A case report on prolonged clinical management of papillomatosis in a heifer

A clinical report submitted in the partial fulfillment of the requirements for the degree of Doctor of Veterinary Medicine (DVM)

A Clinical Report Submitted By:

Nurul Amin

Roll No:19/78

Reg No: 03288

Intern ID: 61

Session: 2018-19

Faculty of Veterinary Medicine

Chattogram Veterinary and Animal Sciences University

Khulshi, Chattogram-4225, Bangladesh

December 2024

A case report on prolonged clinical management of papillomatosis in a heifer

A Clinical Report Submitted as per approved style and contents

Approved by

(Signature of Supervisor)

Professor Dr. Md. Mizanur Rahman

Department of Medicine and Surgery

Faculty of Veterinary Medicine

Faculty of Veterinary Medicine

Chattogram Veterinary and Animal Sciences University

Khulshi, Chattogram-4225, Bangladesh

December 2024

TABLE OF CONTENTS

List of tables

List of the figures

List of Abbreviations

Abbreviation	Elaboration
CVASU	Chattogram Veterinary and Animal Sciences University
TAD	Trans-boundary animal disease
BPV	Bovine papilloma virus
PCR	Polymerase chain reaction
RBC	Red blood cell
WBC	White blood cell
PCV	Packed cell volume
AST	Aspartate aminotransferase
ALT	Alanine transaminase

Abstract

A 2-year-old heifer (weight-150 kg) was referred to Shahedul Alam Quadary Teaching Veterinary Hospital (SAQTVH), CVASU, Chittagong, Bangladesh with a history of wart-like growth found on the head, neck, shoulders, limbs and occasionally on the back and abdomen. On clinical examinations the heifer showed gray, brown, and black-colored pea to ball-size cauliflower-like growth. Hematological, biochemical investigation and urine analysis were performed. On the basis of history, clinical examination and laboratory findings , the case was presumptively diagnosed as papillomatosis. As treatment, there used autohemotherapy, ivermectin, antihistaminic, autogenous vaccine, homeopathy and surgical excision. In this case, autohemotherapy, ivermectin, and autogenous vaccines were not show any improvement. Although the size of the warts did not regress after administering homeopathy, no new wart growth was observed. After surgical excision, there was no new wart growth. Outcomes of the study will be useful to determine the cure rate of papillomatosis in cattle after using different treatment regimen.

Keywords: Autohemotherapy, autogenous vaccine, surgical excision.

Chapter 1 : Introduction

Infectious papillomatosis or wart is a disease occurring in cattle, goats, dogs, rabbits, horses and humans in various parts of the world. Bovine papillomatosis, a contagious and trans-boundary animal disease (TAD) caused by Bovine papilloma virus (BPV) type 1 to 10. Bovine papillomatosis occurs as a result of multiplication of the virus in basal cells, leading to wart formation. However, most warts are benign and do not proliferate indefinitely (Sreeparvathy, 2011). It produces characteristic gross lesions that are either exophytic (proliferating outwards) or endophytic (inverted) and the growth is composed of a hyperplastic epithelium supported by a discrete dermal tissue containing dilated capillaries. In Cattle, warts occur all over the body, the common sites for the development of cutaneous warts are head, eyelids, ears, neck, dewlap, brisket, shoulders and legs, occasionally on the back, para-genital region and along the lower line of abdomen (Mukherjee, 2013).

Bovine papillomavirus infects large ruminants worldwide, abundance of cases are recorded in regions having great density in United Kingdom, Germany, India, Japan, United States of America and Brazil as per these sources (Campo, 1995; Ogawa, 2004; Schmitt, 2010; Singh, 2009; Carvalho, 2012) . Prevalence (3.4%) and distribution of bovine cutaneous papillomatosis in Bangladesh have been reported by Nooruddin, 1984. This neoplastic disease could occur as a co-infection, a study findings in Japan (Ogawa, 2004) verified the presence of BPV in up to four BPV types and new putative BPV types in the same papilloma in a cattle herd. In Italy, BPV-1/2 DNA was discovered in the placenta of pregnant cows suffering from chronic enzootic hematuria (Roperto, 2012). Bovine papilloma virus-1 infection in the water buffalo was demonstrated by which was associated with cutaneous, perivulvar and vulvar fibropapilloma (Silvestre, 2009). In Scotland, BPV was reported to be associated in the occurrence of cancers of the gastro-intestinal tract and urinary bladder in cattle (Campo, 1985). In India, as per reports of recent researches conducted on cutaneous warts in bovines indicate that the DNA of BPV-1 and BPV-2 is present in both cutaneous warts as well as in normal skin, while in buffaloes, the tested samples were diagnosed with single or mixed infections, corroborate the high prevalence of the infection in bovines (cattle and buffaloes) (Pangty K, 2010). The occurrence of BPV-5 infection along with BPV-1 and BPV-2 has been reported in wart-like lesions in rumen of buffaloes, indicating the increase of the prevalence of these virus types and their ability to jump the species (Kumar, 2015).

