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Minimally Invasive Techniques and Diagnostic Approaches for Milk Flow Disorders in Bovine

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Abstract:- Udder health plays an important role in modern dairy farming and is the basis for an economical and hygienic milk production process. Milk flow disorders are a central problem in the field of udder health. Consequently leading to a loss in milk production, detrimental changes to the milk components and raw milk quality. Therefore, a rapid and accurate diagnosis and prognosis is mandatory in patients with udder diseases, and requires the use of state-of-the-art examination techniques and therapeutic treatments. While many studies had been done on teat morphology of machine milked animals, no major studies are available for hand milked cows. Interpretation of ultrasonographic measurement is very crucial for better understanding on the recovery of teat tissue before and after milking. Towards this ultrasonographic assessment of teat morphology was undertaken on milked cows. Ultrasonography of the teat was found to have better diagnostic yield in the detection of Milk Flow Disorder when compared to clinical detection as well as in location of the lesion and in identifying the type of lesion.

I. INTRODUCTION

The dairy industry in India has emerged as a significant sector within the agrarian economy, with milk ranking as the second most substantial agricultural commodity in terms of GDP contribution, following rice. One of the primary sources of income and the foundation of an agriculture-based society is milk production. Teat injuries are a common occurrence in dairy animals, with an estimated 20% incidence rate (Agger & Hesselholt, 1986). Animals with high lactation production are more susceptible to injuries. At this time, the primary concerns of dairy producers are the maintenance of efficient udder health and the production of clean milk for financial gain. Economic losses can result from teat affections due to treatment expenses, reduced milk production, and an elevated risk of mastitis (Agger & Hesselholt, 1986). Additionally, premature elimination from the herd can further compound these issues (Beaudeau *et al.,* 1995). Ensuring the health of the mammary organ is critical for modern animal husbandry to attain high-quality milk production (Wendt *et al.,* 1994). Regarding udder health, both the quantity and quality of milk produced are directly proportional. The challenges that result in a decline in milk production are a bane for dairy farms. Teat affections may be categorised as either open or closed wounds. In contrast to open injuries to the teat, which involve wounds such as lacerations, fistulae, and gangrene that are visible on the surface of the skin, internal injuries affect the cisterns of the teat and the papillary ducts (Sreenu *et al.,* 2014). In closed teat affections, the outer skin remains undamaged and normal, while the injury is situated within the teat. Injuries to covered teats may result in teat stenosis and milk flow disorders (Kubicek, 1976). External teat injuries are more prevalent as a result of several factors, including the anatomical location of the teat, an expansion of the udder and teat size during lactation, suboptimal milking techniques, repetitive trauma to the mucosa of the teat, damage to the veal teeth, self-infliction, contact with barbed wires, spiny bushes, or other animals stamping on the teat, among others (Tiwary *et al.,* 2005; Singh *et al*., 2017). A precise diagnosis is an essential prerequisite for the successful implementation of surgical therapy. Current diagnostic approaches for teat affections rely on clinical observations and patient histories, which are insufficient and imprecise. Therefore, theloscopy (Geishauser *et al.,* 2005; Singh *et al*., 2017) and ultrasonography (Fasulkov *et al.,* 2014) may be crucial for accurately determining the anomaly. Ultrasonography is a non-invasive, radiation-safe technique that is exceptionally valuable in the diagnosis of a wide range of physiological and pathological soft tissue conditions. Carteeet *et al.* (1986) initiated B-mode USG of the mammary gland for the diagnosis of milk flow disturbances and structural changes in the teat of cows using a 5–10 MHz sector transducer. The utilisation of B-mode USG enables the differentiation of various morphological structures, including the parenchyma of the mammary gland, teat and gland cisterns, teat canal, and rosette of Furstenberg (Franz *et al.,* 2001). Both direct and water bath ultrasonography have been utilised by Cartee et al. (1986), Franz et al. (2003), and Güngoret et al. (2005). Ultrasonography has been recommended as the preferred technique for diagnosing milk flow disorders, including pathological changes such as stenosis, inflammation, foreign bodies, and hematomas, as well as measuring teat canal length, teat cistern diameter, and teat wall thickness with favourable outcomes (Fasulkov, 2012). However, it should be

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noted that ultrasonography does not offer any insight into the lesion's external aspect. This drawback can be addressed through the application of theloscopy. Theloscopy, also known as teat endoscopy, is an expedient, minimally invasive, and atraumatic technique that does not result in any post-operative complications, including haemorrhage or mastitis (Tulleners & Hamir, 1990). Theloscopy enables accurate localization of the lesion within the teat that is causing changes in milk flow. It offers a clear depiction of the mucosa, which provides insight into the severity and extent of the injury (Rathod *et al.,* 2009). Surgical interventions involving theloscopy are employed to address conditions such as stenosis caused by tissue fibrosis (Franz *et al.,* 2009). Recently, theloresectoscopy has become increasingly prevalent in the treatment of concealed teat injuries. Bleul *et al.* (2005) utilised a combined instrument consisting of a rigid endoscope and a high-frequency snare to manage teat flow obstructions. This approach resulted in improved outcomes as the high-frequency loop assisted in resecting the obstructing tissue and reduced tissue hyperplasia in structures formed by scar tissue. (Joth *et al.,* 2018) Minimally invasive loresecetoscopy is the method of preference for the treatment of covered teat injuries. Disorders of milk flow that are caused by ruptured and dislocated tissue of the teat canal, papillomas, polyps, and so forth, can be precisely removed via teat endoscopy. By restoring milk flow and yield in the afflicted quarter with minimal economic loss to the farmer and reduced risk of mastitis predisposition, minimally invasive theoresectooscopic surgery improves the quality of the milk produced (Taponen & Pyorala, 2009). As a result, video-assisted theloscopic electro-resection (theloresecetoscopy) may be regarded as a novel and promising alternative to traditional surgical techniques when treating covered teat injuries in valuable dairy cattle.

II. MATERIALS AND METHODS

The incidence of teat affections related to milk flow disorders in cows and buffaloes reported to the Large Animal Surgical Outpatient Unit, to the Department of Veterinary Surgery and Radiology of Nagpur veterinary college, Maharashtra Animal and Fishery Sciences University, Nagpur between August 2022 to January 2024. Details of the etiological occurrence and side/quarter affected have been documented in cows and buffaloes.

