Comparative Studies on Unsutured and Sutured Urethrotomy Wounds by Different Suturing Materials in Male Buffalo Calves (Bubalus bubalis)

THESIS

SUBMITTED TO THE

RAJENDRA AGRICULTURAL UNIVERSITY

PUSA, (SAMASTIPUR) BIHAR

(FACULTY OF VETERINARY SCIENCE AND ANIMAL HUSBANDRY)

In partial fulfilment of the requirements

FOR THE DEGREE OF

Master of Veterinary Science

(SURGERY & RADIOLOGY)

By

Sanjeev Kumar

(Registration No. - M/Vety. Surg./20/1998-99)

Department of Surgery and Radiology

BIHAR VETERINARY COLLEGE

PATNA - 800 014

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Dedicated

To

My Maternal Grand Mother

Dr. K. B. P. Agrawal
M. V. Sc., Ph. D.
Ex-Assoc. Prof. & Head,
Deptt. of Surgery and Radiology,
Bihar Veterinary College,
Patna – 14. (India)

CERTIFICATE – I

This is to certify that the thesis entitled ***"Comparative studies on unsutured and sutured Urethrotomy wounds by different suturing materials in male buffalo calves (Bubalus bubalis)"*** submitted in partial fulfilment of the requirement for the Degree of **Master of Veterinary Science** (Surgery & Radiology) of the Facultry of Post-Graduate Studies, Rajendra Agricultural University, Bihar is the record of bonafide research carried out by **Dr. Sanjeev Kumar** under my supervision and guidance. No part of the thesis has been submitted for any other Degree and Diploma.

It is further certified that such help or informations received during the course of this investigation and preparation of the thesis have been duly acknowledged.

30/12/2000

(Dr. K. B. P. Agrawal)
Major Advisor

Endorsed

30/12/00

(Chairman / Head of the Department)

CERTIFICATE - II

We, the undersigned, Member of the Advisory Committee of **Dr. Sanjeev Kumar**, a candidate for the Degree of **Master of Veterinary Science** with Major in Surgery & Radiology have gone through the manuscript of the thesis and agree that the thesis entitled ***"Comparative studies on unsutured and sutured Urethrotomy wounds by different suturing materials in male buffalo calves (Bubalus bubalis)"*** submitted by **Dr. Sanjeev Kumar** in partial fulfilment of the requirements for the Degree.

Agrawal 30/12/2000

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2. **Dr. M. K. Roy,** 30/12/2000
3. **Dr. C. Jayachandran** 30/12/2000
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CERTIFICATE - III

This is to certify that the thesis entitled ***"Comparative studies on unsutured and sutured Urethrotomy wounds by different suturing materials in male buffalo calves (Bubalus bubalis)"*** submitted by **Dr. Sanjeev Kumar** in partial fulfilment of the requirements for the Degree of **Master of Veterinary Science** (Surgery & Radiology) of the Faculty of Post-graduate Studies, Rajendra Agricultural University, Bihar was examined and approved on 26.6.2002

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Place : Patna

Date : 30/12/2000

Sanjeev Kumar.

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INTRODUCTION

INTRODUCTION

The history of scientific progress is linked with progressive specialisation. The quest for knowledge has enabled to enter into newer domains of veterinary surgery and as a result during the recent past surgical manoeuvers have advanced greatly in all branches of the sciences with very encouraging results. Such endeavors could be possible due to day by day increasing the value of animals, economic viability, awareness of people regarding safe surgery and quick recovery.

In India inspite of mechanised farming, bullock still form the backbone of agricultural operations due to small holding of lands and other relevant causes. For farmers in India bullocks are equally important as dairy cow. In bullocks one of the very common surgical malady is obstructive urolithiasis resulting into heavy mortality. The occurance of urolithiasis in animals during the prime working seasons create great economic problem for the farmers (Anjaria, 1969).

Urolithiasis is more common in bullocks but buffaloes are also affected (Kataria and Rao, 1969; Bhatt *et al.*, 1973 and Gera and Nigam, 1979) owing to predisposing factors like early castration,which leads to decrease in the testosterone level. This inturn decreases the hydrophilic colloids in the urine, which predisposes the animal to obstructive urolithiasis (Williams, 1955; Marsh and Safford, 1957 and Kopper, 1967).

In cattle, the site for lodgment of calculi might be either in the kidney, ureter, bladders or urethra. Urethral obstruction is more common in bullocks than any other type of urinary obstruction. This might be due to the fact that the calculus in cattle is of migratory type and usually gets arrested in distal part of sigmoid flexure during its passage. Later on, with the concentric

deposition of salts over the calculus the lumen is blocked, which results in retention of urine that is characterised by anuria, rupture of urinary bladder and heavy mortality due to uremia (Prasad *et al.*, 1978). In 37.5 percent cases of obstructive urolithiasis was leads to rupture of urinary bladder and mortality rate as high as 50 percent inspite of medicinal treatment (Donecker and Bellamy, 1982 and Prasad *et al.*, 1978).

Pressure necrosis and perforation of urethral wall are common complications following urethral obstruction. These usually result in subcutaneous accumulation of urine on the ventral abdominal wall around the sheath and umbilicus. In advance stage, complete necrosis and sloughing of the skin have also been seen. Rupture of urethra in cattle may occur due to direct trauma or irregular sharp edged calculus. It may also be encountered due to impaction of urethra with sabulous material and gravel (Shastry and Rao, 1981).

It has been found in the literatures that treatment for urinary calculi has been tried medicinally and surgically. Various workers in this field have expressed their opinion that therapeutic management has some value in early phase of the disease and is directed mainly towards its preventions rather than cure, but in well established state of the disease, surgical approach has become more practical. So the established surgical management of obstructive urolithiasis is urethrotomy. After performing urethrotomy, urethra is intubated by polythene catheter to provide free flow of urine.

Post surgical management of urethrotomy wound is still controversial. There is difference of opinion, whether to suture the urethra or not. Some workers sutured the urethral incision while others left it to heal of its own (Oehme and Tillaman, 1965; Anjaria, 1969; Prasad *et al.*, 1978; Kolhekar

1960 Gera and Nigham 1979; Singh *et al.*, 1978 and Sharma *et al.*, 1983). The most common complications of unsutured urethra with indwelling catheter are seepage of urine and formation of urinary fistula at the site of operation, particularly when the indwelling catheter is not snugly fitted. The subsequent infiltration of urine at urethrotomy site and abdominal area leads to cellulitis, local tissue necrosis and wound dehiscence. Unsutured urethrotomy wounds preclude the risk of stricture formation and allow the mucosa of urethra to reestablish its continuity and heal by second intention (Bozrab *et al.*, 1983).

In operative surgery, incised wound edges are closed by suture to obtain healing by first intention. As suture hold the wound edges together, the process of healing continues, meanwhile the reunion of anatomical layers take place. Thus, wound regain ability to withstand stress and strain on its own. Healing of the wound is very much affected by type, size, nature of suture material and pattern of suturing. Urethral wound is closed by two layers of continuous suture pattern to minimise the chance of infiltration of urine at the site of operation (Gera *et al.* 1973 and Gera and Nigam 1979), where as Vig and Tyagi (1970) sutured the urethral wound in three layer.

Silk is inexpensive, readily available and easily sterilized by autoclaving. Silk is handled comfortably, holds a knot well, maintains adequate strength and causes moderate tissue reaction. The utility of catgut has been demonstrated by many workers in urological surgery of large animal and men, either experimentally or clinically. (McLean and Fais, 1952; Goldstein and Gualteiri 1967; Vig and Tyagi 1970; Sharma and Khan 1978; Larson 1997 and Prakash *et al.* 1999).

Polyglactin – 910 is synthetic braided suture material consisting of glycolic acid and lactic acid in the ratio of 9:1. It is absorbed by hydrolysis and absorption is slower than chromic catgut. It elicits minimum tissue reaction and does not predispose infection and calogenic potency (Stewart *et al.* 1990). It has surgical potency similar to silk and non-toxic upon implantation into animals (Herrmann *et al.* 1970).

It is generally feared that suturing of urethra may lead to stricture formation, that enhance the chance of further lodgement of calculi. Some reports regarding urethral wound closure are available in bovine, but without any recommendation (Chatuphale, 1932; Barshikar, 1932-33; Vig and Tyagi, 1970; Gera and Nigam, 1979; Larson, 1996 and Sharma and Agrawal 1997). Keeping in view the above mentioned facts the present investigation was under taken with the following specific aims and objectives.

1) To compare the healing of sutured urethrotomy wound with that of unsutured.
2) To study the clinical changes during wound healing of urethra.
3) To study the haematological changes during urethrotomy wound healing.
4) To study the histopathological changes of unstured urethrotomy wound and different type of suturing materials undertaken for closure of urethrotomy wound.
5) To compare the efficacy of three suture materials used for closure of urethrotomy wound.

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Chapter - 2

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Urethrotomy is usually performed for the management of obstructive urolithiasis. For surgical management of urethrotomy wound, various techniques have been adopted by various workers. Some sutured the urethral wound edge and some left it to heal by second intention. In case of suturing of urethra various technique were also applied. Hence, in this study an attempt has been made to review all available relevant literature under the following heads.

A) Urethrotomy and urethral wound management .

B) Haemotological and biochemical changes.

A) urethrotomy and urethral wound management

Fleming (1902) outlined the ischial and post-scrotal uresthrotomy in male bovine. He advocated ischial urehrotomy when the bladder is in danger of rupture or when the calculi could not be located and advised for pre or post-scrotal urethrotomy to extract out the calculi. He neither mentioned about the suturing of the urethra nor sutured the skin incision and left the wound to heal spontaneously.

Wooldridge (1924) practised ischial urethrotomy and used a catheter through the urethrotomy opening to evacuate the bladders. He stated that post-scrotal urethrotomy was less successful.

Chatuphale (1932) and Barshikar (1932-33) after performing urethrotomy in a bullock sutured the urethral edges with catgut. Infiltration of urine was observed at the site of operation.

Carey (1940) performed ischial urethrotomy and exteriorization of the urethra after amputation of the penis.

Sambamurthi (1947) observed a case of urethral calculi with rupture of the urethra. He preferred ischial urethrotomy for easy drainage of urine and overcoming urine infiltration with subsequent complication.

O'conner (1950) suggested suturing of urethra in order to prevent urine infiltration as subsequent complication.

Guard (1953) emphasized that the length of the incision should be shortened gradually from the skin wound to that of urethral wound for better facility in drainage.

Puntriane (1954) viewed that by mere incising the urethra for the removal of stone in not completely successful., becomes animals may not fully recover and may show recurrences.

In order to minimize the possibility of stricture formation, it has been advocated that the longitudinal urethral incision be sutured transversely (Motta and Walmsley 1954).

Gibbons (1956) justified the surgical removal of urethral calculus in valuable breeding bulls and reported that incision in the urethra heals better as an open wound.

Kingrey (1956) discussed about the repair of urinary bladder. He stated that a routine urethrotomy of these animals are usually attended by poor prognosis. Spontaneous healing of the bladder may be an accidental feature. The defect of the bladder closed surgically gave encouraging results. The initial procedure was paracentesis to remove the accumulated urine from the abdominal cavity accompanied with ischial urethrotomy to facilitate urinary flow and finally repaired the bladder through ventral abdominal approach in prepubic region just lateral to penis under 2 to 4 % procaine

hydrochloride infiltration at the proposed line of incisions. The operation was relatively simple.

Verraya (1957) described a case of urethral calculus with rupture of bladder in a bull. He performed urethrotomy and saved the life of the animal and stated that the opened bladder was closed due to Nature's miracle.

Symthe (1959) stated that urethrotomy for removal of urinary calculus is only confined to the relief of the distended bladder.

Kolhekar (1960) performed post scrotal urethrotomy to remove urethral calculi located in the 'S' curve of urethra in bullock. The urethral wound was closed with fine nylon suture in lambert pattern. Skin was sutured separately with nylon. Animal was uneventfully recovered.

Lowe (1960) surgically removed the equine uroliths by superpubic cystotomy. He stated that the conventional method of sub-ischial urethrotomy for the removal of cystic calculi through the urethra has several disadvantages viz, the surgeon operates only by tactile sense, fragments of calculus are often free in the bladder or incorporated in the mucosa of the bladder which may act as nuclei for the formation of more calculi in future.

Scheel *et al.* (1960) sated that urethrotomy was performed in case of distended urinary bladder with urethral occlusion in steers, although there was considerable inflammatory changes in the urethra and even after surgery urination was often difficult. They further stated that urethrotomy does not constitute completion of treatment. Even then animals may show the sign of stranguria.

Carey (1961) followed sub-ischial urethrotomy when there was complete stoppage of urine and the bladder was in danger of rupture. He sutured the urethral leap to the margins of the wound on each side and

allowed the wound to heal spontaneously and to form a permanent urinary fistula.

Vasko *et al.* (1962) experimentally produced vesicle calculi in steer calves and then performed suprapubic cystotomy. In all such cases bladder healed up without infection.

Khan (1963) reported the treatment of 12 clinical cases of urethral calculi. Out of twelve cases one was a heifer, one was a male buffalo calf and others were male cow calves. He adopted different surgical techniques viz., post – scrotal urethrotomy, ischial urethrotomy with polythene tube, manual removal of the calculi, dilatation of the urethra and laparocystotomy with ischial urethrotomy using a polythene tube for the treatment of all those cases. He drew the conclusion that in case of rupture of the bladder with or without cystic calculi laparotomy with ischial urethrotomy was indicated.

Mohanty *et al.* (1963) reported a surgical procedure which consisted of cystotomy in prepubic area along with post-scrotal urethrotomy. He introduced a catheter through the bladder and took out at urethrotomy opening and indwelled the catheter for 7 to 10 days. He adopted this surgical procedure in five clinical cases of obstructive urolithiasis: one of them had a rupture in the bladder and got favourable result in all cases.

Noordsy and Trotter (1963) described a urethral by-pass technique for those animals in which free flow of urine through normal urethral passage cannot be restored following retention of urine due to trauma or necrosis of tissue at the neck of bladder or urethra adjacent to anterior pelvic region. A rubber tubing, transfixed into the fundus of bladder, and exteriorised through abdominal floor midway between the umbilicus and pubis, served as urethral by pass.

Frank (1964) did not sutured urethrotomy wound and allowed to heal spontaneously. In most cases strictures were formed, which may be predispose to obstruction by future calculi.

Hudson (1964) performed laparotomy in a cow and treated the case of urethral obstruction by removal of a blood clot from the vesico-genital pouch of the peritoneum which resulted into the obstruction.