The diagnosis of papillomatosis usually includes a clinical examination, cytology, parasitological examination, and histopathology. Polymerase chain reaction (PCR) has been used as a sensitive method for the identification and genotyping of BPV(Mesfin Mathewos, 2021).

This disease can lead to a serious economic loss if not diagnosed and treated promptly. The warts can break the skin, making it easier for bacteria to get in. This can lead to infections, especially in the udder (mastitis) or on the skin. Surgery and vaccination, or a combination of both is the most common forms of treatment and prevention. There are no exact drug regimens for warts to prevent its reoccurrence. The homeopathic medicine, autogenous vaccine, autohemotherapy and anthiomaline can be used for the treatment (Archana, 2019). The clinical manifestations of these conditions can lead to significant economic losses if not identified and treated promptly.

The objective of the present study were to,

1. To observe the treatment strategy of a clinical case on papillomatosis in a heifer.

Chapter 2 : Materials and Methods

2.1 Case presentation

A 2-years-old heifer suffering from severe generalized papillomatosis was presented in S. A. Qaderi Teaching Veterinary Hospital (SAQTVH), CVASU, Chattogram, Bangladesh with a history of two months of infection. The animal has a history of not receiving vaccinations or deworming treatments. The house has wet floors and receives insufficient sunlight, making the rooms appear gloomy. The feed habit consists of bran, husk, hay, and banana residue, administered twice daily. No treatment was given until 2 months after the initial infection. The animal was apparently healthy. The case was diagnosed as papillomatosis based on the clinical signs.

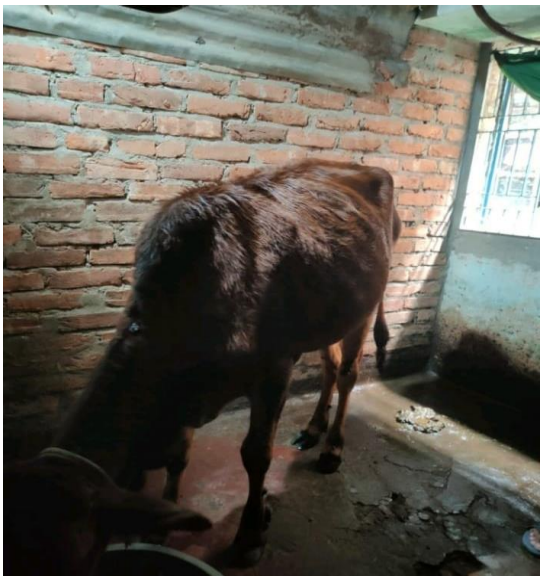

Fig 2.1.1 Housing condition of the heifer

Fig 2.1.2 Affected hind limb for the heifer with papillomatosis.

2.2 Hematological parameter analysis

Blood sample (5 ml) was collected from the affected heifer in a sterile vial. The test was complete blood count (CBC). The examination was conducted in the laboratory of the Department of Physiology, Biochemistry and Pharmacology, CVASU.

2.3 Biochemical parameter analysis

Aseptically blood sample (5 ml) was collected from the affected heifer in a sterile vacotainer. The tests were glucose, total protein, albumin, AST and ALT.

The lab test was conducted in the laboratory of Department of Medicine and Surgery, CVASU.

2.4 Urine analysis

Urine sample (10ml) was collected from the affected heifer in a sterile falcon tube. The tests were p^H , specific gravity, blood, ketone, bilirubin, protein, nitrite, leukocyte, urobilinogen and glucose. The examination was conducted in the laboratory of the Department of Physiology, Biochemistry and Pharmacology, CVASU.

