of papilloma lesion but not a fibroma suggests that the infection is due to the virus-host interaction, and that the BPVs transmits horizontal, whereas HPV transmits vertically to humans (Sarkola et al., 2008), horses (Savini et al., 2019) and sheep (Savini et al., 2020).

Conclusion

Gross diagnosis of warts revealed a high prevalence of infection in study areas. Also, a wide variety of irreversible skin changes were occurred due to warts. However, application of other advanced diagnostic assays such as electron microscope and immunohistochemistry could provide a moreover data about the virus and their types/strains.

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Conflict of interest

No.

Authors' contributions

NED: Tissue processing. MAS: Collection of wart samples and statistical analysis. Both authors contributed equally in microscopic examination of slides, reading and approving the final copy of the manuscript.

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