Metcalf (1965) evolved a non-suture technique in closure of the urethral wound in cattle. He employed methyl-2 cyanoacrylate monomer in closure of urethral incision. It was adhesive in nature and produced a strong band between the tissues. He discussed about some common complications when urethral incision is closed by suturing or allowed to heal without suturing. Urine leakage often occurs around the sutures and when sutures are not used infiltration of urine is found in the surrounding tissues resulting into necrosis, scar formation and stricture. Stricture of the lumen resulting from scar formation reduces the animal's ability to pass even small calculi if formed. So the material which he used in closure of urethral wound in cattle helped to overcome all those complications.

Oehme and Tillman (1965) reported the cases of ruptured urinary bladder in ruminants. Following urethrotomy and drainage of accumulated urine after performing cystotomy and catheterisation, they observed spontaneous healing of the bladder in fifty percent cases.

Gulati *et al.* (1966) and Gera and Nigam (1979) found area of distal sigmoid flexure adjacent to attachment of retractor peni muscle, to be the most common site of calculus lodgement, However, in many cases calculus may get lodged between ischial arch and sigmoid flexure (Gera et al. 1973) and at ischial arch (Jubb and Keneddy, 1970).

Sharma *et al.* (1966) successfully performed pre-pubic cystotomy and ischial urethrotomy in which they used a rubber catheter for treating a male calf with ruptured urinary bladder.

Sutherland (1968) discussed a case with urethral obstruction in a cow due to a haematoma in the vaginal wall. This resulted into distention of the bladder which was later on relieved by neilsen catheter.

Anjeria (1969) experienced that complications of subcutanous urine stagnation in bovines were less by avoiding suturing of skin at the post scrotal urethrotomy site. Urethra was also left unsutured. Wound healing in such cases was earlier as compared to the cases in which skin was sutured. For pubic level urothrotomy and complication at sigmoid level a permanent urethral opening could be made.

Bone (1969) indwelled catheter (a piece of intra-venous tube) following urethrotomy in male cats to reduce effectively the incidence of post surgical blockage during the healing process. His idea was to maintain normal potency of the urethra following urethrotomy. The catheter was removed after 10-14 days post operatively.

Vig and Tyagi (1970) studied the response of urethral tissue to various suture materials in eighteen apparently healthy buffalo calves. Microscopically suture materials employed in study were not observed inside the lumen of urethra, whereas it could be easily traced in the periurethral tissue. The tissue reaction, however, could be appreciated in both the urethra and periurethral tissues. Microscopic examination of the biopsies taken on 10^{th} day of surgery revealed proliferation of epithelium more marked in catgut, whereas a very mild proliferation of epithelium could be noticed in case of cotton. Linen and silk revealed moderate proliferation,

severe reaction incited by catgut resulted in obliteration of corpus cavernosum urethrae, whereas, no obliteration was noticed in cotton. Heavy cellular infiltration observed around catgut suture with abundance of polymorphs and zone of reaction was represented by preponderance of immature fibroblasts. Cotton had comparatively less zone of reaction characterized by few polymorphs, lymphocytes, immature and mature fibroblasts. Microscopic examination of biopsies taken on 30th day of surgery with catgut revealed comparatively less hyperplastic changes in the epithelium than that on 10th day, but they were appreciably more than cotton. Linen revealed increased proliferation of the epithelium than on 10th day. Proliferation of epithelium in case of silk at 10th and 30th day was nearly the same but was decidedly less than catgut. It was also reported that suturing of facia and other ventral penile tissue over the sutured urethral passage provided double support and minimum chances of leakage.

Kelami *et al.* (1972) did replacement of the total resected urethra with alloplastic materials like human dura tube, collagen tube or collagen-Decron tube in an experimental study in dogs. Encouraging results were noted with the use of collagen-Decron tubes and lyophilized human dura tubes to eliminate stenosis at the site of anastomosis.

Rao *et al.* (1972) treated the cases of ruptured bladder and urethral obstruction in oxen with the help of indwelling catheter without repairing the rent of bladder and urethral wound edge.

Gera *et al.* (1973) successfully treated eight cases of urolithiasis by post scrotal urethrotomy and laparotomy. In all the cases urethral wounds were closed by two layers of continuous sutures using 1-0 silk thread with

atraumatic needle. Sutures were applied avoiding deep bites and narrowing of urethral passages.

Archibold and Owen (1974) preferred suturing the corpus spongiosum and urethral mucosa at urethral incision in canines. The wound gradually closed by granulation.

Prasad *et al.* (1978) preferred leaving urethral wound unsutured after urethrotomy in bullocks after removal of uroliths. Urine leakage could successfully be prevented by indwelling polythene catheter in urethra for 10-12 days. Healing was achieved uneventfully. Singh *et al.* (1978) treated three cases of urethral rupture in bullocks by creating a permanent fistula in post scrotal region. However, in none of the cases the obstructing calculi could be recovered. Recovery in all the cases was encouraging.

Sharma and Khan (1978) used catgut and silk for colocystoplasty and caecocytoplasty in buffalo calves. They observed that healing was complete with both these sutures. Microscopical studies revealed complete regeneration of the transitional epithelium after cystoplasty and tissue reaction was moderate and identical in both catgut and silk groups.

Singh *et al.* (1978) treated three cases of urethral rupture in bullocks by creating a permanent fistula in post scrotal region. However, in none of the cases the obstructing calculi could be recovered. Recovery in all the cases was encouraging.

Gera and Nigam (1979) reported that post scrotal urethrotomy to be the treatment of choice in case of urolithiasis in bullocks with moderate distension of bladder. They repaired the urethral wound with 3-0 chromic catgut using a atraumatic needle in two layers. This resulted in uneventful recovery without any seepage of urine.

Shastry and Rao (1981) successfully treated a case of bull calf by performing post scrotal urothrotomy. The opening in the urethra was closed by No.1 chromic catgut passing through the overlying facia.

Sharma *et al.* (1981) encountered cystorrhexis in 21 out of 36 clinical cases of obstructive urolithiasis. The treatment executed involved urethrotomy, cystorraphy and administration of analgesics and appropriate fluid therapy.

Bozrab *et al.* (1983) preferred to allow the prepublic urethrotomy to heal as an open wound in canines and to allow the mucosa to re-establish continuity to preclude the risk of stricture formation associated with suturing.

Sharma *et al.* (1983) observed recumbency, tympany, anemia, protrusion of rectal mucosa, problematic catheterization, cystic adhesions, cystic calculi, defective urination during recovery due to blockade and dislodgement of urethral catheter, urethral wound dehiscence, subcutaneous infiltration of urine and atony of bladder as major complications during treatment of 21 cases of bladder rupture in calves.

Tyagi and Singh (1983) reviewed the status of uraemia in cattle and buffaloes, inclusive of its etiology, varying origin, biochemical changes and treatments for respective causes. Post scrotal urethrotomy was advocated for removing calculi in cases of obstructive urolithiasis.

Waldron *et al.* (1985) compared first and second intention healing in canine urethra after performing urethrotomy on 12 normal male dogs. Surgical site was allowed to heal by second intention in six dogs. All dogs were maintained for 60 days after surgery. Packed cell volume and total serum proteins were measured before surgery and at 2, 4, 7, 14 and 60 days

after surgery. Suturing of urethra resulted in less haemorrhage during post-operative recovery period. Urethral mucosa irregularity was seen in urethrograms performed on all dogs 60 days after surgery. Urethral stricture was not observed in any of the dogs.

Weber *et al.* (1985) compared the healing of sutured prescrotal urethral incisions (12 dogs) with that of non-sutured incisions (6 dogs). Comparison was also made of healing of 5-0 polydioxanone (6 dogs) with that of 5-0 polygalactin – 910 (3 dogs), the dogs were sacrificed 3 weeks and 6 weeks respectively after surgical procedure. Surgical sites were examined grossly and urethral circumference measurements were taken at three locations (surgical site, 1.0 centimeter cranially and 1.0 centimeter caudally). Transverse sections of surgical sites were prepared and examined by light microscopy. Haemorrhage occurred post-operatively in dogs in which incisions were not sutured. Surgical sites from 6 dogs in which incisions healed by second intention had fibrosis and less inflammation than those which were sutured. There was little difference between incisions sutured with polygalactin-910 and those sutured with polydioxanone suture material. Post operative urethral stricture did not occur in any of the dogs.

Layton *et al.* (1987) compared the three anastomosis techniques of urethra. Suturing of urethra over an indwelling catheter, suturing of urethra without catheter and opposition of urethra without sutures and without an indwelling catheter in dog. Sutured and catheterized urethra had the least amount strictures as compared to other two techniques mentioned above.

Barua *et al.* (1988) made comparative study for finding best surgical technique for repair of urinary bladder, manifested healing proliferation of fibrous connective tissue. In unsutured urinary bladder, the union of wound

was effected by proliferation of transitional epithelium fibrous connective tissue and mesothelial cells respectively.

Oestenrhage *et al.* (1988) compared the new mono file absorbable suture material like maxon with chromic catgut in bladder surgery on rabbits. Maxon offered an advantage over chromic catgut in extramuscular suturing. It caused less inflammatory reaction reaction and the absence of any complication that might be attributed to it as confirmed histologically. Regarding the calculogenic potential both were no way different with each other. In their openion concretions were formed due to contact of either maxon or chromic catgut. Even *E. coli* or *Proteus* species infection did not influence stone formation.

Stewart *et al.* (1990) compared PDS, Polyglactin 910 and chromic catgut in bladder surgery of rat to study the propensity for infection, degree of inflammation, calculogenic potential, change in urine pH and suture absorption. They found that there was no difference in the calculogenic potential. None of these predisposed to infection and no correlation was found with urine pH. Absorption of PDS and polyglactin 910 was slower than that of chromic catgut. They mentioned that PDS, Polyglactin – 910 and chromic catgut would be equally valuable in bladder surgery.

Tiwari (1990) after performing urethrotomy in a buffalo bull, did suturing of urethral wound with 3-0 chromic catgut. Animal recovered uneventfully and started feeding normally.

Patil *et al.* (1992) performed urethrotomy in a Doberman dog and sutured urethral wound with catgut. Dog recovered completely on 10^{th} day without any complication.

Scherz *et al.* (1992) observed injury due to urethral catheterization in the New Zealand white rabbits. Urethral catheterization resulted in increased inflammation and fibrosis as compared to non-catheterized animals.

Haven *et al.* (1993) examined 21 cases of urolithiasis in small ruminants. He observed that cystotomy was effective method than perineal urethrotomy and urethral amputation in management of urolithiasis in small ruminants.

Ashturkar (1994) after performing post-scrotal urethrotomy in 8 bullocks, sutured urethral wound over the polythene catheter with 3-0 catgut in two rows. All the eight bullocks showed uneventful recovery.

Bose *et al.* (1994) successfully treated a case of urethral fistula in a bull calf by applying dermal graft over the damaged urethra. Suture material used was 3-0 chromic catgut. Dermal graft was covered with overlying facia of penis. The animal recovered uneventfully.

Mohinder *et al.* (1995) sutured urethra of a ram after doing perineal urethrotomy with 2-0 catgut. Animal recovered normally.

Mohindra *et al.* (1996) compared healing and degree of stricture among sutured (5 dogs), non-sutured (5 dogs) and non-sutured with amniotic membrane casings (7 dogs) by prescrotal urethrotomy. Healing was also compared between the urethral incisions sutured by 4-0 chromic catgut (3 dogs) and 4-0 polygalactin 910 (2 dogs). In dogs where urethral incisions were not sutured (with or without amniotic membrance casing), marked inflammation at urethrotomy site was noticed, marked fibrosis and severe necrosis of epithelium was observed at surgical site. Intraluminal urethral diameter tended to be large in unsutured urethra (with or without amniotic membrane). Mild inflammation at urethrotomy site was observed in dogs

with sutured urethral wound. Urethritis was a common finding in dogs in which incision was sutured and was characterized by polymorphonuclear leucocytes, plasma cells, lymphocytes and macrophages. Urethrotomy site sutured with polygalactin-910 showed less cellular reaction than those sutured with chromic catgut. In dogs with amniotic membrane casing of urethra, amniotic membrane was completely adhered without any evidence of metaplasia. The amniotic membrane showed necrotic epithelial cells with submembranous haemorrhage and severe neutrophillic infiltration and fibroplasia at periphery. The results suggested that sutured urethrotomy incisions healed uneventfully without any stricture and leakage of urine from urethrotomy site. Leakage of urine and hematuria were of lesser degree in urethrotomy incision covered by amniotic membrane.

Larson (1996) recommended the suturing of urethral wound after performing urethrotomy in bovines. The urethra can be sutured with 5-0 silk, 3-0 chromic catgut or 3-0 polygalactin 910 with closely placed interrupted sutures.

Sharma and Agrawal (1997) sutured the post – scrotal urethrotomy incision with 2-0 black braided silk after catheterisation and removal of sandy deposition . They also used 2-0 black braided silk in cystoplasty with itwact caecum in ruptured urinary bladder of Zebu cattle calf.

Sharma (1997) reconstructed partially cystectomised urinary bladder on 9 apparently healthy male buffalo calves in which anterior half of the organ was resected and remaining bladder stump was sutured with 3-0 black braided silk on serosa of invaginated caecal apex. All of the operated animals survived well. Gross and histological examination revealed complete

regeneration of bladder tissue with typical transitional cell epithelium on caecal serosa on 90th post-operative day.

Prakash *et al.* (1998) compared the catheterised sutured urethrotomy with polyglactin-910 in simple continuous pattern with unsutured catheterised wound and observed non-significant increase in the diameter of unsutured wound.

Prakash *et al.* (1999) reported that suturing of urethral mucosa with covernous spongiosum and tunica albuginea in simple continuous pattern was best technique for management of urethrotomy wound.

Prakash *et al.* (2000) conducted urethrotomy on twelve clinically healthy cross bred male cow calves. They were divided into two group of six animals in each. Group-I served as control group in which after doing urethrotomy and intubation, urethral mucosa was left unsutured where as in group-II urethral mucosa was sutured with 3-0 polyglactin-910. They observed non significant variation in rectal temperature, heart rate and respiration rate in both groups. In group -I, two animals showed severe urine infiltration on dependent subcutaneous abdominal area. No post-operative complication was observed in group-II. They also observed no stricture formation in any group of animal.

Haematological and biochemical changes :

Rajmani and Ganapathy (1965) created a zig-zag tear in the bladder wall and produced uremia in calves. The highest level of BUN was found to be 468 mg per cent. The normal values of BUN in buffalo calves varied from 8-26 mg per cent. Haematological studies were also carried out by them.