2.5 Preparation of autogenous vaccine

Autogenous vaccine was prepared using 5 g of fresh active growth tissue that aseptically collected from the lesions side and washed with saline solution. Then the collected tissue sample was cut into tiny pieces, homogenized in morter and pastle with few drops of formalin solution to inactivate the virus and also added 30ml of distilled water. After that, the mixture was filtered through a whatman filter paper then the filtration was taken in a sterile vial and penicillin (200,000 IU of procaine penicillin) and streptomycin (250 mg of dihydrostreptomycin sulfate) was added to prevent bacterial growth. At last, the preparation was refrigerated at 4 °C until used.

Chapter 3 : Results and Discussion

Table 3.1 Hematological analysis of blood parameters of affected heifer

Test name	Result	Reference Value
Haemoglobin (mg/dl)	7.9	10-15
Total count of RBC (million/mm^3)	5.0	5-10
Total count of WBC (Thousand/ mm^3)	20.4	4-12
PCV (%)	24	24-46
Differential count of WBC		
Lymphocytes (%)	75	50-75
Neutrophils (%)	20	15-40
Eosinophils (%)	3	0-5
Monocytes (%)	2	1-15
Basophils (%)	0	0-2

Note : RBC, WBC, & PCV refer to Red Blood Cell, White Blood Cell, & Packed Cell Volume respectively.

Table 3.2 Biochemical analysis of blood sample of affected heifer

Serum Type	Units	Results	Normal ranges
Glucose	mg/dl	30.31	45-75
Total protein	g/dl	5.63	5.7-8.1
Albumin	g/dl	3.02	3.3-4.3
AST(SGOT)	U/L	178.2	78-132
ALT(SGPT)	U/L	52.28	11-40

Note: AST refers to Aspartate aminotransferase and ALT refers to Alanine Transaminase.

Table 3.3 Urine analysis report of the affected heifer

Name of the test	Result	Normal ranges
P^H	8.0	7.27-8.71
Specific Gravity	1.015	1.030-1.040
Blood	Negative	
Ketone	Negative	
Bilirubin	Negative	
Protein	Negative	
Nitrite	Negative	
Leukocyte	Negative	
Urobilinogen	Negative	
Glucose	Negative	

Findings of hematological investigation were shown in table 3.1, The biochemical investigation of blood serum was shown in table 3.2, and urine analysis report was shown in table 3.3. Here, we found an abnormal value of total count of WBC, and that was 20.4 Thousands/mm^3, of which the normal range is 4-12. In biochemical examination, we found an abnormal value of AST and ALT, and that were 178.2 U/L and 52.28 U/L, of which normal range is 78-132 and 11-40 respectively. In urine analysis report, we found a normal value of p^H and specific gravity, and that were 8 and 1.015 respectively.

White blood cells are an important and necessary part of your immune system. Produced in your bone marrow, they defend the body against infections and disease. But when the body is fighting an infection or inflammation, the bone marrow produces more white blood cells, leading to an elevated WBC count. It usually means infection or inflammation in the body.(Joaquin Barca, 2021)

Cattle are prone to liver disease as the bovine liver is involved in many metabolic disorders as well as infectious and parasitic diseases because of its central role in metabolism. Papillomatosis in cattle is not directly linked to liver damage. It's a viral infection that causes benign tumors (warts) on the skin and mucous membranes. Severe cases of papillomatosis can cause discomfort and reduce feed intake in cattle. This nutritional stress can impact liver function and lead to elevated liver enzymes. Severe cases of papillomatosis can trigger a systemic immune response. This could lead to temporary increases in liver enzymes(West, 1997).

According to (Tagesu, 2018) , Under normal conditions the urine of ruminants and horse is alkaline while in canines and feline it is acidic. The pH of alkaline urine is 7.4-8.4, while that of acidic urine is 6-7. Specific gravity in health varies with the state of hydration and fluid intake. The range of specific gravity of urine in normal cattle is 1.025-1.045 with an average of 1.035 and in the obstructive urolithiasis it ranges from 1.008 to 1.025.