Table 1: Table Showing of Animals and Grouping

Group I : Open Teat Affections								
Group I	**Open Teat Affections**			**Animals**				**Total Teats Involved (n= 30)**
				Cows (n=22)	**Teats (n=22)**	**Buffaloes (n=8)**	**Teats (n=8)**	
Ia	Teat fistula			5	5	2	2	7
Ib	Teat laceration	i	Distal end	7	7	2	2	9
		ii	Mild	5	5	1	1	6
		iii	Full teat	3	3	1	1	4
Ic	Teat Tip Injury			2	2	2	2	4
Group II : Covered Teat Affection								
Group IIa	**Congenital Anomalies**			**Animals**				**Total Teat involved (n=8)**
				Cows (n=5)	**Teats (n=5)**	**Buffaloes (n=4)**	**Teats (n=4)**	
IIa1	Supernumerary teat			2	2	1	1	2
IIa2	Imperforated teat			2	2	2	2	4
IIa3	Teat spider			1	1	2	2	2

A. Clinical Evaluation of the Patient

A comprehensive clinical examination was performed on each animal in order to identify the underlying causes of milk flow disorders, which are detailed below.

B. History and Signalment

Following information regarding patient history and signalment was recorded:

- Age: The age of the animals were recorded in years
- Species: The species was recorded as cow/buffalo.
- Stage of lactation or Parity: If the animal had already parturated, value was recorded as positive where as if some days were still remaining in parturition, it was recorded as negative.
- Number of lactations/parities was recorded in all the animals.
- Quarter affected: The quarter affected was recorded and labeled as left fore (LF), left hind (LH), right fore (RF) or right hind (RH).
- Milk yield: The total milk yield of the quarter in milliliters were recorded.
- Duration of milk flow disorders: Was recorded as number of days from observation of illness by the owner to presentation at university clinics

C. Gross Examination

Upon gross examination of the affected tissue, it was determined that the lesion was present in the left fore and rear quarter, as well as the right fore and hind quarter of the mammary gland. The inclusion criteria encompassed includes pain upon contact, swelling, complete or partial obstruction in any region of the teat, and a comparison between the affected and contralateral teats. The affected tissue was palpated by rolling it between the thumb and index finger, beginning at the base and progressing towards the apex. Any abnormality, fibrosis, or nodule observed in the teat cistern or teat canal was documented.

Fig. 1: Instruments used for Somatic Cell Count, DELTA Instrument, BV Kelvinlaan 3,9207 JB Drachten, The Netherland used for Somatic Cell Count

Fig. 2: Plastic Paddle with Four Cups Marked as LF, LH, RF and RH used for CMT

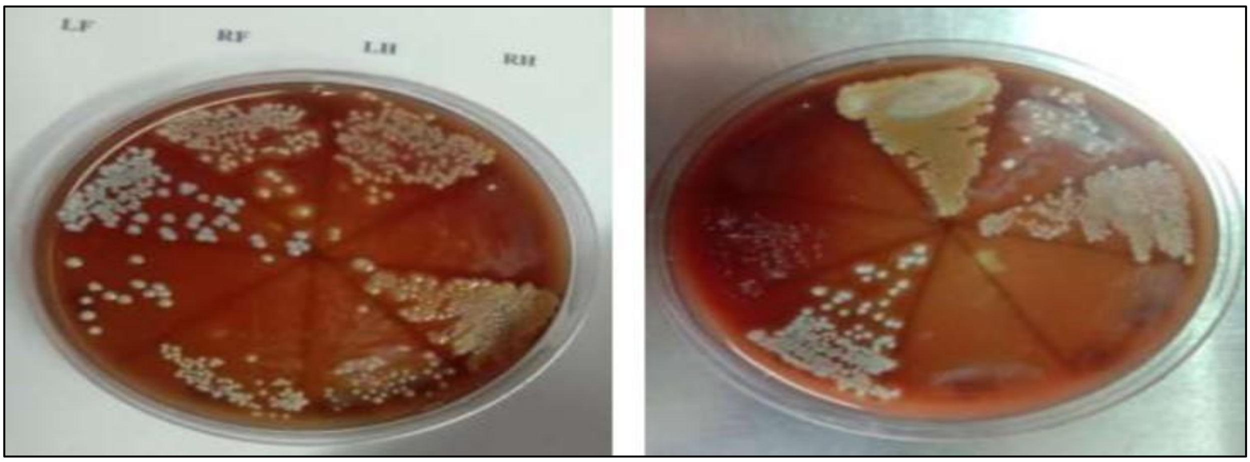

Fig. 3: Bacterial Isolation by Direct Plating on Blood Agar Revealed Hemolytic and Non-Hemolytic Colonies of Staphylococcus sp.

D. Haematology

Using the Advia 2120 Haematology System, the hemological parameters haemoglobin (gram %) and total leukocyte count (x 10^3/L) were determined. The differential leukocyte count (cells/L) was determined using a thin blood smear stained with Leishman's stain in the Veterinary Diagnostic Laboratory of Nagpur Veterinary College, Nagpur @ 100X power Olympus BX51 microscope was used to observe the cells.

E. Milk Quality Assessment

The teats were carefully dried and cleansed before milk samples were collected. After soaking cotton wool in 70% alcohol (spirit), the teat opening was cleansed. After discarding the initial milk streaks, 15 millilitres (ml) was carefully measured out and placed into sterile, marked test tubes for each quarter sample. Within one to two hours of collecting the milk samples, they were transported to the mastitis lab of Nagpur Veterinary College, Nagpur for further examination.

F. Somatic Cell Count

A milk somatic cell counter from DELTA Instrument, BV Kelvinlaan 3, 9207 JB Drachten, The Netherlands, was used to find SCC in milk samples. A sample of about 1ml of warm milk was needed by the machine. It took about 40 seconds to do each test. A Light Emitting Diode (LED) was used as the light source for the Soma Scope Smart cell counter. Flow cytometry was used to count somatic cells from the flow of cells through the flow cell using a dual detection method. It was given in terms of 10^3 cells/ml.

G. California Mastitis Test

The standard method explained by Pandit and Mehta (1969) was used to do the California Mastitis Test (CMT) and figure out what it meant. The test was done in a plastic paddle that had four cups marked LF, LH, RF, and RH. Equal amounts of milk and test solution (2–3 ml each) were added. When the paddle was moved in a circle over the flat surface, the contents were fully mixed. In less than 10 seconds, the results were recorded and interpreted as

Table 2: Reference Table of California Mastitis Test

Outcome	Score	Condition
No mastitis	-	Liquid with no precipitate
Doubtful	+	Traces of precipitate which disappeared quickly or precipitate with little gel
Positive	++	Precipitate thickness and moves towards the centre on swirling. When movements were stopped, mixture levelled again covering bottom of the cup
Strong positive	+++	A distinct gelling which tended to stick to bottom of the paddle and a central was seen during swirling

H. Bacterial Isolation and Identification

It was possible to separate bacteria from cow and horse milk samples by plating them directly on Muller Hinton agar, blood agar, and nutrition agar. The colony shape was looked at after 24 hours of incubation at 37ºC, and the separated organisms were labelled as either Gram positive or Gram negative bacteria using the Gramme staining method. The bacterial colonies were then grown in a selective medium, like Mannitol Salt agar for Staphylococcus sp., Edwards medium for Streptococcus sp., MacConkey agar for Enterobactor sp., and others. The cultures were then lifted on blood agar slants and identified by their shape, opacity, consistency, surface, size, coloration, and how the media changed. To make sure the results were correct, microscopic, cultural, and biochemical traits were used.