Watts and Campbell (1970) performed nephrectomy in four bullocks and studied various biochemical constituents in the blood. Plasma urea

increased following operation and showed a mean rise of 35 mg per cent/day. Creatinine value also showed an increase like blood urea nitrogen. There was no rise in plasma inorganic phosphate. No changes were noticed in plasma sodium, potassium and chloride except a low value of sodium in one bullock.

Prasad (1972) observed the fluctuations, within physiological norms, in the values of sodium, potassium, chloride and urea concentrations following caecocystoplasty in buffalo calves.

Mohanty (1973) noticed increased value of blood urea nitrogen, creatinine, inorganic phosphate and potassium, and very little changes in the values of sodium and chloride after experimental ligation of urethra in calves. He further noticed the values of blood urea nitrogen, serum creatinine, inorganic phosphate and potassium came down within four days of caecocystoplasty and sodium, chloride, haematocrit and haemoglobin values were fluctuating.

McIntosh *et al.* (1974) studied 93 cases of urolithiasis and found protein as an important starting component in the calcium carbonate types of calculi and concluded that dietary and managemental factors could be important for the onset and development of outbreaks.

Munakata *et al.* (1974) concluded that there was hardly any change in haemoglobin and haematocrit value in cattle affected with urolithiasis syndrome. However, little change occurred to the serum levels of calcium, inorganic phosphorus, magnesium and blood sugar. In severe cases, blood urea nitrogen showed an increase.

Gera (1976) following ligation of urethra, observed that there was a progressive increase in haemoglobin values from pre-ligation mean value of

9.20 gm per cent to 12.85 gm per cent at the time of death and there was also rise in haematocrit value at 12 hours post-ligation and then from 48 hours post-ligation till death.

Finco and Cornelius (1977) induced urethral obstruction in adult male cats and noticed clinical signs identical with those observed in naturally occurring disease.

Sharma and Khan (1978) were used catgut and silk for colocystoplasty and caecocystoplasty in 16 buffalo calves. There was not much variation in the level of blood urea nitrogen (BUN) in either of the suture materials used.

Gera and Nigam (1980) studied haematological and biochemical changes of urethral ligation in calves. There was progessive increase of haemoglobin, haematocrit, total erythrocyte and leucocytic count post ligation. There was no significant different between the values of erythrocyte sedimentation rate at different intervals. There was initial increase in BUN, serum creatinine and inorganic phosphorus, the values declined after 36 hours after ligation and again there was progressive increase in these values upto the terminal stages. There was a gradual increase of plasma potassium during this study.

Gera *et al.* (1980) studied biochemical changes following trans-plantation of preserved bladder allografts in buffalo calves. They found significant increase in BUN value to a mean value of 38.03 mg/100 ml after 24 hours of urethral ligation. The value further increased to 41.05 mg percent at the interval of 24 hours post cystoplasty and decreased afterwards and returned to within normal limits at 120 hours post Cystoplasty. Creatinine levels ranged between 2.00 to 4.00 mg/100 ml after 24 hours of urethral ligation, continuously decreased afterwards. The values were not

significantly different from the preligation values at 48 hour post cystoplasty.

Singh *et al.* (1981) observed that biochemical examination of blood in clinical cases of bovine urolithiasis, significant increase in the levels of blood urea nitrogen, creatinine, inorganic phosphorus, Magnesium and Potassium when compared to normal ones. During postobstruction period these levels decreased progressively. Haemoglobin slightly increased and clotting time prolonged.

Amer *et al.* (1982) studied clinicopathological picture after urethral obstruction in rams. Surgical occlusion of the urethra was carried out in four rams and blood samples collected at different intervals after the operation. Progressive increase occurred in blood urea nitrogen, blood non preoteinous nitrogen, serum creatinine, serum potassium and serum inorganic phosphorus. Gross and histopathological lesions characteristics of uraemia were observed.

Donecker and Bellamy (1982) carried out the survey of blood chemical abnormality in cattle with ruptured urinary bladder and ruptured urethra. They observed increase in serum creatinine and BUN.

Kulkarni *et al.* (1985) studied the alteration in certain biochemical parameters following experimentally induced rupture of urinary bladder and development of uraemia in male calves. They observed that increases in BUN and createnine value at 48 hours of uroperitoneum were highly significant.

Barua, *et al.* (1990) studied certain blood biochemical parameters of cow calves due following experimental rupture and repair of the urinary bladder. There was elevation of bicarbonates at 48 hours following ligation

of urethra but it declined slowly to almost normal limits within 108 hours following repair of bladder. There was significant increase in blood urea nitrogen and creatinine values following 48 hours of urethral ligation. With treatment these values gradually declined and reached almost normal at 108 hours of repair of urinary bladder.

Gangwar, *et al.* (1991) observed biochemical profile in experimental uraemia after urethral obstruction in crossbred calves. They found a more pronounced and consistent increase in serum creatinine level during the terminal stage of uraemia. Serum creatinine had almost ten fold increased where as serum urea nitrogen increased by four fold.

Kumar *et al.* (1991) observed that obstructive urolithiasis in bullocks was associated with marked increase in haemoglobin, packed cell volume, total erythrocytic count, blood urea nitrogen, serum creatinine and potassium significant decrease in these values during post-operative period was noted serum sodium level was subnormal to normal. There was hypochloremia and mixed acidosis or alkalosis.

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Chapter - 3

MATERIALS AND METHODS

Materials and Methods

For the present study twenty four male buffalo calves, about one year of age and weighing between 80-110 kilogram were obtained from the experimental animal supplier of Bihar Veterinary College, Patna. The selected animals were apparently healthy and in good condition. All animals were kept under similar environmental condition and kept under close observation for period of one week, during that period a preliminary examination was done just to check up their health and ensure that animals were healthy. All animals were dewormed with **Albendazole**[1] at dose rate of 5 mg per kg body weight, a fortnight before the start of the experiment.

Materials :

1. *Polythene tube :* Polythene tubes of 3.6 to 3.9 mm of diameter were used in the present experiment.The wall of the tubes were about 0.5 mm in thickness (Fig. 1).

2. *Suturing materials :*

   i. **Chromic catgut**[2] - 3 – 0

   ii. **Polyglactin- 910**[3] - 3 – 0

   iii. **Black braided silk**[4] – 3 – 0

Design of Experiment :

The selected animals were randomly divided into four groups consisting of six animals in each group. Out of these four groups, one group

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1. Albomar, Glaxo Ltd., Dr. Annie Besant Road, Mumbai-400025.
2. Mersutures, Ethicon, Johnson & Johnson Ltd., 30, Forjett, street Mumbai-400 036.
3. Vicryl, Ethicon, Johnson & Johnson Ltd., Aurangabad-431136.
4. Merisilk, Ethicon, Johnson & Johnson Ltd., 30, Forjett, street, Mumbai –400 036.

(Group A) was kept as control and the remaining three group of animals were allotted to three different types of suturing materials to close the urethrotomy wound.

Group A : Animals of this group were kept as control group. Urethral wound edges were left unsutured after performing urethrotomy.

Group B : In this group after performing urethrotomy, urethral incisions were sutured with 3-0 chromic catgut in closely placed interrupted pattern suture.

Group C : In this group after performing urethrotomy urethral incisions were sutured with 3 – 0 polyglactin – 910 in closely placed intrupted pattern suture.

Group D : In this group after performing urethrotomy urethral incision were sutured with 3 – 0 black braided silk in closely placed intrrupted pattern suture.

Preparation of animals and anaesthesia :

Each animal was secured in lateral recumbency with the upper hind leg stretched forward and lower hind leg pointed posteriorly. About 25-30 centimeter wide area extending from scrotum to ischial arch was shaved and washed with **carbolic soap**[5] and finally scrubbed with 1 in 30 **chlorhexidine solution**[6] before covering with the sterlized drape Anaesthesia was insured by infiltration of 2% **Lignocaine hydrochloride**[7] in median raphe extending behind the scrotum towards ischial arch.

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5. Lifebuoy soap, Hindustan Liver Ltd.

6. Acetik, ICT India Ltd., Industrial Area, Chandigarh.

7. Hoechst India Ltd., Hoechst House, Nariman Point, Mumbai-400021.

Operative technique :

After draping the site of operation, about 6 cm long incision was given on the median raphe post scrotally After incising the skin and subcutaneous tissues, the aponeurosis of muscles was bluntly separated. Penis was levered out by passing a curved scissor underneath it (Fig.2). The peripheral facia around the penis was removed with gentle incisions, while avoiding the penile blood vessels. A considerable part of penis was exteriorised by straightening the sigmoid flexure. About one to two centimeter of incision was given on the urethra just above the attachment of retractor peni muscles. After urethrotomy urethral intubation was done with sterilized snugly fitted polythene catheter upto ischial arch in all the groups of animals (Fig.3). Urethral incision was sutured or unsutured as per the grouping of animals. The penile incised sheath and facia were sutured with same suturing material, which was used in closure of urethral incision (Fig.4 & 5). After sprinkling of **Hostacycline powder**[8] at operative site penis was placed in normal position. Skin was closed by Halsted stitch with silk. (Fig.6). Polythene catheter was anchored to sheath using nylon thread. Post Operative Care

Surgical wounds were dressed with **Povidone iodine**[9] thrice daily for three day followed by once daily until the cutaneous wound healing. **Sulphadiazine and trimethoprim inj.**[10] was given intramuscularly at the dose rate of 1 ml / 15 kg body weight for five days. Skin stitches were

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8. Hoechst India Ltd. Hoechst House, Nariman Point, Mumbai –400021.

9. Wokadin, Wockhard Veterinary Ltd., Mumbai-400018.

10. Biotrim, Ranbaxy Laboratory Ltd., New Delhi-110020.

Removed on 10th post-operative day. Polythene catheter was removed from urethra on 25th day of surgery in all the animals.

Parameters :

1. *Clinical signs :* Rectal temperature, respiration rate, pulse and heart rate of all animals were taken on 0 day (day before surgery), 1st, 2nd, 3rd, 7th, 21st and 28th day after surgery. Gross examination of operation site, infiltration of urine, feeding, urination, defecation, hydration status (skin tenting methods), visible mucous membrane was observed daily till the end of study. Gross examination of incision site for leakage, concretion or any untoward pathological lesion on operated area was observed at the time of collection of urethra for the histopathological examination.

2. *Haematological studies :* Blood samples were collected from the jugular vein on 0 day (day before surgery) 1st, 2nd, 3rd, 7th, 14th, 21st and 28th days after surgery. Blood samples were analysed for

i Total leucocyte count (TLC)-It was done in a haemocytometer (Neubauer, S. Chamber).

ii.Differential leucocyts count (DLC)-It was done as per method given by Schalm (1962).

iii. Haemoglobin (Hb)-It was determine by Sahli's method.

3. *Biochemical observation :* Blood samples were collected from jugular vein on 0 day (day before surgery) 1st, 2nd, 3rd, 7th 14th, 21st and 28th days after surgery for studies of

i. Blood urea nitrogen (BUN) (Levina *et al.,* 1961)

ii. Serum createnine (Folin and Wu, 1919)

4. *Histopathological studies :* Urethral tissues from the operated area including pre and post operated portion of urethra were collected on 45th post operative day and preserved in 10% neutral buffered formalin solution. After

48 hours fixation of urethra, tissue sample collected from three location viz. operated area, about 1 cm. posterior to operated area (towards ischial arch) and about 1 cm. anterior to operated area. (towords glans penis) The pieces of one centimeter of each sample were processed by cedar wood method and section of 5-6 μm thickness were cut with rotatory microtome. Section were stained with following stain.

i. Haematoxylin and eosin stain (Luna, 1968)

ii. Van Giesson's method (Druvy and Wallington, 1967).

iii. Verhoeff's stain (Pearse, 1968)

iv. Periodic acid schiff stain/ Alcian blue (Seeshan and Hropchak, 1973).

5. *Micrometry* : Micrometry was done to measure the luminal size of the urethra. For this purpose a fillar micrometer was used. The fillar micrometer was duly calibrated with stage micrometer at different magnifications. To measure the luminal size of urethra, two diameters were taken i.e. major diameter and minor diameter. The average of major and minor diameter was considered as the Luminal diameter of urethra at pre, post- surgical sites.

Statistical analysis :

Statistical analysis of the data obtained from clinical studies haematological studies biochemical studies and micrometic observation at different stages of observation were carried as per method suggested by Snedecor and Cochran (1967). Parameters in percentages were analysed after converting them to corresponding angle (Where, angle = $\arcsin\sqrt{\text{percentage}}$), as given by C.I. Bliss (Snedecor and Cochran, 1967).