Treatment protocol:

Phase	Treatment Intervention	Dose	Route, Frequency & Duration	Trade Name	Prognosis
First phase	Ivermectin	0.4mg/kg BW	S/Cly, SID at 1st and 14th days	Inj. Acimec 1%	No improvement
	Autohemotherapy	15 ml	I/Mly, Five times at a one-week interval		
	Pheniramine maleate	1mg/kg BW	I/Mly, SID for 4 days	Inj. Histavet	
	Autogenous vaccine	10ml	S/Cly, Four times at 2 weeks interval		
Second phase	Thuja 200	Few drops	Topically to the affected area, twice for 7 days	Liq.Thuja 200	The wart size did not regress, but the wart areas got infected and started bleeding.
		10 drops	orally mixed with 2 ml of water, SID for 15 days		
Third phase	Combination of Ivermectin and clorsulon	5ml	S/Cly, SID at 1st and 14th days	Inj. Amectin Plus Vet	The infected bleeding areas were healed, but the warts were not eliminate.
	Terpentine oil	few drops	Topically to the affected area	Turpen oil	
	Povidone iodine	few drops	Washed the infected bleeding area	Liq. Povin Vet	

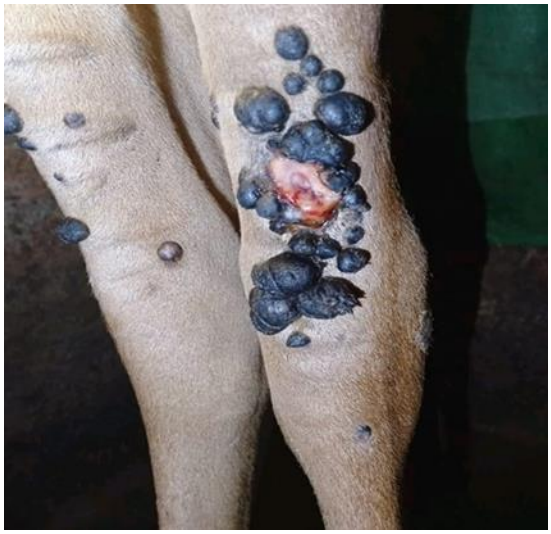

Fig 2.6.1 Secondary bacterial infection despite initial treatment with Liq. Thuja 200

After being brought to the hospital, initially autohemotherapy, ivermectin, antihistaminic drugs, and an autogenous vaccine were administered. No improvement was observed. Rather, the growth of some new warts was seen.

There, autohemotherapy, a treatment that involves injecting a small amount of a patient's own blood back into their body, has been shown to stimulate the reticuloendothelial system. This stimulation leads to an increase in the number of macrophages circulating in the blood, which may play a role in enhancing the regression rate of papillomas(Turk, 2005). It may enhance the body's immune response by helping it recognize and target virus-infected cells and also help to correct the actual reason of failure in immunological protective mechanism of body which allowed such abnormal cell proliferation(Patnayak, 2004).

The regression or healing of warts may be attributed to the immune-modulating and anti-tumor properties of Ivermectin. Avermectins, a class of compounds derived from soil microorganisms, possess antibiotic and significant anti-tumor activity(Drinayaev, 2004).

There, antihistamine used as an indication of itching at different areas of the body, drug allergy, urticaria, inflammation, and skin edema.

Autogenous vaccines are highly specific to the individual patient's strain of the pathogen, potentially offering better protection than a standard vaccine. Some papillomas are often very

location-specific, with their development linked to unique antigen reactions and DNA sequences of the infecting agents. This specificity means a vaccine effective against one strain may not protect against others, or even against different virus types, as not all papillomaviruses share immunological similarities. While repeated inoculation can build resistance to bovine papillomavirus (BPV), spontaneously infected cattle can still experience reinfection(Terziev, 2015). Autogenous vaccines obtained from affected animals within the same herds could have a prophylactic but not a therapeutic effect(Leishangthem, 2008).

After administering the homeopathic drug (Liq. Thuja 200), the wart size did not regress, but the wart areas got infected and started bleeding. However, new growth of wart was not observed. It acts on skin, blood, gastrointestinal tract, kidney and brain but mainly the action is on skin and genitourinary organs. This is of real importance in the treatment of skin conditions accompanied by the development of warty growths which bleed easily. Papillomatous warts are especially amenable to this drug. It is indicated in polyp, epithelioma(Madrewar).

