**Abstract**

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| A case study was conducted to evaluate the treatment strategy of gangrenous mastitis in a cross breed doe at S. A. Quadery Teaching Veterinary Hospital of Chittagong Veterinary and Animal Sciences University, Chittagong. Confirmatory diagnosis was performed from the obvious physical changes of the udder. The underlying causal microorganisms (*Staphylococcus sp* and *E. coli*) were isolated from milk sample. Parenteral (Gentamycin + Sulphadimidine+ Trimethoprim intramuscularly) and intramammary infusion of antimicrobials (Streptomycin + Penicillin) along with supportive treatments for 7 days could not save the quarter rather condition deteriorated. After checking antibiogram only ciprofloxacin was found sensitive. Decision was taken for mastectomy to save the animal. Ciprofloxacin was used as antibiotic after mastectomy. The doe recovered without other complications. |

**Key words:** goat, gangrenous mastitis, etiology, diagnosis, treatment.

**CHAPTER-I**

**Introduction**

The term mastitis comes from the Greek words *mastos* (“breast”) and *itis* (“inflammation of”), refers to an inflammation of the parenchyma of the mammary gland, regardless of the cause **(Spanue, 2009).** Mastitis is characterized by a range of physical and chemical changes in the milk and pathological changes in the glandular tissue **(Radostits *et al*., 2007).** However, cases of mastitis can be divided into clinical and subclinical categories **(White, 2007).** Mastitis is considered one of the most important diseases of domestic animals, caused by several etiologic agents **(Anderson et al., 2005).** Inflammation of the mammary gland/udder may theoretically be caused by trauma of various kinds but far the most common causes are the infectious agents. In severe cases of mastitis caused by virulent strains of organisms, thrombosis of the mammary vessels occur resulting in infarction and gangrene. It frequently causes partial or complete damage to clinically affected glands leading to reduce milk yield and retarded growth of lambs. Additional losses associated with gangrenous mastitis are cost of treatment and culling of does due to permanent damage to udder. Gangrenous mastitis is one of the most difficult forms of mastitis to be managed **(Bloway, 1993).** In very severe cases, gangrene leads to toxemia and animal may die **(Ribeiro et al., 2007).**

Mastitis reflects the inflammation of the mammary gland, which may occur due to any bacterial infection secondary to teat injury or poor management **(Marogna *et al*., 2012; Egwu *et al*., 2001).** *Staphylococcus aureus* is recognized as the most common causal agent of goat mastitis, followed by minor occurrence of *Mannheimia* (Pasteurella) *haemolytica*, *Escherichia coli*, *Clostridium perfringens*, *Streptococcus*, *Pseudomonas* and *Nocardia* genera **(Radostits et al., 2007).** *Staphylococcus aureus* in goat mammary gland system, can produce any type of mastitis like subclinical, chronic and acute. Increase pathogenic potential of *S. aureus* produces disease in more severe form i.e. gangrenous mastitis **(White and Hinckley., 1999).** High morbidity and mortality was observed along with 4% to 40% frequency of *S. aureus* among all isolated microorganisms recorded **(Marogna *et al*., 2012).** Besides bacteria, mycoplasmal and yeast may cause mastitis **(C.Boscos., 1996).** Several risk factors, milking hygiene, management practice, feeding, number of days not lactating, number of lactation days and geographical locality may influence the type and the frequency of isolation of organisms causing mastitis in animals **(Smith & Hagstad., 1986).** The females of all animal species especially cows and goats during steaming up and in the first few weeks after parturition are vulnerable to mastitis **(Heidrich and Renk., 1968).** Mastitis causing bacteria produce very different rates of clinical mastitis. *Coliform* and environmental *Streptoccocus* infections tend to be associated with relatively high incidence rates of clinical mastitis **(Ingalls., 2003).** The organisms infecting the udder of goats are similar to those in cows. Coagulase-negative staphylococci are generally the most prevalent and can cause persistent infections that result in increased cell counts and low-grade mastitis with some recurring clinical episodes. The level of infection and incidence of gangrenous mastitis due to *Staphylococcus aureus* tends to be low (<5%) but can result in persistent infections that do not generally respond to therapy **(Merck Veterinary Manual).**

Previously there were many goats registered at SAQTVH affected with gangrenous mastitis. But the causal agent and antibiotic sensitivity had not been screened. Therefore I choose this case to prepare my clinical report with the following objectives :

So the objectives of this study were :-

1. To identify the causal agents.
2. To detect the efficacy of antibiotics used for treatment.
3. To know the overall management of a gangrenous mastitis case.