I. Antibiotic Sensitivity Test

For the antibiotic susceptibility test, Bauer's method was used (Bauer et al., 1959). A sample of milk was put into nutritional soup and kept at 37^0 c for 24 hours. A clean cotton swab that had been soaked in broth culture was spread out evenly on a plate with 5% cow blood agar. Antibiotic discs were then put over the spot that had been infected. Antibiotics like doxycycline, cefaperazone, ampicillin, gentamicin, amoxicillin-sulbactum, cloxacillin and cefaperazone were used. The area where growth was stopped after 24 hours at 37^0 c was called "sensitive," and the area where growth was not stopped was called "resistant." As part of the treatment, the drug that killed the microorganisms the fastest was picked.

J. Bromothymol Blue (BTB) Paper Test

The idea behind this test comes from the fact that when a cow has mastitis, bicarbonate salts from the blood enter the milk and raise its pH. This card had a drop of quarter milk on it. The colour of the dye changed from yellow (normal) to greenish-yellow (+), green (++), and blue (+++), based on how healthy the quarter was and its pH.

Table 3: Reference Table of Bromothymol Blue Paper Test

Outcome	Score	Condition
No change in color	-	Healthy
Colour turning light green	+	Mild sub clinical condition
Colour turning dark green	++	Moderate sub clinical condition
Colour turning blue	+++	Clinical condition

K. pH

Using a digital pH meter Mettler Toledo, Five Easy Plus, the pH of the milk was determined.

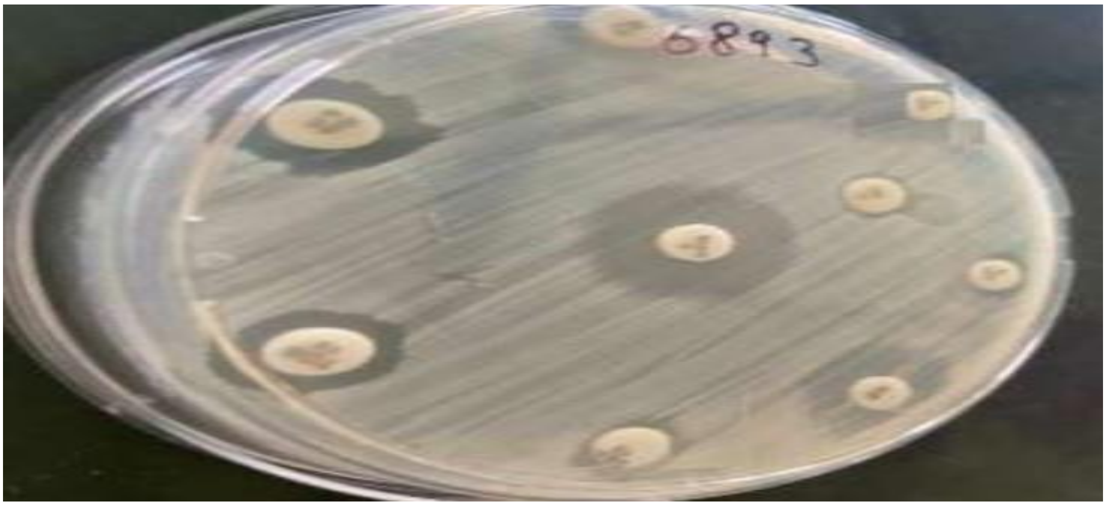

Fig. 4: Antibiotic Sensitivity Test used to Record the Zone of Growth Inhibition as Sensitive and Lack of Growth Inhibition as Resistant

Fig. 5: Milk Tested by using Bromothymol Blue (BTB) Paper

Fig. 6: Placement of Cassette at the Inter-Mammary Groove for Radiography

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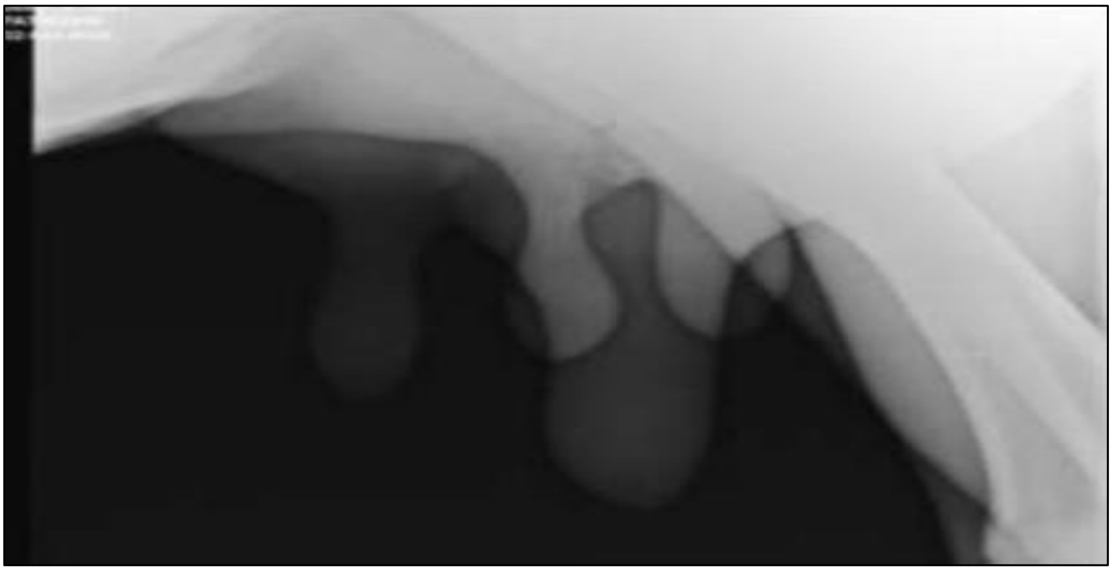

Fig. 7: Plain Radiograph of the Teat

III. RADIOGRAPHIC EXAMINATION OF NORMAL AND DISEASED TEAT

A. Preparation and Positioning of the Animal

Radiographs with plain and negative contrast were used on the animals in groups I and II. The x-ray research in this study used 1000 mAs and 150 kV from the Modern Prorad Prognosys Company. The x-rays were looked at with a Kodak Direct View Classic computerised radiograph machine. The teat that was going to be looked at was washed, scrubbed, dried, and then 70 percent medical spirit was used to remove any grease. The animal was laid on its side with the mammary gland that needed to be studied facing the X-ray machine. The leg was kept stretched at the back so that the structures of the limb wouldn't get in the way of the images. The tape was put in the groove between the breasts and behind the parts of the mammary gland that were hurt. There was no trouble with the plain x-ray study of the udder and teat, but it did not show any obstructive lesions or pathological changes in the tissues of the udder and teat. The x-rays of the udder and teat tissues showed them as solid grey tones with no features in the soft tissues. Radiographic procedures with negative contrast were also done on the first day of the study. There was a clean teat cannula put in, and the milk from the teat was taken out. In order to do negative contrast x-rays, air from the atmosphere was pumped into the teat as a contrast medium. A 50 ml empty syringe was used to add the air. The needle was pulled out and side x-rays were taken after the teat had been inflated enough, which could be seen by the syringe's distension and backpressure. The results were analysed and written down.