Turpentine has been used in veterinary medicine topically as a rubefacient and counterirritant for treating sprains, muscle pains, and swollen udders. It has also been given internally as an antiseptic, carminative, expectorant, anthelmintic, abortifacient, and treatment for bloat.(Plumlee, 2004)

Povidone iodine is a broad-spectrum antiseptic for topical application in the treatment and prevention of wound infection. It may be used in first aid for minor cuts, burns, abrasions, and blisters. It is used on the skin to decrease the risk of infection. For pre- and post-operative skin cleansing; for the treatment and prevention of infections in wounds, ulcers, cuts, and burns; for the treatment of infections in decubitus ulcers and stasis ulcers.(Burks, 1998)

When all those treatment didn’t show substantial improvement, then the doctor proceeded for surgical intervention to inhibit the spread of the warts in remaining external organs. The animal responded well to the surgical excision, and lesions completely disappeared by the 2nd week of therapy.

Surgical excision and thermocautery

The animal was well restrained and tied before surgical excision of warts. Finally, Warts were surgically excised under 2% lidocaine at stalk, and thermocautery was done. Postoperatively, wounds were cleaned routinely.

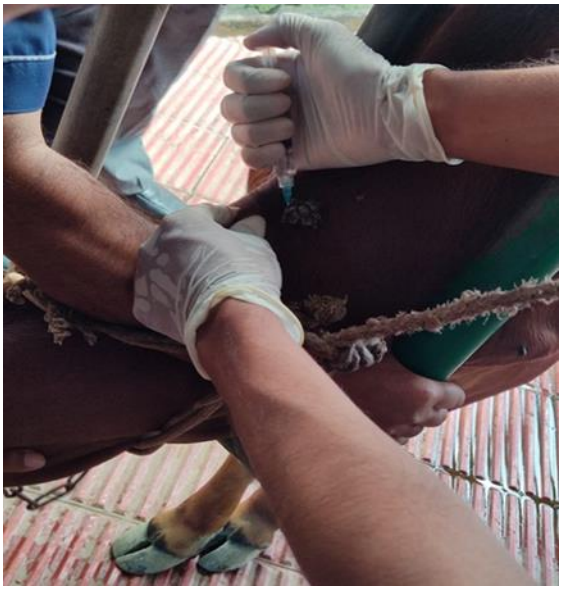

Fig 2.7.1 Administering 2% lidocaine around warts as local anesthetic before surgery.

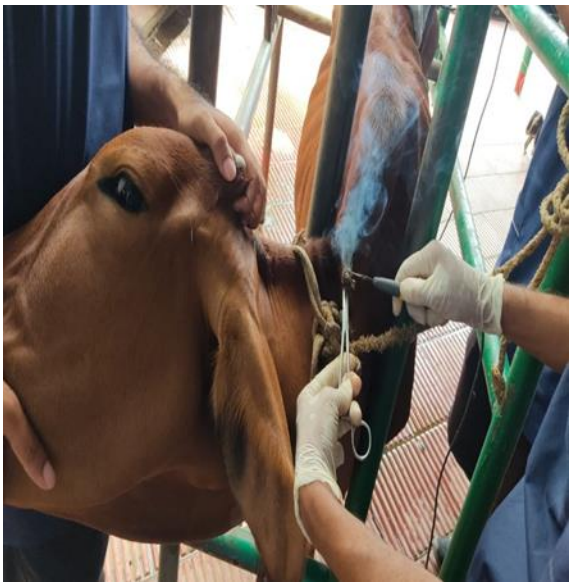

Fig 2.7.2 Thermocauterization of warts.