**CHAPTER-II**

**Materials and Methods**

**Case history**

A 3.5 years of cross breed (Black Bengal × Jamuna Pari) lactating doe was admitted to the S.A Quadery Teaching Veterinary Hospital (SAQTVH) of Chittagong Veterinary and Animal Sciences University, Chittagong with a history of two weeks of lactation, sudden anorexia and reduced milk production.

**Clinical Examination**

Clinical examination revealed that heart rate was 80/minute and respiratory rate was 28/minute. Mucous membranes were slightly pale. Rectal temperature was 1040F. Large swelling of mammary gland at the right quarter. Changing the milk color, reddish milk came out through teat canal. Pain on palpation was found. Affected quarter was indurated. Reddish to Black discoloration of the affected quarter with sharp demarcations from grossly normal tissue were noticed **(Fig: 5.1.A)**

**CHAPTER-III**

**Diagnosis**

From clinical signs and symptoms the disease was diagnosed as gangrenous mastitis. Blood sample was collected to see the biochemical profile of the doe. For gram’s staining, isolation of causal agents and testing antibiotic sensitivity, milk sample was collected.

For biochemical test (Ca & P) blood sample was collected aseptically. Detection of Ca level was very sensitive. After measuring the sample absorbance (Asample) according to the assay procedure, one drop of EDTA was added to the samples and turned colorless (after about 10 seconds). The absorbance of the sample was read. Blank against the reagent blank (reagent blank) was set to obtain Asample/EDTA. Mix, absorbance of the sample (Asample) and standard (Astandard) against the reagent blank after 5 to 50 minutes. **(Randox Laboratories Limited).**

**Table- 3. 1**

**Detection of Ca level:**

|  |  |
| --- | --- |
| Wavelength | Hg 578 nm (550- 590) |
| Spectrophotometer | 570 nm |
| Cuvette | 1 cm light path |
| Measurement | against reagent blank  only one blank required per series |
| Temperature | 20-25 0C/ 37 0C |

For detection of P level wavelength was 340 nm, Hg 334 nm and Hg 365 nm. Optical path was 1 cm, temperature maintained 20-25 0C / 37 0C and measurement against reagent blank. Mixed sample, DW and reagents were incubated for 5 min. at 20-25 0C / 37 0C. After 60 minutes later the absorbance against the reagent blank was read. **(Diasis Diagnostik Sistemler Tic. Istanbul, Turkey).**

Milk samples were aseptically collected and submitted to microbiological culture on Mannitol salt media, and MacConkey agar, incubated at 37 0C for 08 hours. Then formation of colonies in culture petridish **(Fig: 5.3).** Microorganisms isolated were identified based on macro and microscopical morphology by Gram stain, culture characteristics and biochemical profile **(Krieg and Holt, 1984; Quinn et al., 1994).**  Culture of milk stained by Gram’s staining revealed combined infection characterized by numerous Gram–positive cocci of *Staphylococcus sp* **(Fig: 5.4.A)** and Gram–negative rods *E.coli***. (Fig: 5.4.B)**

Antimicrobial sensitivity test was performed by Kirby-Bauer Technique **(Fig: 5.5).** A thin uniform inoculum of the test strain is exposed to a disk **(Oxoid limited, United Kingdom)** of known concentration of antimicrobial agent. The antimicrobial agent from the disk gradually diffuses into the agar and creates a concentration gradient of the drug. The susceptibility of the organism to the agent is indicated by a clear zone of inhibition around the disk. The diameter of the zone of inhibition is directly proportional to the susceptibility of the organism tested. Absence of a zone of inhibition around the drug reservoir indicates complete resistance **(Bauer et al., 1966)**

**CHAPTER-IV**

**Treatments**

Combined preparation of Gentamycin, Sulfadimidine and Trimethoprim (Inj. Gentasone plus, 10 ml and 100 ml vial) [Chemist Laboratories Ltd., Bangladesh] @ 2 ml were administered intramuscularly daily for 7 days.As Intramammary infusion Penicillin and Streptomycin combination (Inj. SP-Vet® 0.5 gm vial) [Acme Laboratories Ltd., Bangladesh] @ 2 ml were administered for 7 days. As a supportive treatment Inj. Aminovit Plus (Preparation of Vitamin, mineral and amino acid, Vial-50 ml, Company-Popular, Bangladesh) @ 2 ml was administered intramuscularly, Dextrose 5% (Inj.Dextrose-500 ml) [Popular Pharmaceuticals Ltd., Bangladesh] @ 300ml and Calcium –Magnesium combined preparation (Inj. Sancal vet oral, 1L pack) [Novartis Pharmaceuticals Ltd., Bangladesh] @ 10 ml twice daily were administered intravenously slowly for 7 days. But there was found no effective result. So decision was taken to mastectomy.