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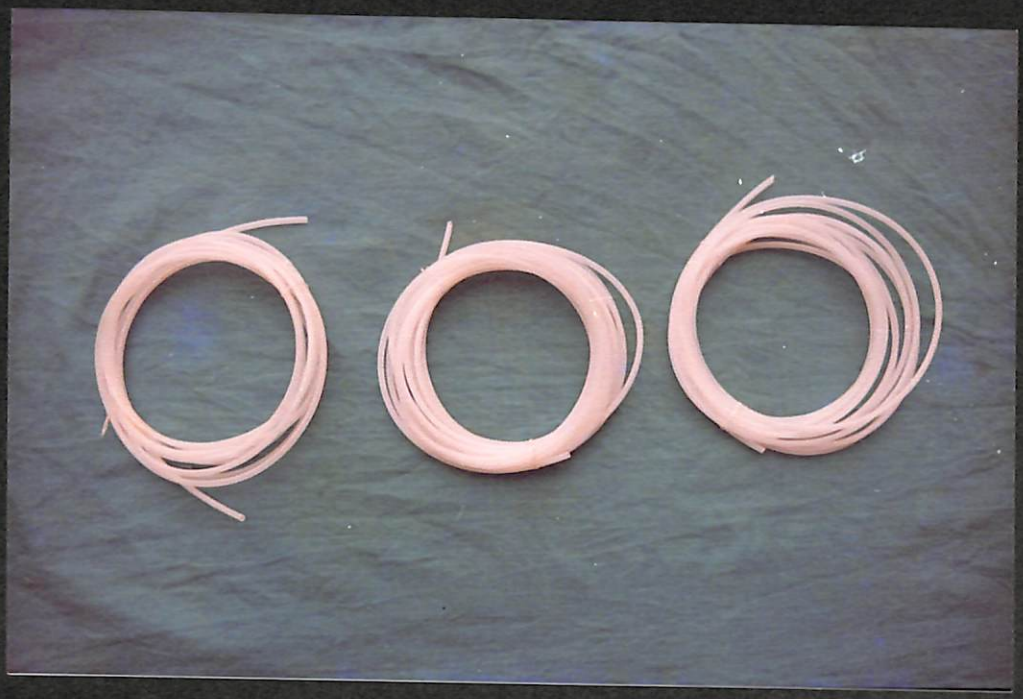

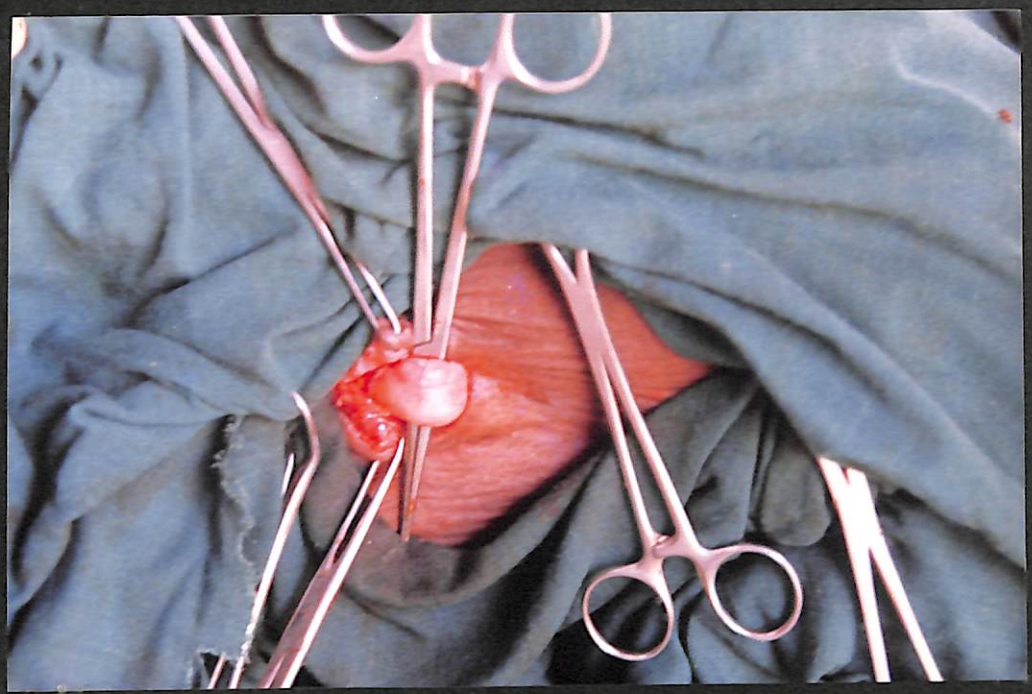

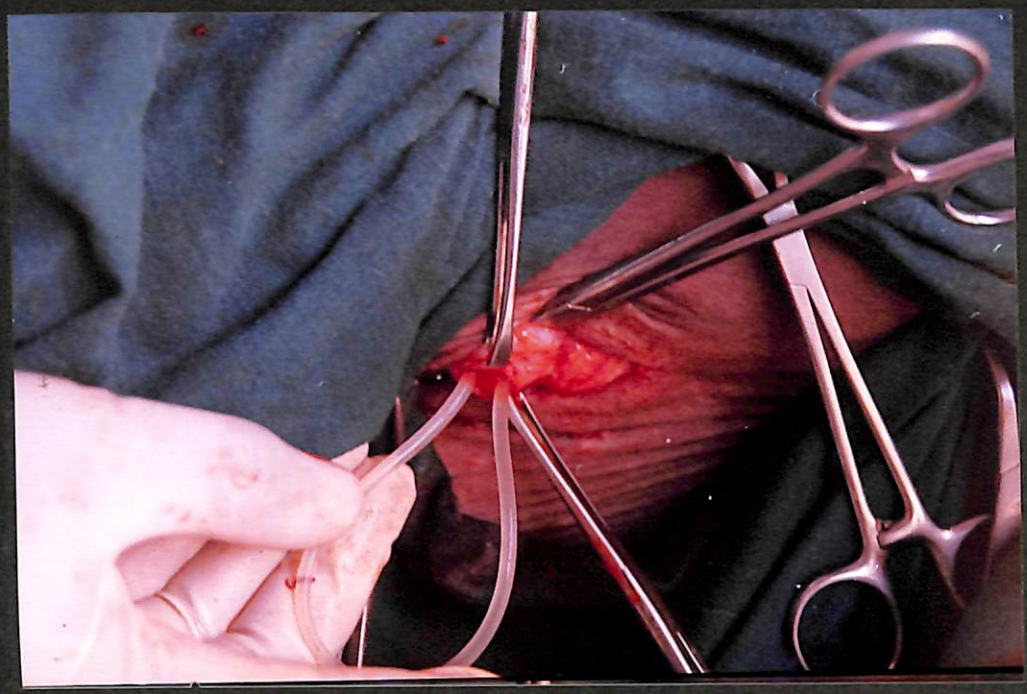

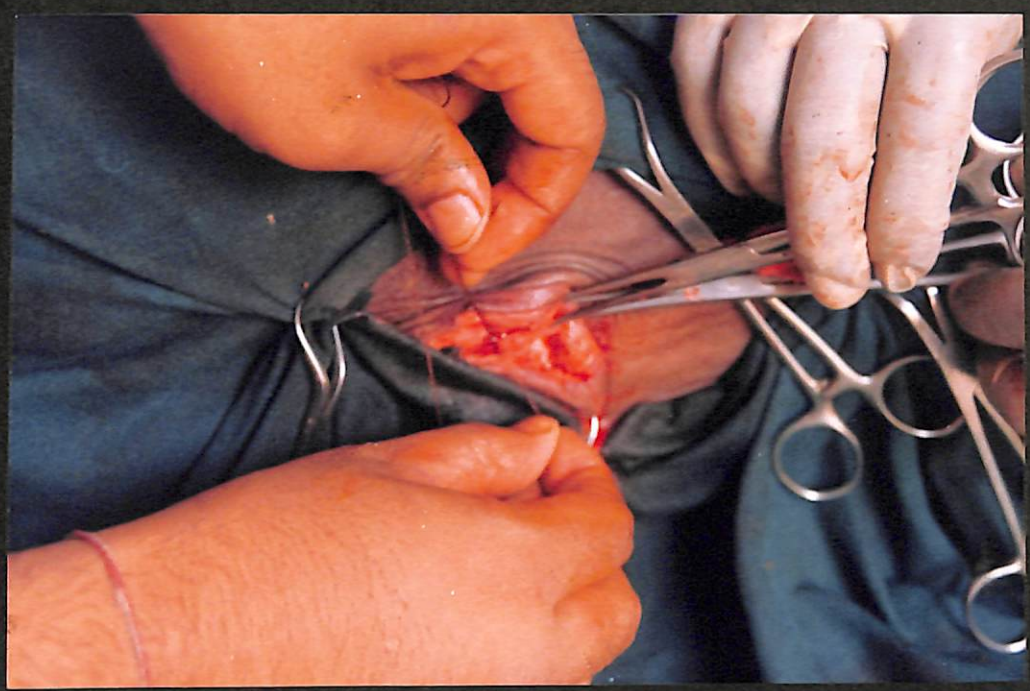

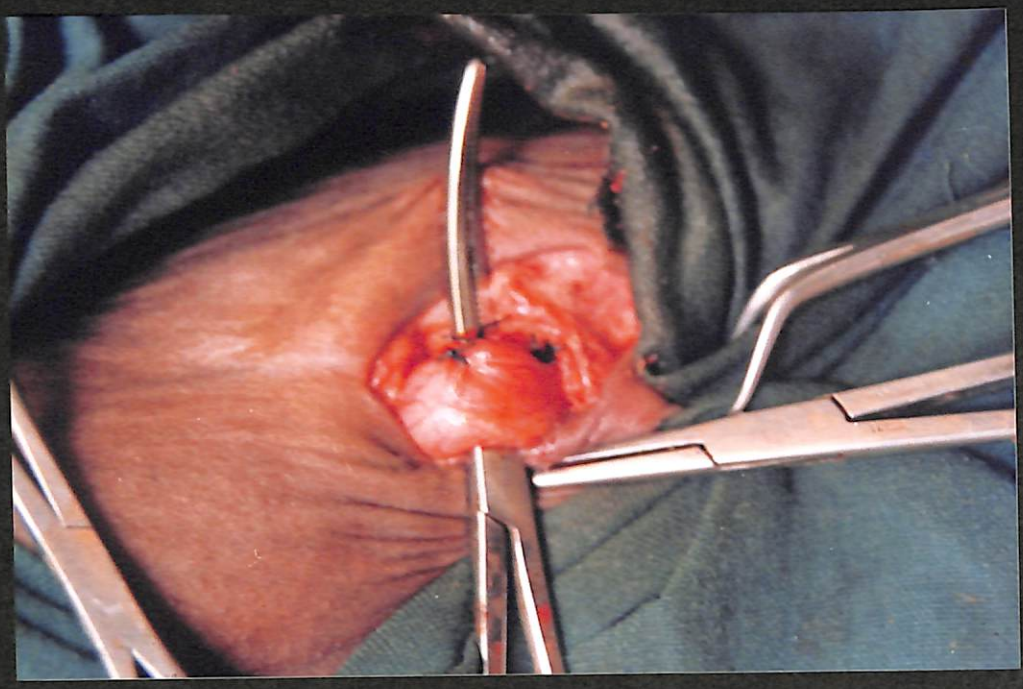

Results

In the present study three types of suturing materials, consisting of chromic catgut, polyglactin-910 (absorbable) and black braided silk (non-absorbable) were used for closure of post-scrotal urethrotomy wound of buffalo calves, where as, in control group post-scrotal urethrotomy wound was left unsutured. Urethral intubation up to the level of ischial arch was done with sterilized snugly fitting polythene catheter in all the groups. In this chapter the results of the following different parameters studied were recorded, to evaluate the results of this experiment.

1. Anaesthesia and surgery.
2. Clinical observation.
3. Haematological observation.
4. Biochemical observation.
5. Macroscopic observation.
6. Micrometric observation of lumen of urethra.
7. Histopathological observation.

Anaesthesia and surgery :

Post-scrotal local infiltration of 2% lignocaine hydrochloride produced satisfactory analgesia. Proper securing of animals and analgesia did not allow much movements of animals during surgery. Mild haemorrhage after urethral incision was checked with gentle pressure. During the course of surgery no other complication was recorded ever. Urethral intubation with proper sized sterlized polythene catheter was performed with ease. After this Hostacycline was sprinkled over urethrotomy wound. Intramuscular injection of sulphadiazine and trimethoprim was given post operatively which did not cause any hypersensitivity reaction.

Clinical observations :

Group A: All the animals showed normal activity upto last date of observation. Appetite , rumination and defecation were normal except in two animals, which developed severe subcutaneous infiltration of urine at the operative site. The mucous membrane of the eyes and hydration status were observed normal. Two animals in this group passed urine normally through polythene catheter and there were no infiltration of urine in the dependent abdominal area till the last day of observation. In other two animals of this group moderate infiltration of urine was noted at the site of operation extending up to sheath and ventral abdominal area after 24 hours of surgery. The infiltration subsided completely on 6^{th} day postoperative. One animal of this group removed the polythene catheter on 16^{th} postoperative day. However, the animals urinating normally throughout the observation periods. Healing of incision wound was normal in all these four animals. Rest two animals of this group had seepage of urine from the incision site along with severe infiltration of urine on dependent ventral abdominal area. To avoid the necrosis of muscles and skin, infiltrated urine was drained off by giving stab incision on either sides of the prepuce in both the affected animals. The stab wounds were dressed regularly with Povidone iodine ointment[11]. In two animals urine was seeping out at post-scrotal incision site, where a gap was formed after removal of skin suture which healed by second intention.

Group B: General activity of all the animals were normal. Appetite, rumination, mucous membrane of eye, hydration status and defecation were also normal. There were no infiltration and seepage of urine in four animals

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11. Betadine Wockhardt Pvt. Ltd. Animal Health Division Dr. Annie Besant Road, Mumbai – 400018.

day. One animal of this group removed the polythene catheter on 15th post-operative day. Inspite of removal of polythene catheter no complication was observed in voiding the urine in this animal.

The mean ± SE and CV% of body temperature of male buffalo calves at different periods of interval have been presented in table-1. The mean body temperature ranged from 101.21 to 102.17 ^{0}F in group A, 101.25 to 102.20 ^{0} F in group B, 101.25 to 101.98 ^{0} F in group C and 101.52 to 102.32 ^{0} F in group D. Body temperature in each group was shown to be increased on 2nd and 3rd day after urethrotomy and then started declining on 7th day onwards. As compared to zero (0) day, the body temperature on 2nd and 3rd day increased by 0.91 and 0.96 ^{0} F in group A, 0.68 and 0.60 ^{0} F in group B, 0.53 and 0.68 ^{0} F in group C and 0.8 and 0.61 ^{0} F in Group D, respectively. However, the analysis of variance presented in table-2 did not reveal any significant effect of urethrotomy on body temperature.

The mean ± SE and CV % of respiration rate of male buffalo calves at different days of interval have been presented in table-3. The mean respiration rate was found to be ranged from 21.33 to 23.33 per minute in group A, 21.33 to 23.17 per minute in group B, 21.00 to 22.66 per minute in group C and 21.33 to 23.00 per minute in group D. Respiration rate in each group was shown to be increased gradually from 1st day onwards and reached to the maximum on 3rd day and then started declining on 7th day onwards. As compared to zero (0) day, the respiration rate on 2nd and 3rd post-operative day were observed to be increased by 1.67 and 2.00 per minute in group A, 1.5 and 1.67 per minute in group B, 1.33 and 1.66 per minute in group C and 1.33 and 1.5 per minute in group D, respectively. The analysis of variance for the effect of urethrotomy on respiration/minute is

of this group. Wound healed normally and stiches were removed on 10th post operative day. One animal of this group showed gaping of wound on 6th post operative day, with seepage of urine leading to permanent fistula formation. This animal was passing urine also through the catheter. Fistula of that animal reduced to pin head size within 25th day of surgery, but did not heal completely and was discharging urine in drops at cutaneous region throughout the observation periods. In one animal moderate infiltration as well as seepage of urine from operation site was observed within 24 hour after surgery. Later on this animal also developed fistula at the site of operation, which did not heal completely till the end of the study. No inflammatory reaction was observed at the site of operation in any of the animal.