B. Ultrasonography

The hair on top of the udder was shaved off. The udder and teats were washed to make sure they were clean, dried with a clean cloth, and cleaned with 70% surgical spirit. They were then ready for the ultrasound test. An ultrasound machine called GE Logiq 3 BT Expert was used for the procedure. The direct contact method or the water bath technique were used, as explained by Franz et al. (2001) and Fasulkov (2012), respectively. To do ultrasonography of the teat, the animal had to be held in place in the trevis while it was standing. The milk man's knot was used to hold animals down. Before the scan, savlon (Aceptik Hospital concentrate) was used to shave and scrub each area of the udder.

In the direct contact method, an acoustic gel was used to put the transducer directly on the teat. There was a linear transducer at 12 MHz and a curved transducer at 5 MHz that were used to scan the teat. It was possible to take both transverse and longitudinal images. The teat was put in a plastic container full of warm water (30–35°C) for the water bath method, and the transducer was placed on top of the container. A gel layer was put on the probe head to help it stick to the plastic glass better. The probe was moved up and down until a clear picture of the teat showed up on the US screen. The scanning began at the tip of the teat and moved forward slowly to the base of the teat. Picture depth on the screen was kept between 4 and 6 cm for teat. Since having milk in the teat improved the quality of the picture, extra care was taken to keep milk from leaking out of the teats that were hurt during the clinical examination. Along with the damaged teat, the contralateral side was also scanned to make a comparison.

C. Sonographic Measurements of Teat Structures

All the animals were scanned prior to milk drainage from the teats. However, in teat fistula cases, the milk drained spontaneously through the fistulous tract so animal was scanned immediately on presentation. After achieving a satisfactory image on the ultrasound machine, it was saved. The measurements of the teat anatomical structures were done by a single observer in all the teats using same technique in all the animals as recommended by Neijenhuis et al. (2001). The parameters measured were as follows:

- Teat Canal Diameter (TCD): This was recorded as the width of hyperechoic lines measured at the half length of the teat canal.
- Teat Diameter (TD 1) was measured at the area of Furstenberg's Rosette.

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- Teat Diameter (TD 2) was measured 1.5cm - Proximal to the area of Furstenberg's Rosette .
- Teat Cistern Diameter (TCsD): This was recorded as the length on a perpendicular line drawn at the level of 1-1.5cm from the area of Furstenberg's Rosette which ended at mucosa of opposite teat wall.
- Teat Wall Thickness(TWT): This was measured at the level where TCsD was recorded. This was recorded by measuring distance of mucosa to the skin
- Teat canal length (TCL): This was measured at the level area of Furstenberg's Rosette to teat tip.

Fig. 8: Restraint of Animals in Standing Position in the Trevis after Applying Milkman's Knot

Fig. 9: Scanning the Teat by Direct Contact Technique and Water Bath

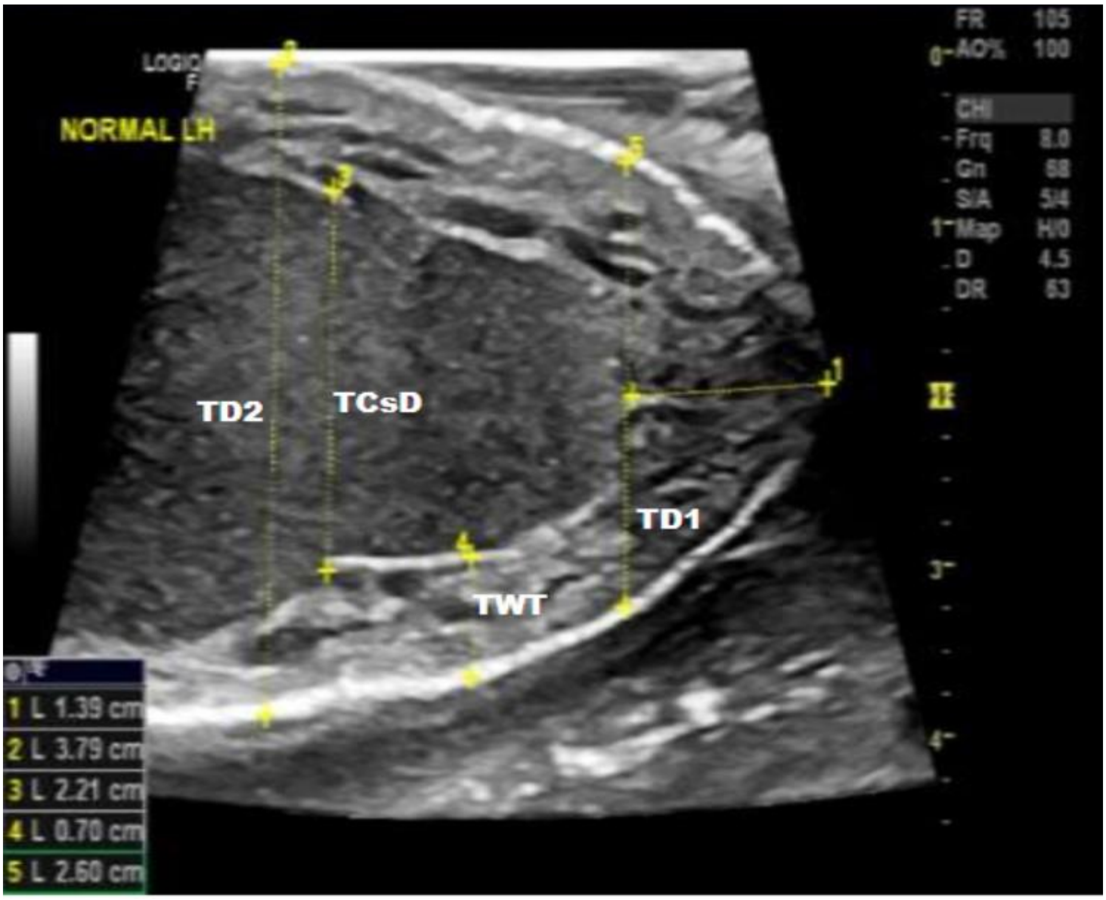

Fig. 10: Measurement of Structures of Teat Cistern

- TD 1 – Teat Diameter measured in the area of Furstenberg's Rosette
- TD 2 - Teat Diameter measured 1.5 cm proximal to the area of Furstenberg's Rosette
- TCsD – Teat Cistern Diameter
- TWT- Teat Wall Thickness

Fig. 11: Measurement of Structures of Teat Canal using inbuilt Calipers

- TCL - Teat canal length, TCD – Teat Canal Diameter

Fig. 12: Seeh/ Hospes Theloresecoscopy Set used for Detailed Evaluation of Teat Canal and Teat Cistern

Fig. 13: Assembled Theloresectoscopy Unit

D. Theloscopy

After a general checkup and an ultrasound, the chosen animals' teat tube and teat cistern, along with any problems with milk flow, were carefully looked at using a theloscopy.