**Mastectomy:**

Doe was controlled by left lateral recumbency with high epidural (last sacral and first coccygeal vertebrae) anaesthesia with 2% solution of Lidocaine HCl @ 4 ml (Inj. Jasocaine® 2%, 50 ml vial) [ Jayson Pharmaceuticals Ltd., Bangladesh]. Ring block was done locally with 2% Lidocaine @ 20ml. Antiseptic washing and incision at the border of the affected right quarter was performed. Removal of the affected quarter and skin suture with silk. **(Fig:5.2.B)**

**Post operative cares**:

As post operative care Ciprofloxacin (Inj.Cipo-A vet, 30 ml vial) [Acme Laboratories Ltd., Bangladesh] @ 2 ml, Ketoprofen (Inj.Keto vet, 10 ml vial) [Techno Laboratories Ltd., Bangladesh] @ 0.5ml and Pheneramine meleate (Inj.Astavet, 10 ml vial) [Acme Laboratories Ltd., Bangladesh] @ 1 ml were administered intramuscularly for seven days.

**CHAPTER-V**

**Results of Laboratory Tests**

Biochemical examination of blood showed that serum calcium level was 8.85 mg/dl and Serum phosphorous level was 7.95 mg/dl **(Table- 5.1)**. Bacteriological examination was done. Microscopic examination revealed positive result with gram stain. Isolation and identification of the organism were done from milk sample grown on Muller Hinton, MacConky and Mannitol salt agar media. Finally *E.coli* and *Staphylococcus* were suspected based on colony morphology and microscopic examination. *Staphylococcus sp* formed a fairly large yellow colony **(Fig: 5.3.A)** and *E.coli* with large pink colony characteristic **(Fig: 5.3.B)**. Under microscope *Staphylococcus sp* was cocci in grape like cluster **(Fig: 5.4.B)** and *E.coli* was just like bacilli **(Fig: 5.4.A).** In antimicrobials sensitivity test only Ciprofloxacin was strongly sensitive. Gentamycin and oxytetracyclin were moderately sensitive **(Table -5.2).**

**Table- 5.1**

Blood examination report:

|  |  |  |
| --- | --- | --- |
| Name of the test | Result | Normal Range |
| Serum Calcium | 8.85 mg/dl | 9.7-12.4 mg/dl |
| Serum Phosphorous | 7.95 mg/dl | 4.2-9.1 mg/dl |

(Goatlink.com, 2009)

**Table- 5.2**

Antimicrobials sensitivity test

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Drug | Strongly sensitive | Moderately Sensitive | Weakly sensitive | Resistant |
| Penicillin |  |  |  | + |
| Ampicillin |  |  |  | + |
| Oxytetracycline |  | + |  |  |
| Gentamycin |  | + |  |  |
| Neomycin |  |  |  | + |
| Cephalexin |  |  |  | + |
| Cotrimazole |  |  |  | + |
| Amoxicillin |  |  |  | + |
| Ciprofloxacin | + + + |  |  |  |
| Colistine sulphate |  |  |  | + |

(Department of Microbiology, CVASU)

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**Figure-5.1: (**A)Changes in udder in the early stage of gangrenous mastitis.Black colored arrow indicates **r**eddish to Black coloration of skin. (B) Red color milk from the affected quarter (blue colored arrow).

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**Figure-5.2: (A)** Before surgery of udder. Black arrow indicates sloughing of tissue leaving an open wound. (B) After surgery (Mastectomy) of right quarter. Blue arrow indicates the skin suture.

|  |  |
| --- | --- |
| **A** | C:\Users\SAMUN\AppData\Local\Microsoft\Windows\Temporary Internet Files\Content.Word\IMG_5792.jpg  **B** |

**Figure-5.3**: (A) Colonies (Large yellow colonies) of *Staphylococcus sp* in Mannitol salt agar media. (B) Colonies (Large pink colonies) of *E.coli* in MacConky agar media.

|  |  |
| --- | --- |
| C:\Users\SAMUN\AppData\Local\Microsoft\Windows\Temporary Internet Files\Content.Word\E_coli_2000_P7201172.jpg  **A** | **B** |

**Figure- 5.4:** (A) Microscopic slide of *E.coli*(Like red color bacilli)

(B)Microscopic slide of *Staphylococcus sp (Cocci in grape like cluster).*

|  |
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| **C:\Users\SAMUN\Pictures\ffff.PNG**  **Figure-5.5:** Antimicrobial Sensitivity Test. Black arrow indicates Ciprofloxacin was strongly sensitive than Penicillin, Ampicillin, Oxytetracycline, Gentamycin, Neomycin, Cephalexin, Cotrimazole, Amoxicillin and Colistine sulphate. |