Group C : General activity of the animals was normal and did not show any sign of ill health till the end of study. The mucous membrane, hydration status, appetite, rumination defecation and urination were normal. The wound healed without any complication in all the animals. Skin stiches were removed on 10th post-operative day. Two animals of this group removed the polythene catheter on 10th and 15th post-operative day. Inspite of removal of polythene catheter no complication was observed in voiding the urine in these animals.

Group D : Animals of this group showed normal general activity upto last day of observation. Mucous membrane of eye and hydration status were also normal. All the animals showed normal feeding, rumination and defecation and were urinating normally till the end of the study. One animal of this group showed moderate infiltration of urine at operation site, which subsided after 5-6th day of surgery. Skin sutures were removed on 10th post-operative

day. One animal of this group removed the polythene catheter on 15th post-operative day. Inspite of removal of polythene catheter no complication was observed in voiding the urine in this animal.

The mean ± SE and CV% of body temperature of male buffalo calves at different periods of interval have been presented in table-1. The mean body temperature ranged from 101.21 to 102.17 ^{0}F in group A, 101.25 to 102.20 ^{0} F in group B, 101.25 to 101.98 ^{0} F in group C and 101.52 to 102.32 ^{0} F in group D. Body temperature in each group was shown to be increased on 2nd and 3rd day after urethrotomy and then started declining on 7th day onwards. As compared to zero (0) day, the body temperature on 2nd and 3rd day increased by 0.91 and 0.96 ^{0} F in group A, 0.68 and 0.60 ^{0} F in group B, 0.53 and 0.68 ^{0} F in group C and 0.8 and 0.61 ^{0} F in Group D, respectively. However, the analysis of variance presented in table-2 did not reveal any significant effect of urethrotomy on body temperature.

The mean ± SE and CV % of respiration rate of male buffalo calves at different days of interval have been presented in table-3. The mean respiration rate was found to be ranged from 21.33 to 23.33 per minute in group A, 21.33 to 23.17 per minute in group B, 21.00 to 22.66 per minute in group C and 21.33 to 23.00 per minute in group D. Respiration rate in each group was shown to be increased gradually from 1st day onwards and reached to the maximum on 3rd day and then started declining on 7th day onwards. As compared to zero (0) day, the respiration rate on 2nd and 3rd post-operative day were observed to be increased by 1.67 and 2.00 per minute in group A, 1.5 and 1.67 per minute in group B, 1.33 and 1.66 per minute in group C and 1.33 and 1.5 per minute in group D, respectively. The analysis of variance for the effect of urethrotomy on respiration/minute is

presented in table-4, which did not reveal any significant effect of urethrotomy on respiration rate.

The mean ± SE and CV% of pulse rate of male buffalo calves at different days of interval have been presented in table-5. The mean pulse rate ranged from 57.34 to 60.83 per minute in group A, 57.17 to 60.5 per minute in group B, 57.33 to 60.17 per minute in group-C and 57.5 to 59.5 per minute in group D. Pulse rate in each group was shown to be increased on 2^{nd} and 3^{rd} post operative days and then started declineing on 7^{th} day onwards. As compared to zero (0) day, the pulse rates on 2^{nd} and 3^{rd} post operative day were observed to be increased by 2.7 and 3.50 per minute in group A, 2.67 and 3.00 per minute in group B, 2.67 and 2.84 per minute in group C and 1.34 and 1.62 per minute in group D, respectively. However, the increase in pulse rate was not found to be statistically significant (Table 6).

The mean ± SE and CV% of heart rate of male buffalo calves at different days of interval have been presented in table-7. The mean heart rate ranged from 78.10 to 80.17 per minute in group A, 78.33 to 80.17 per minute in group B, 78.33 to 79.83 in group C and 78.66 to 80.33 in group D. The heart rate in each group was shown to be increased gradually and became maximum on 2^{nd} and 3^{rd} post-operative days and then started declining from 7^{th} day onwards. However, the analysis of variance (Table-8) did not reveal any significant effect of urethrotomy on heart rate. The heart rate was observed to by increased by 2.07 and 2.07 per minute in group A, 1.5 and 1.33 per minute in group B, 1.5 and 1.33 per minute in group C and 1.34 and 1.67 per minute in group D, respectively on 2^{nd} and 3^{rd} post operative day as compared to heart rate on zero day.

Table:- 1. Mean ± S.E. and CV % of Body temperature (^{0}F) in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	101.21±0.61	1.48	101.35±0.64	1.56	102.12±0.79	1.89	102.17±0.87	2.09	101.53±1.14	2.74	101.70±0.80	1.94	101.35±0.69	1.68	101.35±0.72	1.75
B	101.52±0.65	1.58	101.25±0.60	1.45	102.20±0.90	2.15	102.12±0.87	2.07	101.75±0.83	1.99	101.48±1.16	2.79	101.72±0.55	1.32	101.57±0.72	1.74
C	101.30±0.60	1.46	101.38±0.66	1.58	101.83±0.87	2.09	101.98±0.85	2.04	101.43±0.73	1.75	101.25±0.50	1.22	101.55±0.72	1.74	101.57±0.72	1.74
D	101.52±0.73	1.76	101.65±0.68	1.63	102.32±0.85	2.03	102.13±0.79	1.84	101.90±0.53	1.28	101.78±0.56	1.34	101.78±0.30	0.71	101.60±0.58	1.39

No.of observation in each group, (n) = 6.

Table:-2 Analysis of variance for the effect of suture material used in urethrotomy on body temprature in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.9604	0.282174 NS
Between days	7	2.0104714	0.5906 NS
Error	181	3.403366	

NS :- Non- significant.

Table:- 3. Mean ± S.E. and CV % of respiration/minute in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	21.33±0.88	10.13	21.67±0.95	10.79	23.00±1.24	13.19	23.33±1.33	14.00	22.00±1.18	13.17	21.67±1.05	11.92	21.83±1.13	12.77	21.39±1.08	12.39
B	21.50±0.92	10.50	21.83±0.87	9.79	23.00±1.06	11.34	23.17±1.11	11.71	21.83±1.01	11.38	21.67±1.05	11.92	21.66±0.92	10.39	21.33±0.88	21.33
C	21.00±0.89	10.45	21.33±1.15	12.56	22.33±1.15	12.56	22.66±1.05	11.39	22.00±1.15	12.86	21.83±0.95	10.61	21.50±0.76	8.70	21.00±1.00	11.66
D	21.50±1.02	11.67	21.50±1.06	12.04	22.83±1.14	12.21	23.00±1.15	12.30	22.50±0.99	10.80	21.66±1.02	11.56	21.5±0.99	11.30	21.33±1.15	13.15

No.of observation in each group, (n) = 6.

Table:-4. Analysis of variance for the effect of suture material used in urethrotomy on respiration rate in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	1.528	0.2089 NS
Between days	7	10.7028	1.9398 NS
Error	181	5.5173	

NS :- Non- significant.

Table:- 5. Mean ± S.E. and CV % of pulse/minute in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	57.34±1.83	7.85	58.33±2.25	9.32	60.00±1.65	6.75	60.83±1.96	7.88	59.33±2.09	8.64	57.83±2.02	8.57	58.16±2.28	9.56	57.50±2.85	12.13
B	57.50±1.92	8.18	58.66±2.46	10.26	60.16±2.07	8.44	60.50±1.95	7.87	59.66±2.08	8.52	57.66±1.80	7.65	57.17±1.64	7.03	57.33±1.82	7.77
C	57.33±2.18	9.31	58.33±1.94	8.16	60.00 ±1.86	7.60	60.17±1.96	7.96	57.66±2.08	8.82	57.55±1.93	8.20	57.66±1.98	8.40	58.00±1.81	7.63
D	57.88±1.83	7.74	57.50±1.83	7.80	59.16±2.01	8.31	59.50±2.09	8.60	59.00±1.90	7.88	58.66±1.86	7.75	58.50±1.57	6.57	58.00±1.48	6.26

No of observation in each group, (n) = 6

Table:-6. Analysis of variance for the effect of suture material used in urethrotomy on pulse rate in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	1.1702767	0.05818 NS
Between day	7	25.384754	1.262078 NS
Error	181	20.113458	

NS :- Non- significant.

Table:- 7. Mean ± S.E. and CV % of Heart/minute in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	78.10±1.07	3.36	79.50±1.18	3.62	80.17±1.16	3.56	80.17±1.60	4.89	79.33±1.66	5.13	78.66±1.81	5.66	78.33±1.66	5.19	78.33±1.56	4.89
B	78.33±1.17	3.66	79.50±1.31	4.04	79.83±1.19	3.67	80.17±1.33	4.05	79.33±1.33	4.10	78.33±1.35	4.22	78.66±1.38	4.30	78.33±1.36	4.25
C	78.33±133	4.16	79.66±12.8	3.94	79.83±1.47	4.51	79.66±1.26	3.86	79.17±1.30	4.03	78.66±1.26	3.91	78.50±1.46	4.54	78.66±1.12	3.47
D	78.66±0.95	2.94	79.83±1.33	4.07	80.00±1.44	4.40	80.33±1.31	3.99	79.33±1.08	3.33	78.83±1.30	4.40	78.67±1.25	3.89	78.83±1.08	3.34

No.of observation in each group, (n) = 6 .

Table:-8. Analysis of variance for the effect of suture material used in urethrotomy on heart rate in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.6251	0.0657 NS
Between days	7	11.2143	1.1790 NS
Error	181	9.5114	

NS :- Non- significant.

Haematological Observation :

The mean ± SE and CV % of total leucocytes count (thousand/cu mm) at different days of interval have been presented in table-9. The mean total leucocyte count ranged from 8.83 to 9.67 thousand/cu mm in group A, 8.76 to 9.66 thousand/cu mm in group B, 8.80 to 9.46 thousand/cu mm in group C and 8.84 to 9.45 thousand/cu mm in group D. The total leucocyte count in each group was shown to be increased on 2nd and 3rd post operative days and then started declining on 7th day onwards. As compared to zero (0) day, the total leucocyte count on 2nd and 3rd post-operative days were observed to be increased by 0.77 and 0.84 thousand/cu mm in group A, 0.62 and 0.67 thousand/cu mm in group B, 0.65 and 0.66 thousand/cu mm in group C and 0.56 and 0.65 thousand/cu mm in group D, respectively. However, this increase in leucocyte count was statistically non-significant.

The analysis of variance (table-10) did not reveal any significant effect of urethrotomy on total leucocyte count.

The mean ± SE and CV% of neutrophil % of male buffalo calves at different period of interval have been presented in table-11. The mean neutrophil count was found to be ranged from 36.35 to 39.56% in group A, 35.96 to 39.86% in group B, 36.10 to 38.75% in group C and 35.95 to 39.09 in group in group D. Neutrophil % in each group was shown to be increased on 2nd and 3rd days of urethrotomy and then started declining on 7th day onwards. Neutrophil % of 2nd and 3rd day of post-operative period was found to be increased by 1.38 and 1.88 % in group, 1.38 and 1.97 % in group B, 1.50 and 1.50 % in group C and 1.68 and 1.88 % respectively as compared to zero (0) day.

The analysis of variance for the effect of urethrotomy on neutrophil %

is presented in table -12 did not reveal any significant variation.

The mean ± SE and CV% of lymphocyte % of male buffalo calves at different intervals have been presented in table-13. The mean lymphocyte % found to be ranged from 52.83 to 55.74 % in group A, 54.02 to 55.90 % in group B, 54.37 to 56.22 % in group C and 54.34 to 56.23 in group D. Lymphocyte % in each group was shown to be decreased on 2nd and 3rd day after urethrotomy and then started inclining on 7th day onwards. Lymphocyte % of 2nd and 3rd after surgery was decreased by 1.68 and 2.91 % in group A, 1.54 and 1.88 % in group B, 1.37 and 1.85 % in group C and 1.19 and 1.55 % respectively as compared to zero (0) day. However, this decrease was statistically non-significant.

The analysis of variance presented in table-14 did not reveal any significant effect of urethrotomy on lymphocyte %.

The mean ± SE and CV % of monocyte % of male buffalo calves at different periods of interval have been presented in table-15. The mean monocyte % ranged from 2.08 to 2.67 % in group A, 2.04 to 2.52 in group B, 2.04 to 2.69 % in group C and 2.06 to 2.56 % in group D. Monocyte % in each group was shown to be slightly decreased on 2nd and 3rd day after urethrotomy and again increased from 7th day onwards. However, this increase and decrease was statistically non-significant.

The analysis of variance (table-16) did not reveal any significant effect of urethrotomy on monocyte %.

The analysis of variance showing the effect of wound on eosinophil percent at different days of interval is depicted in table-18, which did not reveal any significant difference of eosinophil percentage between the groups as well as at different days of interval.

The mean ± S.E. and C.V.% of haemoglobin (gm%) of male buffalo calves at different interval of days have been presented in table-19. There were slight variation observed between different group and between differ interval of days. However this variation is non-significant.

Biochemical observation :

The mean ± SE and CV% of blood urea nitrogen of male buffalo calves at different period of interval have been presented in table-21. The mean blood urea nitrogen ranged from 21.33 to 23.73 mg % in group A, 21.45 to 23.18 mg % in group B, 20.38 to 22.82 mg % in group C and 21.48 to 23.43 mg % in group D. Blood urea nitrogen in each group was shown to be increased on 2^{nd} and 3^{rd} day and then started decline on 7^{th} day onwards. As compared to zero (0) day 2^{nd} and 3^{rd} day the blood urea nitrogen was increased by 1.4 and 2.1 mg % in group A, 1.32 and 1.73 mg % in group B, 1.49 and 0.72 mg % in group C and 1.78 and 1.95 mg % in group D, respectively. However, this increase was statistically non-significant.

The analysis of variance for the effect of urethrotomy on blood urea nitrogen is presented in table-22 did not reveal any significant difference on blood urea nitrogen between groups.