E. Instrumentation

Endoscopy of the injured teat was done with the Seeh/Hospes theloresectoscopy set (Karl Storz Gmbh & Co. KG, Tuttlingen, Germany). Figure 1 shows the parts of the loresectoscopy unit and Figure 2 shows the unit put together.

IV. INSTRUMENTATION

A. Theloscopic and Theloresectoscopic Instrument Set Included:

Theloscopic and theloresectoscopic instrument set included:

- Theloscope (Hopkinsoptic, 0º, 1.9/2.1mm, 18cmlength, Tuttlingen, Germany)
- Operating unipolar cautery with circular loop.
- LUCER-lock adaptor with monopolar cautery unit.

- Sleeve (Metal schaft, 11charr and Metal sheath, 11 Fr).
- Endovision TELE PACK VET X an is a compact system that combines lighting, picture orientation, image processing, and a full treatment documentation software.
- USB stick (data storage).
- Unipolar and Bipolar cautery Device with cutting and coagulating processing unit.
- Cutting and Coagulating Cautery foot switch.
- TELECAM 1-chip camera head with 2 freely programmable camera head buttons, colour system PAL, with integrated focal-zoom, focal length f = 25-50mm (2x). TELECAM 2-camera control unit

B. Preparation of the Animal

For the theloscopic exam, the animal had to go without food for at least 24 to 36 hours before the operation. Keep the injured quarter of the animal above while it is lying on its side. Before the process starts, wash the udder and teats with savlon (Aceptik Hospital concentrate). Once the teat was clean and germ-free, a soft rubber patch was put around the base of it.

C. Preparation of the Instruments

All of the parts of the ophthalmology set were cleaned with pure water that was kept at room temperature. For sterilisation, all of the tools used for the endoscopy were put in the formalin room.

D. Anaesthesia

Hold the animal so that it is lying on its side.At the base of the teat, 5–10ml of 2% lignocaine hydrochloride was used as ring block anaesthetic to make the area less painful. Before putting the theloscope into the teat tube, lignocaine gel should be put on it. When needed, 0.1 mg/kg b.wt. i/m of xylazine hydrochloride was used to put the animal to sleep.

E. Theloscopic Examination

Once the theloscopic tools were put together, the light source was turned on and the scope moved forward. The endoscopic process was shown as a continuous moving picture. The picture was taken, and the details were studied and analysed.

Axial theloscopy was used to fix the surgical problem with the blocked teat lesion in the first place. Geishauser et al. (2005) say that the axial method was used to do the laparoscopy.

After holding the animal in place, a clean theloscopic canula with a hard trocar was put in through the teat canal. With the help of a blunt trocar, the teat tube was made big enough. Keep the canula in place while you take out the trocar. The canula was pulled out until it reached the tip of the teat tube, and milk was drained from the teat cistern. The theloscope's white balance was set by comparing it to a white gauge piece or cotton before the scan began. Through the tube, the theloscope was carefully put into the teat canal after it had been put together. The Telepack Vet X unit's insufflation pump was used to fill the teat tank up to level 2 of the machine's settings.

The teat cistern and teat stream were looked at from the very top and worked their way down. On the camera piece, manual settings were used to focus on the target spot. By dipping the endoscope in distilled water or standard saline, any blood or foreign bodies were taken out. Once the lesions are found, the pictures should be saved. After the laparoscopy was over, normal saline solution was used to flush the teat, and antibiotics were introduced into the quarter. Take the theloscopy set apart, clean it with distilled water, and then put it back together in a formalin box.

F. Treatment

For the treatment of underlying conditions, standard methods were used based on the size, type, and location of the abnormality.

Theloresectoscopic surgery was done once the location and size of the blockage were confirmed. After that, a clean baby feeding tube about 9 to 10 numbers long was put into the teat canal and brought all the way up to the glandular cistern. Adhesive tapes were put around the teat to hold the catheter in place, and the tip of the catheter was sealed off. An unused baby feeding tube was put into the teat canal of 9–10 babies and entered all the way to the glandular cistern before this treatment. The baby feeding tube was put over the teat with sticky tape, and the end was sealed. told owner to open the baby feeding tube's cap twice a day to drain the milk. Gentamicin sulphate (5ml, 200 mg) was infused into the surgical teat through the catheter. To stop an infection from spreading, a column of Gentamicin sulphate was left in the catheter.

For animals with full teat fibrosis and udder fibrosis, it was suggested that they take 6 grammes of potassium iodide powder by mouth every day for a week and 2 grammes of serratiopeptidase every day for two weeks. A teat tumour remover was used to get rid of the fibrous tissue in the case of everted mucosa. After the teat hole was made surgically, a clean feeding tube for babies was put into it and held in place. When the teat tip was hurt, it was cleaned out. Since the teat tube was open, the skin on the outside of the teat was sewn back together. A teat plug was then put in the teat tube.

When animals were found to have extra teats, they were medically removed before they started nursing. The surgery wound was stitched up as usual. Teat dilator was used to loosen up the sphincter muscles and Hug's knife was used to make the teat opening bigger when the teat wasn't punctured. Then, a teat syphon was put into the area where the teat canal was supposed to go to make the hole bigger and let the milk drain. After the teat hole was made surgically, a clean feeding tube for babies was put into it and held in place.

A mix of ampicillin and cloxacillin (AC-VET Intas Pharmaceuticals Ltd.) was given intramuscularly twice a day at a dose of 10 mg/kg body weight, and enrofloxacin (Fortivir, Virbac) was given intramuscularly once a day at a dose of 5 mg/kg body weight. Meloxicam (Melonex, Intas Pharmaceuticals Ltd.) @ 0.2 mg/kg b.wt. I/M was given once a day for three days. Follow-up calls were made at the right times (0-day, 15-day, 30-day, and long-term follow-up).