The cure rate of cutaneous papillomatous growths through surgical excision was higher than after the application of an autogenous vaccine. The results from surgical excision were good, there were no recurrences in operated heifers. After application of the autovaccine, satisfactory results were obtained, although the results on papillomas of larger size (< 3 cm diameter) were disappointing (Terziev, 2015). A study showed that autogenous vaccine was ineffective in regression of warts (Pence, 2005). In contrast to the present study, (Ranjan, 2013)reported that autogenous vaccine was found to be effective in treating bovine papillomatosis. According to (Jarrett, 1990), vaccination remains the best prevention method against papillomatosis. Partial excision combined with autogenous vaccine has some therapeutic value in cattle with small pedunculated papillomas, but not in cattle with large confluent lesions (Ssenyonga, 1990)

Chapter 4: Conclusion

Most of the cases of papillomatosis are severe though recovery is possible if taking proper clinical management during early stage of the disease. The present study revealed that treatment of bovine papillomatosis using Thuja (homeopathic medicine) was found to be most effective therapy combined with surgical excision. Zero cure rates were noticed in the wart after four weeks of treatment using prepared autogenous vaccine, autohemotherapy and ivermectin. This allowed recommending this method for treatment of cutaneous papillomatosis but only in cases where the specific conditions, available options and clinical signs (size, shape, location) were appropriate for total excision. When the localization, the number and size of growths are not eligible for surgery, the use of autogenous vaccine is another treatment option but possibly, it could be applied over a prolonged period of time.

References

Burks, R. I. (1998, February). Povidone-iodine solution in wound treatment. *Physical Therapy Rehabilitation Journal, 78*(2), 212-218. doi:https://doi.org/10.1093/ptj/78.2.212

Campo, M. S. (1995, March). Infection by bovine papillomavirus and prospects for vaccination. *Trends in microbiology, 3*(3), 92-97. doi:0.1016/S0966-842X(00)88889-7

Carvalho, B. S. (2012). Detection of bovine papillomavirus types, co-Infection and a putative new BPV11 subtype in cattle. *Transboundary and Emerging Diseases, 59*(5), 441-447. doi:10.1111/j.18651682.2011.01296.x.

Drinayaev VA, M. V. (2004). Anti-tumour activity of ivermectin. *European Journal Pharmacol, 501*, 19-23.

G. S. Z. Ssenyonga, J. S.-S.-O. (1990). Therapeutic value of partial excision of lesions combined with administration of an autogenous vaccine during an episode of cutaneous papillomatosis in cattle of Uganda. *Journal of American Veterinary Medical Association, 197*(06), 739-740.

G. Terziev, R. R. (2015). Papillomatosis in heifers-Comperative studies on surgical excision and autogenous vacccine therapies. *Trakia Journal of Sciences, 13*, 274-279.

Geeta Devi Leishangthem, R. S. (2008). Detection of bovine papillomaviruses in cutaneous warts in cattle. *Indian J. Vet. Pathol., 32*(1), 15-20.

Jarrett, O. G. (1990). Studies on vaccination against papillomaviruses: the immunity after infection and vaccination with bovine papillomaviruses of different types. *The Veterinary Record, 126*(19), 473-475.

Joaquin Barca, Y. H. (2021, January). Increase in white blood cell counts by pegbovigrastim in primiparous and multiparous grazing dairy cows and the interaction with prepartum body condition score and non-esterified fatty acids concentration. *PLOS ONE, 16*(1), 1-13. doi: https://doi.org/10.1371/journal.pone.0245149

Kumar, P. N. (2015). Detection of bovine papilloma viruses in wart- like lesions of upper gastrointestinal tract of cattle and buffaloes. *Transboundary and Emerging Diseases, 62*(2), 264-271.

M Nooruddin, A. D. (1984). Prevalence of dermatophytosis in cattle of Bangladesh. *Indian Journal of Veterinary Medicine, 4*(1), 25-27.

M. Sreeparvathy, C. H. (2011, December). Autogenous vaccination as a treatment method for bovine papillomatosis. *Journal of Livestock Science, 2*, 38-40.

Madrewar. (n.d.). Therapeutics of veterinary homeopathy and repertory. B. Jain Publishers, 2nd Ed.New Delhi, India.

Mesfin Mathewos, T. T. (2021). Cytopathogical characterization of papillomatosis in cattle of Wolaita Sodo district, Southern Ethiopia. *Elsevier, 13*, 1-9.

MS Campo, M. M. (1985). The presence of bovine papillomavirus type 4 DNA is not required for the progression to, or the maintenance of, the malignant state in cancers of the alimentary canal in cattle. *The EMBO Journal, 4*(7), 1819-1825.