The mean ± SE and CV % of serum createnine of male buffalo calves at different periods of interval have been presented in table-23. The mean serum createnine ranged from 2.13 to 2.36 mg % in group A, 2.15 to 2.55 mg % in group B, 2.20 to 2.50 mg % in group C and 2.17 to 2.38 mg % in group D. serum createnine in each group was shown to be increased on 2^{nd} and 3^{rd} day and then started declining from 7 day onwards. The serum createnine was increased by 0.17 and 0.23 mg % in group A, 0.28 and 0.35 mg % in group B, 0.13 and 0.30 mg % in group C and 0.11 and 0.21 mg %

Table:- 9. Mean ± S.E. and CV % of Total leucocyte count (10^3/cu mm) in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	8.83±0.38	10.52	8.96±0.37	10.04	9.60±0.67	17.02	9.67±0.75	18.94	9.38±0.75	19.56	9.19±0.69	16.78	9.00±0.50	17.49	9.00±0.44	12.08
B	8.99±0.28	7.63	9.03±0.23	6.21	9.61±0.59	14.22	9.66±0.56	14.23	9.58±0.57	14.45	9.48±0.42	10.95	8.85±0.45	12.44	8.76±0.31	8.79
C	8.80±0.24	6.81	8.91±0.29	7.88	9.45±0.53	13.69	9.46±0.57	14.70	9.23±0.26	6.90	9.19±0.22	5.97	9.05±0.30	7.98	8.91±0.30	8.25
D	8.84±0.32	8.84	8.90±0.40	11.00	9.40±0.33	8.60	9.45±0.40	10.42	9.00±0.25	6.71	8.99±0.23	6.24	8.87±0.39	10.77	8.87±0.41	11.32

No.of observation in each group, (n) = 6.

Table:-10. Analysis of variance for the effect of suture material used in urethrotomy on TLC (10^3/cu mm) in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.385986	0.4034166 NS
Between days	7	1.9078451	1.994 NS
Error	181	0.9567924	

NS :- Non- significant.

Table:- 11. Mean ± S.E. and CV % of angles (angle = arcsin $\sqrt{\text{percentage}}$) corresponding to neutrophil percent in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	37.15±0.88 (36.35)	5.81	37.84±1.07 (37.45)	6.91	38.59±0.92 (38.68)	5.82	39.03±0.78 (39.56)	4.87	38.01±0.89 (37.48)	5.76	37.84±0.99 (37.48)	6.39	37.64±0.99 (37.15)	6.43	37.35±0.94 (36.66)	6.16
B	37.25±0.89 (36.52)	5.85	37.65±0.94 (37.14)	6.13	38.63±0.94 (38.84)	5.94	39.22±0.89 (39.86)	5.58	38.14±0.92 (38.17)	5.89	37.85±0.73 (37.56)	4.76	37.56±0.68 (37.07)	4.44	36.95±1.03 (35.96)	6.85
C	37.04±1.11 (36.10)	7.34	37.54±1.06 (36.95)	6.90	38.54±0.78 (38.72)	4.98	38.54±0.67 (38.75)	4.28	37.45±0.87 (37.70)	5.69	37.35±0.82 (36.70)	5.38	37.05±0.91 (36.17)	6.01	37.25±0.87 (36.52)	5.69
D	36.94±1.06 (35.95)	7.01	37.36±0.78 (36.56)	5.10	38.62±1.15 (38.74)	7.27	38.82±1.15 (39.09)	7.27	38.14±0.97 (37.99)	6.22	37.24±1.02 (36.46)	6.72	37.34±1.16 (36.57)	7.63	36.95±0.95 (35.99)	6.29

Figures in Parenthesis indicating geometric mean obtained from obsolute percent.
No.of observation in each group, (n) = 6 .

Table:-12. Analysis of variance for the effect of suture material used in urethrotomy on neutrophil percent in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	1.89	0.390 NS
Between days	7	9.1871	1.910 NS
Error	181	4.8086	

NS :- Non- significant.

Table:- 13. Mean ± S.E. and CV % of angles (angle = arcsin $\sqrt{\text{percentage}}$) corresponding to lymphocyte percent in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	48.36±0.82 (55.74)	4.16	48.16±0.80 (55.41)	4.06	47.40±0.87 (54.06)	4.51	46.73±1.13 (52.83)	5.94	47.59±1.07 (54.35)	5.52	47.50±1.06 (54.18)	5.48	47.88±0.99 (54.86)	5.66	47.97±0.83 (55.07)	4.26
B	48.45±0.83 (55.90)	4.19	48.45±0.91 (55.89)	4.58	47.59±1.03 (54.36)	5.30	47.39±1.06 (54.02)	5.47	47.67±1.14 (54.02)	5.86	47.79±1.10 (54.67)	5.64	48.07±1.17 (55.15)	5.97	48.17±1.10 (55.33)	5.60
C	48.64±0.90 (56.22)	4.54	48.55±0.89 (56.06)	4.47	47.88±1.05 (54.85)	5.38	47.59±1.00 (54.37)	5.13	47.97±0.92 (55.05)	4.71	48.26±0.95 (55.54)	4.82	48.26±1.03 (55.52)	5.22	48.45±0.81 (55.92)	4.07
D	48.45±0.88 (55.89)	4.46	48.65±0.85 (56.23)	4.29	47.78±0.99 (54.70)	5.09	47.59±1.08 (54.34)	5.57	47.89±1.06 (55.00)	5.42	48.17±1.15 (55.32)	5.87	48.56±1.16 (55.99)	5.85	48.46±1.03 (56.00)	5.19

Figures in Parenthesis indicating geometric mean obtained from obsolute percent.
No.of observation in each group, (n) = 6.

Table:-14. Analysis of variance for the effect of suture material used in urethrotomy on lymphocyte percent in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	2.7501	0.5256 NS
Between days	7	3.9696	0.7586 NS
Error	181	5.2326	

NS :- Non- significant.

Table:- 15. Mean ± S.E. and CV % of angles (angle = arcsin $\sqrt{\text{percentage}}$) corresponding to monocyte percent in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	9.23 ± 1.12 (2.38)	29.77	9.11 ± 1.05 (2.33)	28.24	8.81 ± 1.05 (2.18)	29.07	8.62 ± 1.08 (2.08)	30.78	8.99 ± 1.16 (2.25)	31.17	9.20 ± 0.80 (2.47)	21.47	9.29 ± 0.57 (2.56)	15.11	9.48 ± 0.52 (2.67)	13.58
B	9.00 ± 1.02 (2.29)	27.84	9.14 ± 1.06 (2.35)	28.29	8.86 ± 0.94 (2.24)	25.99	8.51 ± 1.00 (2.04)	29.04	8.99 ± 1.05 (2.29)	28.75	9.16 ± 0.67 (2.47)	18.04	9.29 ± 0.74 (2.52)	19.50	9.11 ± 0.80 (2.39)	21.73
C	9.01 ± 0.96 (2.30)	26.25	9.07 ± 1.01 (2.33)	27.24	8.92 ± 0.95 (2.27)	26.16	8.50 ± 0.95 (2.04)	27.49	9.15 ± 0.93 (2.40)	24.88	9.25 ± 0.67 (2.52)	17.77	9.20 ± 0.54 (2.52)	14.44	9.52 ± 0.50 (2.69)	12.90
D	9.23 ± 1.12 (2.37)	29.77	9.15 ± 0.98 (2.37)	26.63	8.91 ± 0.83 (2.29)	22.94	8.53 ± 0.96 (2.06)	27.65	9.26 ± 0.74 (2.51)	19.51	9.13 ± 0.67 (2.45)	18.07	9.27 ± 0.50 (2.56)	13.32	9.21 ± 0.63 (2.49)	16.65

Figures in Parenthesis indicating geometric mean obtained from obsolute percent.
No. of observation in each group, (n) = 6.

Table:-16. Analysis of variance for the effect of suture material used in urethrotomy on monocyte percent in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.07256	0.0174 NS
Between days	7	1.51744	0.3630 NS
Error	181	4.1798	

NS :- Non- significant.

Table:- 17. Mean ± S.E. and CV % of angles (angle = arcsin $\sqrt{\text{percentage}}$) corresponding to eosinophil percent in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	8.21±0.94 (1.91)	28.00	7.50±0.94 (1.59)	30.64	7.33±1.18 (1.47)	39.51	7.73±1.14 (1.65)	36.19	8.21±0.94 (1.91)	28.00	8.30±1.31 (1.85)	38.67	8.04 (1.76)	36.66	8.44±1.11 (1.78)	32.32
B	8.47±1.06 (2.00)	30.78	8.07±1.16 (1.78)	35.21	7.50±0.94 (1.58)	30.64	7.34±1.18 (1.47)	39.49	7.73±1.14 (1.65)	36.19	8.04±1.20 (1.76)	36.67	8.30±1.31 (1.85)	38.68	8.21±0.94 (1.91)	28.00
C	8.78±1.09 (2.14)	30.39	8.21±0.94 (1.82)		7.90±0.87 (1.78)	26.99	7.33±1.18 (1.47)	39.51	8.13±1.07 (1.84)	32.24	8.21±0.86 (1.91)	28.00	8.04±1.20 (1.76)	36.67	8.84±0.98 (2.22)	27.28
D	8.21±0.94 (1.91)	28.00	7.81±1.03 (1.70)	32.18	7.64±0.67 (1.70)	21.44	7.50±0.94 (1.58)	30.64	7.73±1.14 (1.64)	36.17	7.90±0.87 (1.78)	26.99	8.47±1.06 (2.00)	30.78	8.13±1.02 (1.85)	32.25

Figures in Parenthesis indicating geometric mean obtained from obsolute percent.
No. of observation in each group, (n) = 6.

Table:-18. Analysis of variance for the effect of suture material used in urethrotomy on eosinophil percent in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.648	0.107 NS
Between days	7	2.923	0.485 NS
Error	181	6.029	

NS :- Non- significant.

Table:- 19. Mean ± S.E. and CV % of haemoglobin (mg %) in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	10.84±0.35	7.87	10.66±0.38	8.75	10.68±0.38	8.74	10.55±0.33	7.47	10.65±0.33	7.60	10.68±0.32	7.30	10.65±0.36	8.26	10.64±0.38	8.74
B	10.76±0.36	8.09	10.57±0.39	9.08	10.69±0.38	8.60	10.66±0.44	10.10	10.68±0.44	10.00	10.68±0.39	8.95	10.63±0.41	9.45	10.66±0.42	9.69
C	10.88±0.37	8.25	10.60±0.38	8.77	10.68±0.38	8.73	10.68±0.40	9.10	10.65±0.42	9.72	10.70±0.43	9.86	10.65±0.43	9.83	10.65±0.42	9.59
D	10.74±0.41	9.36	10.60±0.41	9.43	10.38±0.42	9.64	10.68±0.41	9.46	10.63±0.42	9.75	10.58±0.43	9.85	10.6±0.42	9.60	10.6±0.46	10.52

No.of observation in each group (n) = 6 .

Table:-20. Analysis of variance for the effect of suture material used in urethrotomy on Haemoglobin (gm %) in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.0123513	0.0141 NS
Between days	7	0.0452857	0.0518 NS
Error	181	0.8748729	

NS :- Non- significant.

Table:- 21. Mean ± S.E. and CV % of blood urea nitrogen (mg/100 ml) in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	21.33±1.51	17.39	21.60±1.44	16.33	22.73±1.39	14.97	23.43±1.31	13.70	22.13±1.12	12.37	21.50±1.06	12.02	21.33±1.34	15.33	21.40±1.58	18.10
B	21.33±1.20	13.78	21.97±1.43	15.94	22.82±1.53	16.42	22.05±1.49	16.60	21.95±1.73	19.31	21.87±1.33	14.89	21.28±1.65	18.98	22.08±1.35	20.87
C	21.45±1.59	18.13	22.17±1.83	20.20	22.77±2.14	23.02	23.18±1.89	19.99	22.68±1.82	19.66	22.43±1.62	17.66	22.33±1.65	18.10	20.38±1.74	14.94
D	21.48±1.65	18.85	22.10±1.65	18.28	23.26±1.86	19.59	23.43±1.99	20.77	22.95±1.57	16.76	22.37±1.59	17.47	22.03±1.99	22.08	22.08±1.67	18.52

No.of observation in each group (n) = 6.

Table:-22. Analysis of variance for the effect of suture material used in urethrotomy on blood urea nitrogen (mg/100ml) in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	5.741472	0.4218 NS
Between days	7	9.2037143	0.6762 NS
Error	181	13.61082	

NS :- Non- significant.

Table:- 23. Mean ± S.E. and CV % of Serum creatinine (mg/100 ml) in male buffalo calves in consequence of urethrotomy wound healing.

Groups	Period (Days)															
	0		1		2		3		7		14		21		28	
	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %	Mean ± S.E.	CV %
A	2.13±0.19	21.51	2.15±0.18	20.95	2.30±0.20	22.30	2.36±0.26	27.21	2.33±0.21	22.32	2.23±0.23	25.75	2.21±0.21	23.12	2.15±0.22	24.92
B	2.17±0.23	21.32	2.23±0.20	21.72	2.48±0.19	19.15	2.55±0.19	18.35	2.37±0.19	20.17	2.25±0.18	19.42	2.23±0.20	21.72	2.15±0.22	20.96
C	2.20±0.19	25.57	2.20±0.23	25.71	2.33±0.23	24.53	2.50±0.26	25.04	2.35±0.25	26.20	2.33±0.25	26.27	2.25±0.24	26.18	2.20±1.90	21.32
D	2.17±0.24	27.65	2.18±0.25	28.34	2.28±0.25	26.67	2.38±0.25	25.40	2.33±0.25	24.58	2.30±0.24	25.05	2.23±0.20	21.72	2.22±0.21	23.04

No.of observation in each group (n) = 6.

Table:-24. Analysis of variance for the effect of suture material used in urethrotomy on Serum creatinine (mg/100 ml) in buffalo calves.

Source of variation	Degree of freedom	MS	F
Between groups	3	0.0511	0.1967 NS
Between days	7	0.2405	0.9257 NS
Error	181	0.2598	

NS :- Non- significant.

Table:-25. Mean ± S.E. and CV % of urethral diameter (mm) in buffalo calve after urethrotomy at the 45th post-operative day.

Urethral diameter	Group-A Mean ± S.E	Group-A CV %	Group-B Mean ± S.E	Group-B CV %	Group-C Mean ± S.E	Group-C CV %	Group-D Mean ± S.E	Group-D CV %
Anterior to operation site (towards glans penis)	1.24 ± 0.16	30.75	1.28 ± 0.18	34.82	1.29 ± 0.17	32.74	1.27 ± 0.16	30.36
At the operation site	1.35 ± 0.20	35.68	1.26 ± 0.17	33.62	1.28 ± 0.18	33.70	1.24 ± 0.17	33.00
posterior ot operation site (towards ischial arch)	1.29 ± 0.18	33.56	1.31 ± 0.16	30.09	1.27 ± 0.17	33.65	1.28 ± 0.15	28.90

in group D, respectively as compared to the mean values estimated on zero day. However, the differences were shown to be statistically non-significant (Table 24).