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V. RESULTS AND DISCUSSION

The present study was carried on 40 animals with milk flow problems that were brought to the Department of Veterinary Surgery and Radiology, Nagpur Veterinary College, Nagpur. The animals were 27 cows and 13 buffaloes. A total of 60 teats were studied (48 cows and 12 buffaloes). The animals were divided into two groups based on the type and location of the affection diagnosed below:

Table 4: Table Showing of Animals and Grouping

Group I : Open Teat Affections								
Group I	**Open Teat Affections**			**Animals**				**Total Teats Involved (n= 30)**
				Cows (n=22)	**Teats (n=22)**	**Buffaloes (n=8)**	**Teats (n=8)**	
Ia	Teat fistula			5	5	2	2	7
Ib	Teat laceration	i	Distal end	7	7	2	2	9
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		iii	Full teat	3	3	1	1	4
Ic	Teat Tip Injury			2	2	2	2	4

Group II: Covered Teat Affections						
Group IIa	**Congenital Anomalies**	**Animals**				**Total Teat involved (n=8)**
		Cows (n=5)	**Teats (n=5)**	**Buffaloes (n=5)**	**Teats (n=4)**	
IIa1	Supernumerary teat	2	2	1	1	2
IIa2	Imperforated teat	2	2	2	2	4
IIa3	Teat spider	1	1	2	2	2

A. Etiology

The most common etiology of milk flow disorders was established as physical trauma in 55% of the total affections in cattle and 45% in buffaloes. The teat affections due to trauma included teat fistula, teat laceration, everted mucosa due to machine milking and teat tip obstruction due to injury.

B. pH

A digital pH metre from Mettler Toledo called Five Easy Plus was used to record the pH of the milk. The average pH levels found in two groups of teat affections as a follow. For group I, the pH level on the day of introduction was 6.87±0.12, on the 12th day after surgery it was 6.90±0.09, and on the 30th day after surgery it was 6.81±0.18. The mean pH for group II on the day of appearance was 6.89±0.11, for the 12th day after surgery it was 6.94±0.06, and for the 30th day after surgery it was 6.15±0.15.

C. Radiographic Examination of Normal and Diseased Teat

For diagnosis of disease condition in the animals, either normal radiography or contrast radiography was used.

D. Plain Radiography

The Simple X-Ray Method for Examining the Teat Was Found to Be Easy, But It Wasn't Always Possible to Show the Obstructive Lesions and Pathological Changes in the Teat. Radiographs of the Udder and Teat Tissues Showed Them as Solid Grey Tones with No Features of Soft Tissues.

E. Contrast Radiography

It was discovered that the contrast study of the teat was a better way to diagnose than the plain radiographic technique. There were some questions about using iodine, a positive contrast agent, in the teat cistern to find the level of the obstructive tumour. This is because the contrast is irritating. Patient who had teats that were partially blocked by the teat tank found this method helpful. To study the diameter and shape of the cistern, the degree to which the teat wall thickness changed, the pattern of the fistulous tract, and to find out how long the teat canal was helpeful. It also used to diagnosed the approximate size and location of the lesion.

The negative study of a normal udder and teat made it easier to see the circular folds of the teat cistern than the positive contrast study. It also made it easier to understand the area of the gland and the teat cistern by showing the pattern of lactiferous ducts. When air was injected into the teat cistern, the cisternal space grew bigger. To find the site of the lesion, x-rays of animals with mid-teeth block were taken before surgery. The infused air didn't go beyond the point of blockage. After theloscopic electroresection was done at the obstruction site, air was pumped into the teat and through the site of the obstruction. On the x-ray taken after surgery, it showed that the obstruction was gone.

F. Ultrasonography

In Present Study, Real-time B mode Grey-scale sonography of the teat was done using either the direct contact method or the water bath technique, which were both described by Franz et al. (2001) and Fasulkov (2012). A GE Logiq BT expert ultrasound machine with a 6-12 MHz linear transducer was used to do grey-scale, real-time B mode ultrasonography on the damaged teat and the normal teat on the opposite side. Using both direct contact and a water bath, the scanning was done in both longitudinal and transverse lines. For examining the teat structures, diagnostic-quality pictures were made with a linear transducer that worked at 6-12 MHz. Though previous researchers had used transducers with different frequencies to study teat structures. These frequencies were 3.5 MHz (Stocker et al., 1989), 8.5 MHz (Franz et al., 2001); (Fasulkov et al., 2014); 5 MHz (Saratis & Grunert, 1993); and 7.5–1.8 MHz linear transducers

(Ramanbhai, 2016). Rambabu et al. (2008) and Ramanbhai (2016) observed that comparing the use of direct contact, water bath, gel application, and standoff methods for ultrasound of the teats in buffalo dogs. It was reported that the images from the water bath and gel application method were good. As Franz et al. (2001) and Will et al. (1990) found, the water bath method of ultrasonography produced good images because the examiner's hand or the transducer did not put any pressure on the teat while scanning it. This meant that the shape of the teat did not change (Franz et al., 2001).

G. Ultrasonography of Normal Contralateral Teat

An 6-12 MHz frequency linear transducer was used to do real-time, gray-scale B mode ultrasound of the usual contralateral teats. Both transverse and longitudinal parts were looked at using the direct contact and water bath methods.

The three layers of the teat wall—the innermost layer, which is hyperechoic (mucosa), the middle layer, which is hypoechoic (muscular layer), and the outermost layer, which is hyperechoic (skin)—were easy to see on the scan. The teat cistern opening filled with milk looked like an anechoic structure. There was a thin, bright (white), hyperechoic line and two horizontal, thick (dark gray-black), hypoechoic bands on either side. This was called the teat canal. The area between the teat cistern and the teat canal, which looked like a hyperechoic structure, was named the Rosette of Furstenberg. The venous ring of the rosette of Furstenberg was made up of anechoic vessels that met at their edge. A hollow spot in the middle part of the teat wall was made up of blood vessels. In the area where the teat cistern meets the gland cistern, a circular fold forms. Franz et al. (2001) and Fasulkov et al. (2014) both wrote about studies that were similar.

Ultra sonography showed that the shape of the teat looked like it did in studies by Cartee et al., 1986; Franz et al., 2001. A 3.5 MHz linear sensor was used by some researchers, who described the teat canal as a hypoechoic, poorly defined area (Stocker et al., 1989). The low frequency transducer they used might have caused the bad image clarity in their study because it doesn't have very good resolution.

H. Ultrasonography of the Affected Teat

In group I, ultrasonography of the animals' fistulated teats showed breaks in the layers of the teat wall, which means the tract is fistulous. In these cases, the teat canal didn't look like an anechoic lumen; instead, it looked like it had collapsed. It could be because milk is leaking out of the fistulous hole on its own. An ultrasonographic study by Singh (2018) also found similar results in cases of teat fistula. In group I, an ultrasonographic examination of the cut teat revealed a break in the teat wall and the presence of a hyperechoic area at the injury site, which showed that the wound was healing.