**CHAPTER-VI**

**Discussion**

Cases of gangrenous mastitis are less reported in goats. **Chauhan., (1997)** reported that it occurs more frequently in ewes, which suffer a severe unilateral attack of staphylococcal mastitis. There was no recurrence of the growth after few days of excision and the animal recoverd normally. *Staphylococcus aureus* can cause subclinical IMI, but also acute clinical mastitis or gangrenous mastitis **(Contreras et al., 2003).** The identification of different causal agents associated to mammary infections in small ruminants has been reported **(Radostits et al., 2007).** Combined infection by *S. aureus, C. perfringens* and *E. coli* in gangrenous mastitis in goats was reported by the first time in Brazil **(Ribeiro et al., 2007).** Gangrenous mastitis is a per acute form of mastitis, characterized by necrosis of the udder tissue, caused by alpha-toxins **(Smith & Sherman.,** **2009).** Gangrenous mastitis in goats is a severe clinical condition of the inflammatory process in mammary glands. Clinical signs commonly occur in the first weeks of lactation, committing one or two sides of the glands and are characterized by fever, anorexia, dyspnea and systemic signs of toxemia. Initially, the udder is hot, painful and occurs swelling of the affected side, with milk watery, containing flocculent pus and/or blood secretion. Evolution of the process is characterized by udder becoming discolored (blue–blackish or blue–greenish), cold, with demarcation line of the affected tissue, development of abscess and draining pus **(Ribeiro et al., 2007)** which are nearly similar to our observed sign. I had found serum Ca and P level was accordingly 8.85 mg/dl and 7.95 mg/dl from biochemical test where as normal range was accordingly 9.7-12.4 mg/dl and 4.2-9.1 mg/dl **(Goatlink.com, 2009).** Treatment with **antibiotics** (usually a combination of two or more antibiotics) is necessary. Antibiotics alone are not effective because they do not enter infected muscles sufficiently **(**[**Patel**](http://www.mdhil.com/author/mdhil_pass#-2-2-2-2-2-2-2-2/)**., 2012).** We used combined preparation of Gentamycin, Sulfadimidine and Trimethoprim (Inj. Gentasone plus) @ 2 ml intramuscularly daily for 7 days andas intramammary infusion Peniciline and Streptomycin combination (Inj. SP-Vet® 2.5 gm vial) @ 2 ml were administered for 7 days. As a supportive treatment Inj. Aminovit Plus @ 2 ml was administered intramuscularly, Dextrose 5% (Inj.Dextrose-500 ml) @ 300ml and Calcium –Magnesium combined preparation (Inj. Sancal vet oral, @ 10 ml twice daily were administered intravenously slowly for 7 days. But treatment failed. So we had to performed surgery. Besides antibiotic sensitivity test had done and found Ciprofloxacin was strongly sensitive. Antibiotic therapy would not give good result due to late stage of mastitis. Surgical removal of the dead and dying tissue is the immediate treatment. Sometimes infected rehabilitation therapy is used **(**[**Patel**](http://www.mdhil.com/author/mdhil_pass#-2-2-2-2-2-2-2-2/)**., 2012).** Treatment of the early and intermediate stages of the disease was successful through the administration of systemic and intramammary terramycin together with [diuretics](http://europepmc.org/abstract/MED/3402223/?whatizit_url_Chemicals=http://www.ebi.ac.uk/chebi/searchId.do?chebiId=CHEBI%3A35498) and topical antiseptic cream. The late stage of the disease was successfully treated only through surgery **(**[**Abu-Samra.,**](http://europepmc.org/search;jsessionid=whdBinF6tchzkmgXNYnQ.0?page=1&query=AUTH:%22Abu-Samra+MT%22) **1988).** We got better outcome by treated with surgery- Mastectomy. Surgicaltreatment of the gangrenous mastitis animal **(Yeshwantkumar and Nirmala., 2008)** restrained and local infiltration anesthesia was done using 2% Lidocaine HCl. Post-operatively combination of Peniciline and Streptomycin (Inj. SP Vet, 2.5 gm pack) [ Acme Laboratories Ltd., Bangladesh] @ 2 ml was given IM once a day for 5 days and the wound was dressed with Povidone iodine ointment. Goat recovered and had a normal a normal parturition **(C.Yeshwantkumar and G.C .Nirmala., 2008).**

**CHAPTER-VII**

**Conclusion**

The prognosis of gangrenous mastitis in goat is not good as septicemia develops. Success depends on early treatment with appropriate antibiotics and proper supportive therapy. Surgery can save the animal but milk production might be lost partially or completely.

**CHAPTER-VIII**

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**CHAPTER-IX**

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