Micrometry of urethral lumen :

The mean ± S.E. and CV% of lumenal diameter of urethral section of operated part of urethra and about 1 cm. anterior & posterior to operated site were presented in table-25. In group A lumenal diameter of urethra at the site of operation was 1.35 ± 0.20 mm, where as 1.24 ± 0.16 mm and 1.29 ± 0.18 mm at the anterior to the operation site and posterior to the operation site, respectively. The luminal diameter at the site of operation was 1.26 ± 0.17 mm. whereas, 1.24 ± 0.16 mm and 1.29 ± 0.18 mm. at anterior and posterior to the operation site, respectively in group B. In the animals of group C urethral diameter was 1.29 ± 0.17 mm at the anterior to operation site and 1.27 ± 0.17 mm at posterior to operation site. Where it was 1.28 ± 0.18 mm at the operation site. In group D the urethral diameter at operation site was 1.24 ± 0.17 mm where as 1.27 ± 0.16 mm and 1.28 ± 0.15 mm respectively anterior and posterior to the operation site. However, Micrometry of urethral tissue section of the animals of group A, B, C and D did not reveal any significant increase or decrease in the diameter at urethrotomy site when compared to posterior and anterior urethral tissue sections.

Macroscopic Observation :

Group A : The line of incision united very well. The moderate adhesion of penis with adjoining tissue was noticed. Palpable scar was moderate. The urethrotomy site was faintly visible though it healed completely. No pus pocket or necrotic tissue was seen at the operative site in any of the experimental animal.

Group B : The line of incision appeared to be united well except one animal, where leakage was observed. The moderate adhesions of penis with adjoining tissue were noticed in one case. Palpable scars were observed. No pus pocket or necrotic tissue was seen at the operative site in any animal of this group.

Group C : The wound edges were well apposed and suture line could not be visualised. The adhesion of penis with adjoining tissue were not observed in any animal. The palpable scar could not be located at all. No pus pocket or necrotic tissue was seen at operative site in any of the experimental animal.

Group D : The wound edges united very well. There was no covering over the suture. The adhesion of penis with adjoining tissue was not observed in any of experimental animals. A very small area of palpable sear was observed. No pus pocket or necrotic tissue was observed at the operative site in any of experimental animal.

Histopathological observation :

Group – A: Microscopic examination of preoperative portion of urethra (just one centimeter anterior to operated site) showed no stricture in lumen of urethra. Lamina epithelialis of this part was transitional type with minor fold thrown into the lumen (fig. 7). Cytoplasm of epithelialis was vacuolated and their nuclear chromatin was euchromatic. The tunica propria-submucosa was made up of fibromusculoelastic tissue with normal corpus cavernosum urethrae, where as corpus cavernosum penis was much denser with less amount of cavernous spaces. There was no inflammatory reaction observed. The lamina epithelialis was negative for neutral and acid mucopolysaccharides, whereas, propria-submucosa had moderate reaction for neutral mucopolysaccharide and acidic mucopolysaccharide.

Group B : The line of incision appeared to be united well except one animal, where leakage was observed. The moderate adhesions of penis with adjoining tissue were noticed in one case. Palpable scars were observed. No pus pocket or necrotic tissue was seen at the operative site in any animal of this group.

Group C : The wound edges were well apposed and suture line could not be visualised. The adhesion of penis with adjoining tissue were not observed in any animal. The palpable scar could not be located at all. No pus pocket or necrotic tissue was seen at operative site in any of the experimental animal.

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The H & E section of operated part of urethra on 45th day of experiment showed urethral lumen slightly diminished (fig. 8). Lamina epithelialis showed hyperplasia and desquamation. Epithelial cytoplasm was vacuolated with pale vesicular nucleus. Fibroblastic changes were observed at the propria-submucosa at operated site. The epithelial necrosis was also observed at certain point. However, no inflammatory changes were encountered (fig. 9). Mild reaction of neutral mucopolysaccharide was seen at the luminal epithelium, where as propria-submucosa reacted moderately with acidic mucopolysaccharide (fig.10).

Group B : Histomorphological examination of urethra collected from 1 cm. anterior to the operation site showed no stricture in lumen of urethra (fig.11). Luminal epithelium was also transitional type. The corpus cavernous urethral space around lumen appeared normal (fig.12).

Slight hyperplastic reaction was observed in the luminal epithelium. In the operated area the superficial cells of epithelium showed necrosis, but basal cells still appeared normal (fig.13). The lumen of operated part of urethra was near to normal as compared to anterior to the operative part of urethral lumen. There was mild cellular reaction seen at the site of operation in the propria submucosa. Suture materials appeared to be absorbed but zone of reaction was represented by polymorphonuclear leukocyte, macrophages and immature fibroblasts with predominating the mature fibroblasts (fig.14). The fibroelastic tissue of corpus cavernous urethral space showed absorption and remodelling. The fibroblastic activity showed obliteration of cavernous space. Histochemically the neutral mucopolysaccharide content in epithelium and propria-submucosa was low, but acidic mucopolysaccharide content was moderate (fig.15).

Group C :In this group, where urethral wound was sutured with polyglactin - 910, histomarphological examination of urethra anterior to operated part showed no stricture in the lumen of urethra (fig.16). The most area of epithelial lining was normal in architecture. The corpous cavernous space around the lumen was normal (fig.17). In the epithelium, there was absence of neutral mucopolysacchride, where as of propria-submucosa had moderate content of neutralmucopolysaccharide and acidic mucopolysaccharide.

The urethral lumen at operated site did not show any stricture. Epithelium of lumen appeared to be normal (fig.18). Mild fibroblastic activity was observed in the tunica propria-ubmucosa. (fig.19). The cavernous sinus appeared to be normal except at certain locations. Slight cellular reaction was observed at the site of operation in properia submucosa, characterised by very few polymorphonuclear leukocytes, plasma cells, lymphocytes and macrophages. Neutral mucopolysaccharide content was high at suture site but adjoining firoblastic tissue reacted more intensely for acidic mucopolysaccharides (fig. 20).

Group D : Histomorphological examination of urethra collected about 1cm. anterior to the operative site showed no stricture in the lumen of urethra (fig.21). The tunica propria submucosa was made up of fibromusculoelastic tissue with developed cavernous space. The lamina epethelialis was transitional type.

The lumen of operated part of urethra was near normal to preoperated part of urethra. (fig. 22). The hyperplastic reaction was observed in the luminal epithelium. The zone of reaction was extended up to the periurethral tissue, where the fragmented suture material was found to be encapsulated by immature and mature fibroblasts, polymorphonuclear leukocytes, plasma

cells and lymphocytes. Fibroblastic activity was less severe as compared to catgut but more than polyglactin-910 (fig. 23). Neutral mucopolysaccharide content was high at sutured site but adjoining fibroblastic tissue showed more acidic mucopolysaccharide (fig. 24).

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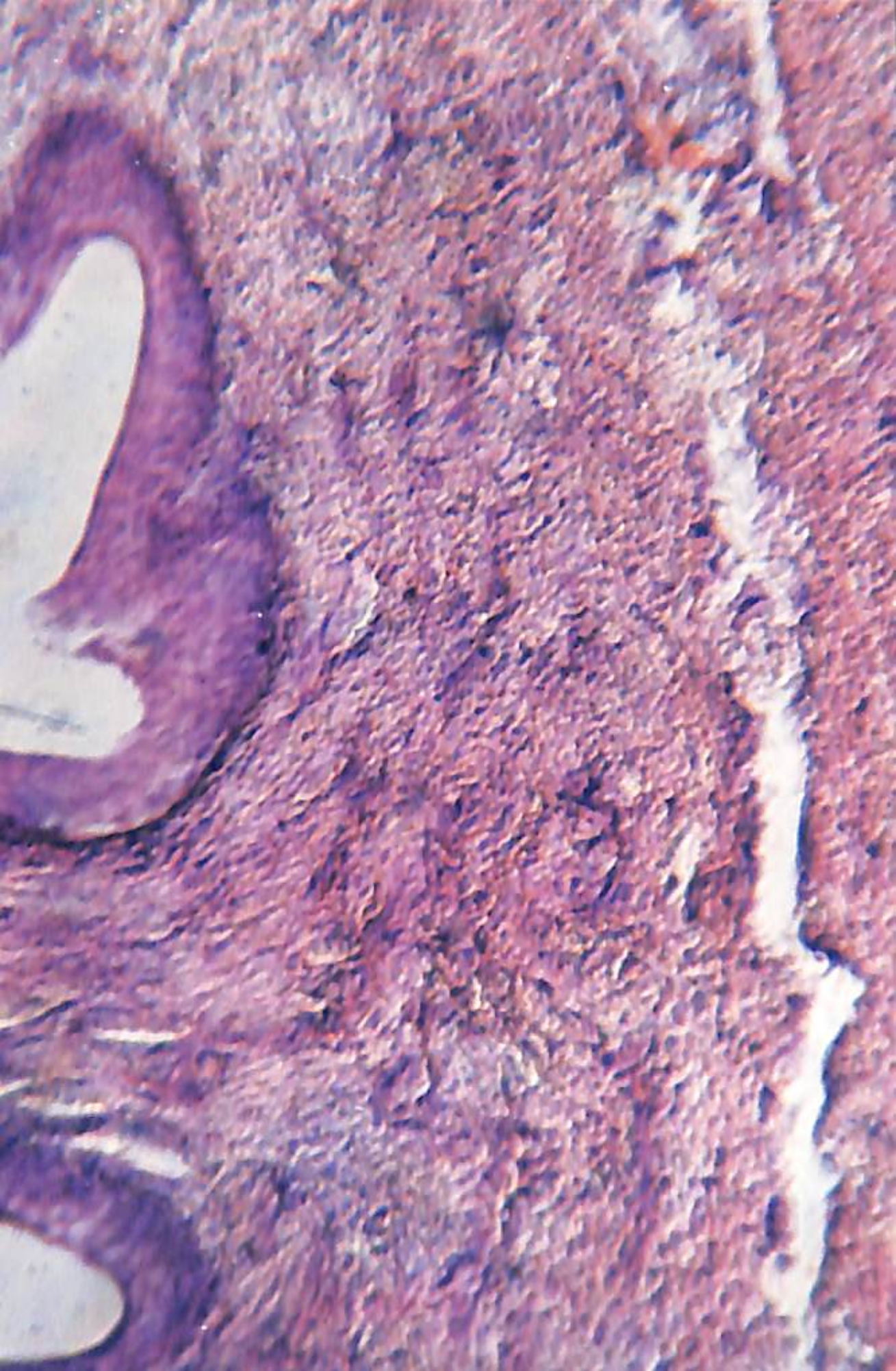