Mukherjee, D. J. (2013). Theraputic management of bovine cutaneous papillomatosis with ivermectin in farm bred calf crops of West Bengal, India. *Exploratory Animal and Medical Research, 3*(2), 123-130.

Ogawa, T. O. (2004). Broad-spectrum detection of papillomaviruses in bovine teat papillomas and health teat skin. *Journal Gen Virol, 85*, 2191-2197.

Pangty K, S. S. (2010). Detection of BPV-1 and-2 and quantification of BPV-1 by realtime PCR in cutaneous warts in cattle and buffaloes. *Transboundary Emerging Diseases, 57*, 185-196.

Patnayak. (2004). Autohemotherapy in bovine papillomatosis. *Intas Polivet, 5*, 16-17.

Pence, M. E. (2005). Cattle warts-and show cattle.

Plumlee, K. H. (2004). *Clinical veterinary toxicology.* doi:https://doi.org/10.1016/B0-323-01125-X/X5001-8

Ranjan, R. G. (2013). Efficacy of autogenous vaccine and autohemotherapy in bovine cutaneous papillomatosis. *Intas Polivet, 14*, 411-415.

Roperto, B. E. (2012). Productive infection of bovine papillomavirus type 2 in the placenta of pregnant cows affected with urinary bladder tumors. *PLOS ONE, 7*(3), 1-9. doi:org/10.1371/journal.pone.0033569

S.N. Archana, A. P. (2019). Bovine papillomatosis and its treatment under farm condition. *International Journal of Current Microbiology and Applied Sciences, 8*(4), 2880-2884.

Schmitt, F. M. (2010). Prevalence of BPV genotypes in a German cowshed determined by a novel multiplex BPV genotyping assay. *Journal Virol Methods, 170*, 67-72.

Silvestre, B. N. (2009). Bovine papillomavirus type 1 DNA and E5 oncoprotein expression in water buffalo fibropapillomas. *Veterinary Pathology Online, 46*(4), 636-641.

Singh, S. &. (2009). Papillomatosis in Indian cattle: occurrence and etiopathology. *Indian Journal of Veterinary Pathology, 33*(1), 52-57.

Tagesu, A. (2018). Examination of urine. *International School of Veterinary Science and Research*, 51-58. doi:http://dx.doi.org/10.17352/ijvsr

Turk, Z. S. (2005). Severe bovine papillomatosis: detection of bovine papillomavirus in tumor tissue and efficacy of treatment using autogenous vaccine and parammunity inducer. *Veterinarski Archiv, 75*, 391-397.

West, H. J. (1997). Clinical and pathological studies in cattle with hepatic disease. *Veterinary Research Communications, 21*(3), 169-185.

Acknowledgements

The author wishes to acknowledge the immeasurable grace and profound kindness of Almighty "ALLAH" the supreme authority and supreme ruler of universe, who empowers the author to complete the work successfully.

The author sincerely acknowledges to Professor Dr. Lutfur Rahman, Vice-chancellor and Dean, Faculty of Veterinary Medicine, and Professor Dr. AKM Saifuddin, Director, External Affairs, CVASU for providing this research exposure through one-year long internship program.

The author feels proud in expressing his deep sense of great gratitude and indebtedness to respected teacher and tutor Dr. Mohammad Mizanur Rahman, Professor, Department of Medicine and Surgery, Faculty of Veterinary Medicine, Chittagong Veterinary and Animal Sciences University for his trustworthy and scholastic supervision.

Special gratitude to the farmer for his cooperation throughout the data collection for the study.

The author also would like to expense deepest sense of respect and appreciation to DR Tishita Sen Ape, MS student, Department of Medicine and Surgery for her cordial co-operation.

The Author

December, 2024

Biography

This is Nurul Amin from Cox's Bazar , Chattogram. He completed his Secondary School Certificate (SSC) examination in 2016 with from Khutakhali High school, Chakaria,Cox's Bazar and Higher Secondary Certificate (HSC) examination in 2018 from Gachhbaria Govt. College,Chandhanish. Currently he has been doing his internship program which is the compulsory of DVM degree under the Faculty of Veterinary Medicine, Chittagong Veterinary and Animal Sciences University . His favorite hoppy is reading newspaper and doing social work. He feels massive interest in the livestock farm management and commercial poultry farm.