In group II, ultrasonography of the extra teat in animals showed that the teat cistern's opening was collapsed and not echoing. As a hyperechoic line, the teat canal could not be told apart. The gastric cistern and the teat cistern could not talk to each other. The reason for this might be that the animal was brought in when it was young and had not its first parturition.

So, there was a possibility that all the teat structures were not completely developed in the teat at the time of presentation of animals. Supernumerary teats were deformed and did not have a potential for delivering milk.

I. Sonographic Measurements of Teat Structures

All the animals were scanned prior to milk drainage from the teats. However, in teat fistula cases, the milk drained spontaneously through the fistulous tract so animal was scanned immediately on presentation. After achieving a satisfactory image on the ultrasound machine, it was saved. The measurements of the teat anatomical structures were done by a single observer in all the teats using same technique in all the animals as recommended by Neijenhuis et al. (2001).

The parameters measured were as follows:

- Teat Canal Diameter (TCD): This was recorded as the width of hyperechoic linesmeasured at the half length of the teat canal.
- TeatDiameter (TD 1) was measuredat the area of Furstenberg's Rosette.
- Teat Diameter (TD 2) was measured 1.5cm proximal to the area of Furstenberg's Rosette .
- Teat Cistern Diameter (TCsD): This was recorded as the length on a perpendicular line drawn at the level of 1-1.5cm from the area of Furstenberg's Rosette which ended at mucosa of opposite teat wall.
- Teat Wall Thickness(TWT): This was measured at the level where TCsD was recorded. This was recorded by measuring distance of mucosa to the skin.

J. Theloscopy

The most important thing for the success of surgery and theloscopic examination is to handle the tools and tissue in a clean way. A theloscopic check was done after milk was drained from the teat that was affectes. The continued irrigation with normal salt made it easier to see things under a microscope. The base of the teat was held in place with a soft rubber band, and a saline-filled teat canal opening made it easier to see the teats. Therefore, lying on the side was better than standing so that milk residue would not leak out of the nipple and make axial theloscopy vision less than ideal. The finding of this study agreed with Medl & Querengasser (1994), Bleul et al. (2005), Geishauser et al. (2005), and Raj (2010).

K. Axial Theloscopy

The teats of the animals that were sick were examined using an axial approach. This method was found to be good for checking the teat cistern and the teat canal, as well as telling different tumours apart. This kind of approaches was used by Adams et al. (1987) and Tulleners & Hamir (1990). Geishauser & Querengasser (2001); Raj, 2010; Jhala, 2009; Walvekar et al., 2017) were among the authors who used both the axial and lateral approaches for theloscopy. It was necessary to keep the animal in a lateral recumbency position and place the injured teat on top of the others, as described by other researchers (John et al., 1998), Seeh & Hospes (1998), Raj (2010), Jhala (2009), and Walvekar et al. (2017). Lateral recumbency made it easier to get to the teat canal and cistern during surgery. It also made it easier to use the theloscopic tools. Sedation was used on animals that would not cooperate. Before surgery, ring block was used to ease pain in the area. In most animals, local anaesthesia with ring block was enough to get the theloscopic procedure done without any problems. Ring block analgesia was used by Gopalbhai (2010) to do theloscopy on farm animals. Other writers used the axial method and injected 2% lignocaine into the teat canal. For the lateral method, ring blocks were mostly used (Bleul et al., 2005; Raj, 2010). During axial theloscopy, it was easier to put the theloscope with metal bands through the teat canal when the teat was swollen. This was done by putting a rubber ring at the base of the teat and continuously flushing it with normal saline. A soft rubber band attached to the bottom of the teat helped to stretch the teat and stopped milk from getting into the teat tank and blocking the view (Gopalbhai, 2010). To make the teat bigger, air was sucked into it through the theloscope (Geishauser & Querengasser, 2001; Raj, 2010; Bleul et al., 2005; Gopalbhai, 2010; Walvekar et al., 2017). Insufflation at level 2 of the machine settings was enough to open up the teat so that the inside structure of the teat tube and teat cistern could be studied for diagnostic purposes. But this treatment wasn't needed when the teat wasn't inflamed. During this study, it was already known that putting the metal sleeves into the teat cistern through the tip of the streak canal hole caused some minor damage. To avoid this harm, the teat streak canal and cuffs with a diameter of less than 3 mm OD might be helpful. It was the same as what Tulleners & Hamir (1990), Medl & Querengasser (1994), Geishauser & Querengasser (2001), Bleul et al. (2005), and Geishauser et al. (2005) found. During teat theloscopy, the teat cistern with spiral folds, Rosette of Furstenberg, and longitudinal blood vessels could be seen through the teat canal. This was similar to what Querengasser et al. found in 2002.Theloscopy through the teat canal was done on 15 of the women in this study. The laparoscopy made the teat cistern and teat channel easy to see. Similar studies were published by Rathod et al. (2009). Theloscopy showed the exact state of the mucosa, its severity, and the length of time it was abnormal. In contrast to what Jhala (2009) said earlier, the teat contractions and relaxations could not be seen when the teat canal was examined endoscopically.

L. Theloscopic Findings

In group I, Theloscopy was not performed in the cases of teat fistula and teat laceration,as the fistulous openings were large and the internal teat structures were already exposed therefore theloscopy was not required. Theloscopy revealed inflammed and hyperaemic teat cistern with injured tissue of the teat. This inflammation and hyperaemia could be due to the traumatic injury to the teat as also stated by Singh (2018).

In group IIa,theloscopy procedure was not performed in animals suffering with Supernumerary teats (group IIa1) and Imperforate teats (group IIa2) due to absence of opening of the teat canal. Teat spider was revealed in 1 case of obstruction at the base of the teat (group IIa3) obstructing the milk flow in the affected teat. Ultrasonography alone did not confirm teat spider, which was observed as a hyper echoic band impeding milk flow at the base of the teat. Jhala (2009) also documented the presence of a teat spider at the base of the teat as a cause of milk flow obstruction from the affected teat. Similar membranous obstruction completely obstructing the milk flow through the obstructed teat were recorded by Brightwell (1969).

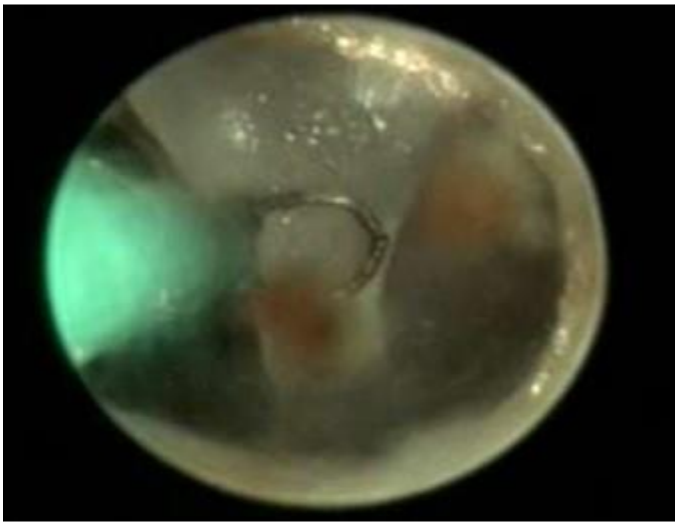

Fig.14: Theloscopy Revealed Presence of Teat Spider

Fig. 15: Theloscopy Revealed Presence of Proliferation at the area of rosette of Furstenberg in case of obstruction at tip of the teat

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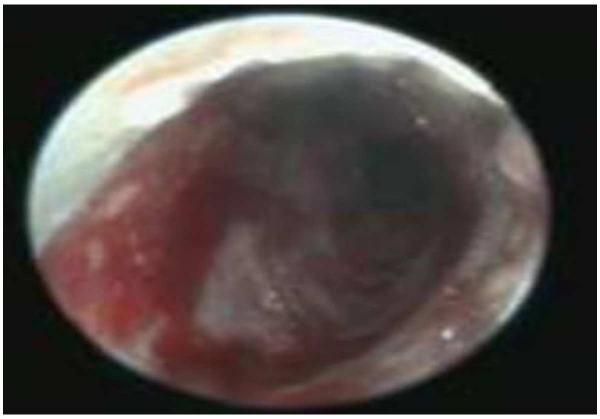

Fig. 16: Theloscopy revealed hyperaemic areas with teat tip laceration in case of obstruction at tip of the teat

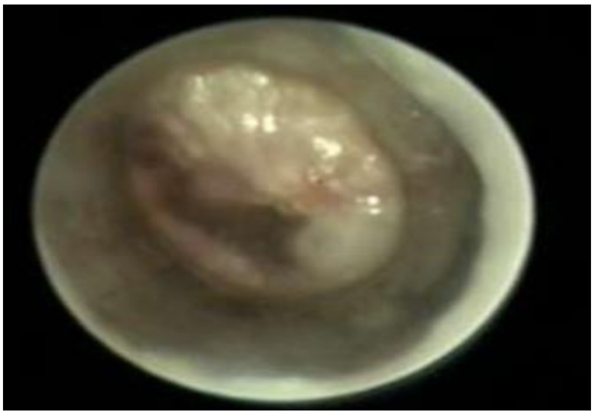

Fig. 17: Theloscopy Revealed Presence of Obstruction in the Middle of the Teat Cistern in Case of the Mid Teat Obstruction

M. Treatment

The present study includes 5 cases of teat fistulae (group Ia) and 15 cases of teat laceration (group Ib); in these cases, surgical intervention was performed as soon as possible, following ring block analgesia with 2% Lignocaine HCl at the teat's base. These cases were repaired with 3-0 Polyglactin-910 (Vicryl, Ethicon Inc.) using three-layer technique as described by Anant(2016). In delayed cases (n=3) i.e. the cases which were already repaired at field level there was recurrence of teat fistula within 3-10 days of the first attempt. No complication was recorded in fresh cases (n=5) and all the animals showed uneventful recovery upto 1 month of follow up. In cases of reoccurrence of teat fistulae, resuturing was done using 3-0 Polydiaxonone (Vicryl, Ethicon Inc.) in 2 layers during the second attempt after debriding and freshening the wound edges. All the 3 cases recovered uneventfully following the second surgery. Kadu(2013) successfully repaired four cases of teat fistula with 3-0 catgut using 2-layertechnique after sedating the animal with xylazine @ 0.05mg/kg B.Wt. in addition to ring block analgesia. Anant (2016) used 2-0 Polyglactin-910 (Vicryl) for repairing teat fistula in three-layer technique. In the present study, Polyglacitin-910 was found suitable for repair of teat fistulae in fresh cases. However, in delayed cases, Polydiaxonone was found ideal for preventing recurrence. This may be due to monofilament nature of Polydiaxonone which prevented infection due to capillary action (Chu &Williams,1984). In cases of teat tip injury (group Ic), debridement of the teat tip was done. As the teat canal was patent so suturing of the outer teat skin was done and a teat plug was placed in the teat canal. Owner was advised to keep the teat plug until suture removal. Suitable intra mammary preparation and anti-inflammatory drugs were used to minimise the inflammatory lesions. Telephonic follow-up at 2-3 months revealed that milking was normal. In animals presented with supernumerary teats, the supernumerary teats (group IIa1), were excised surgically prior to start of the lactation. The surgical wound was closed routinely. Antibiotics and analgesics were administered as described earlier. All animals recovered uneventfully. Similar treatment procedure of supernumerary teat excision were followed by Mulon (2016); Kadu(2013). According to Tiwary et al. (2005), the presence of super numerary teat had no influence on milk output, lactation length, age at calving, or conception rate. In cases of imperforate teat(group IIa2), teat dilator was used to relax the sphincter muscles and Hug's knife to widen the teat orifice . A teat siphon was then inserted into the proposed site of teat canal to enlarge the opening and drain the milk. A sterile infant feeding tube was then placed into the teat canal created surgically and fixed as described earlier. Suitable antibiotics and analgesics were administered as described earlier. Telephonic follow up revealed the recovery of the teat. However, Kadu (2013) treated the condition by introduction of teat knife in the teat canal to regain the patency of the teat canal. O'Connor (2001) directly inserted the teat siphon to obtain the desired effect of regaining the patency.

N. Based on the Results of the Present Study Following Conclusions Were Drawn.

- Cows are more prone to milk flow disorders as compared to buffaloes. Partial obstruction of milk flow was more common as compared to complete obstruction.
- Physical trauma followed by growth/fibrosis was major causative factor for milk flow disorder in animals and hind quarters are more commonly affected than fore quarters.
- Ultrasonography of the normal and diseased teats was done using direct contact technique and water bath technique with 6-12 MHz linear transducer in standing animals and was reliable for diagnosis of extent and location of the lesion in cases of teat obstructions. Ultrasonography by water bath technique was found to be of greater value as compared to the direct contact technique.
- Theloscopy is reliable for diagnosis of the internal lesions in cases of teat canal obstructions and to detect the presence of foreign body but was not applicable in cases of full teat fibrosis, Imperforated teat and Supernumerary teat, open teat injuries.
- Theloscopic electro resection was a minimally invasive technique for management of covered teat lesions. It aided in restoring the functional capacity and anatomical structure of the teat with milk flow disorders. It allowed safe intervention for covered teat lesions with comparatively less inconvenient and risk to the animal as compared to the conventional methods for diagnosis and treatment of milk flow disorders.

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