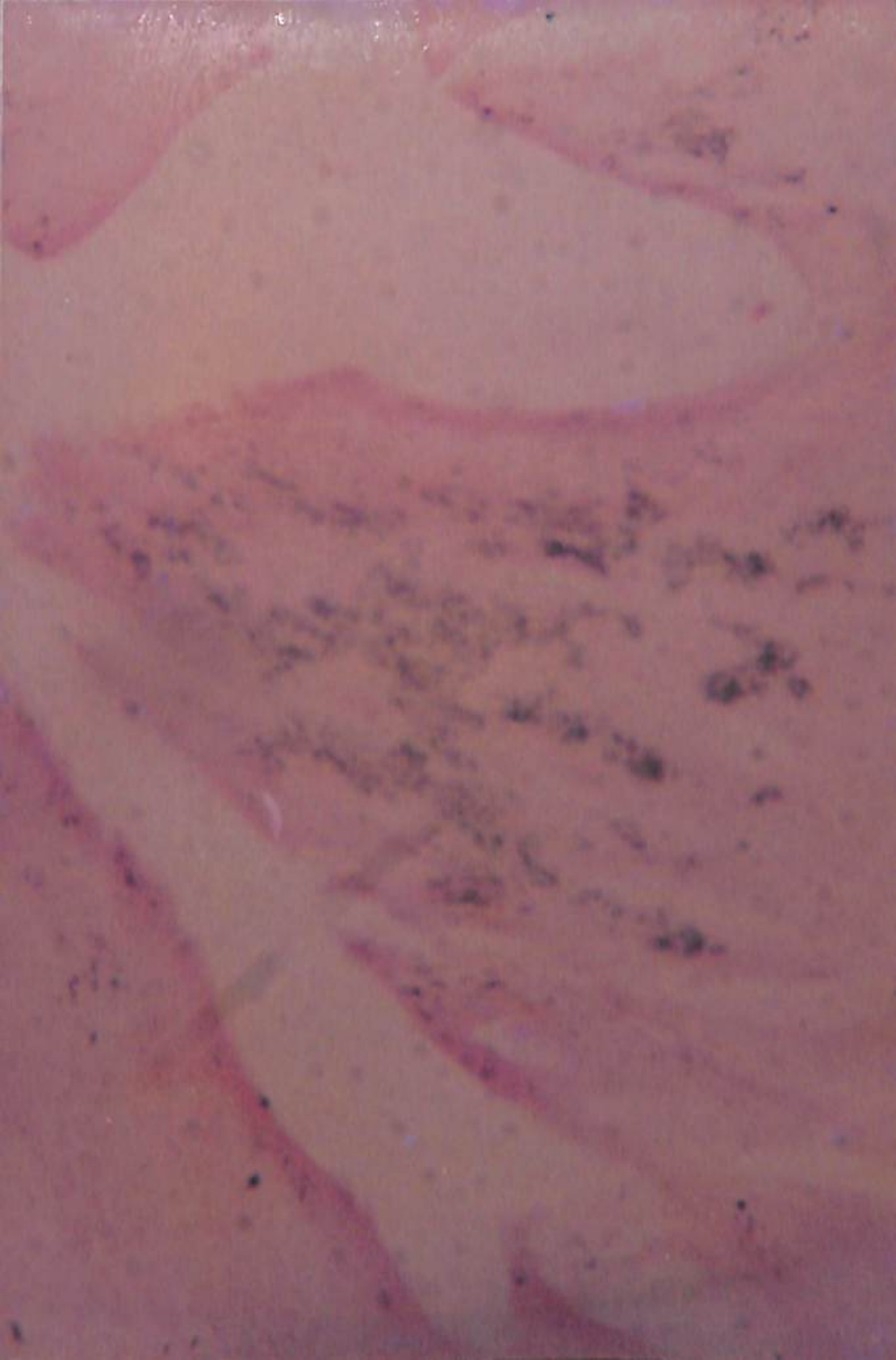

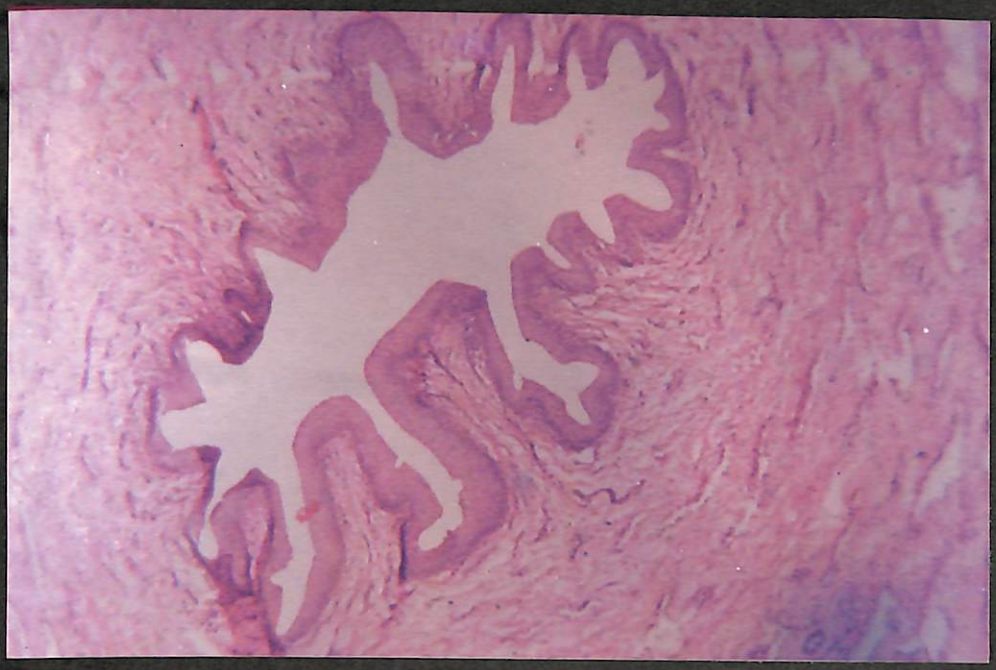

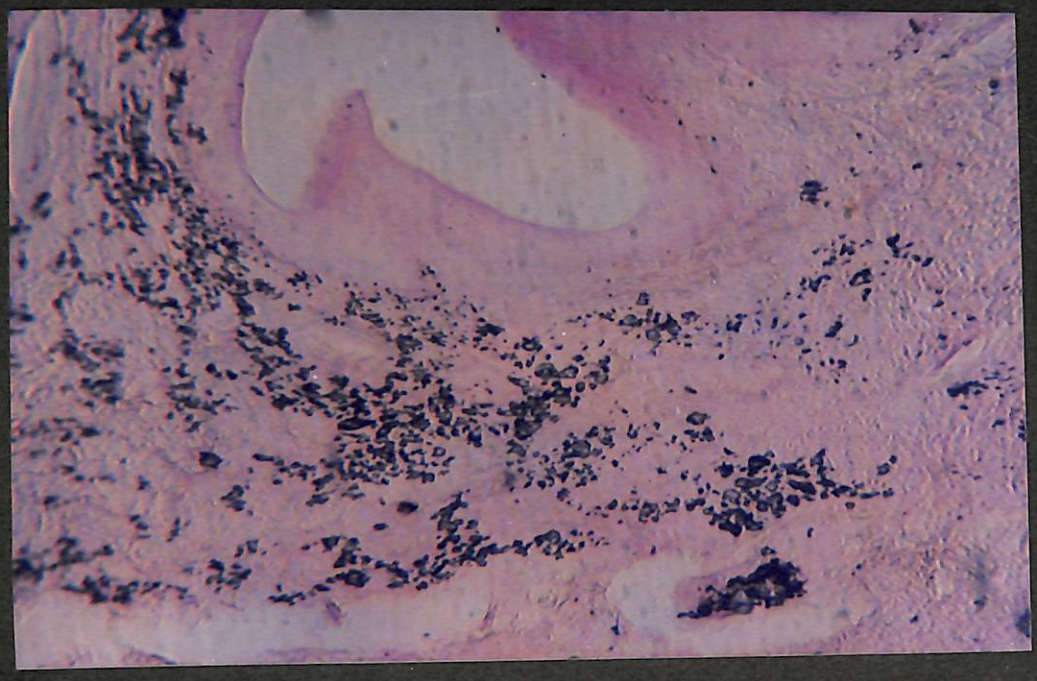

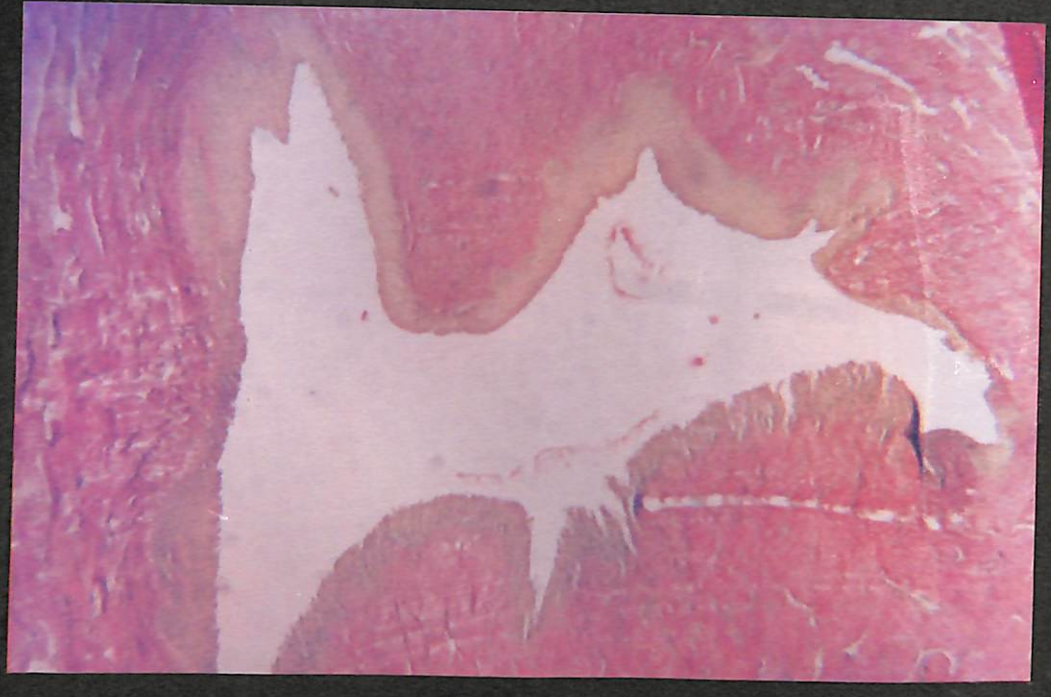

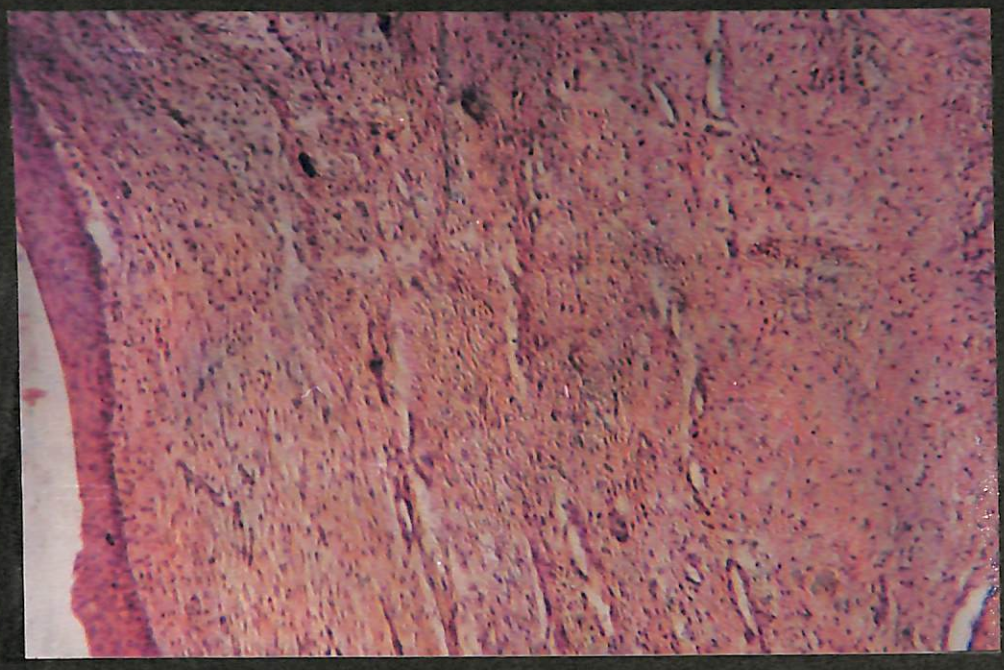

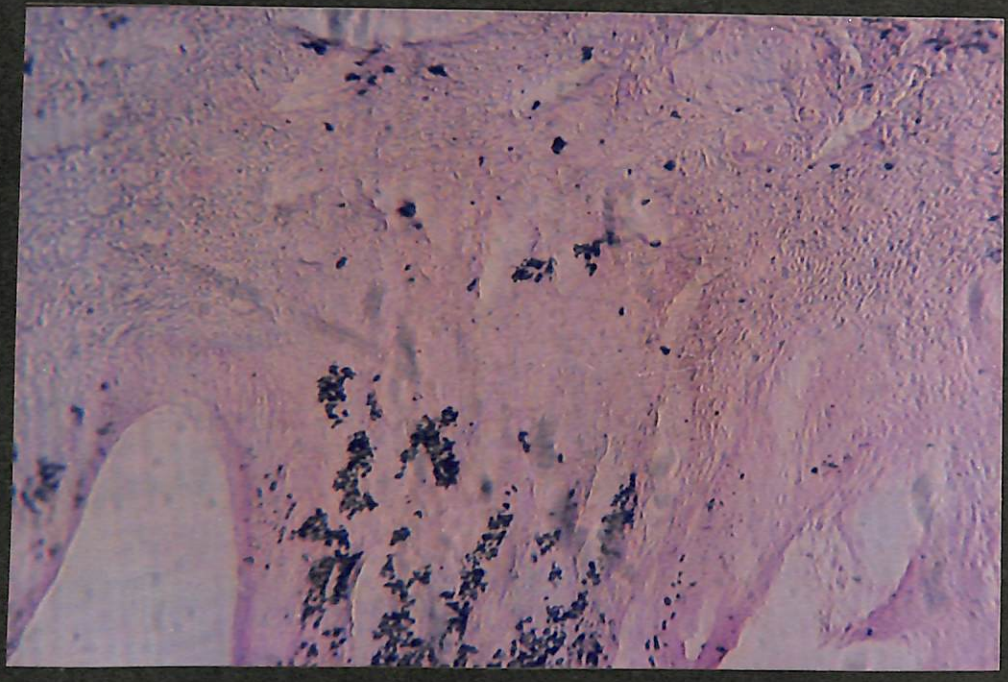

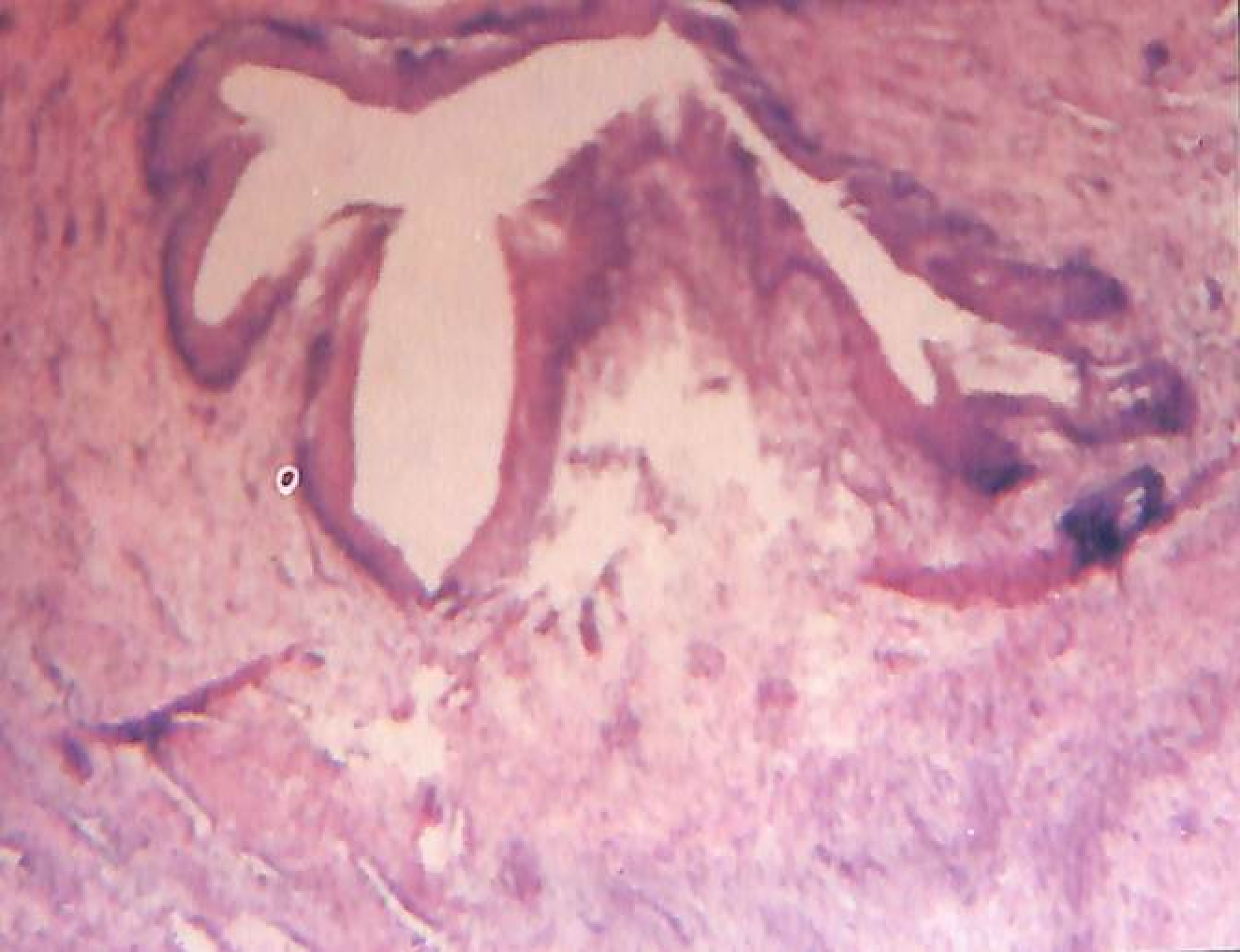

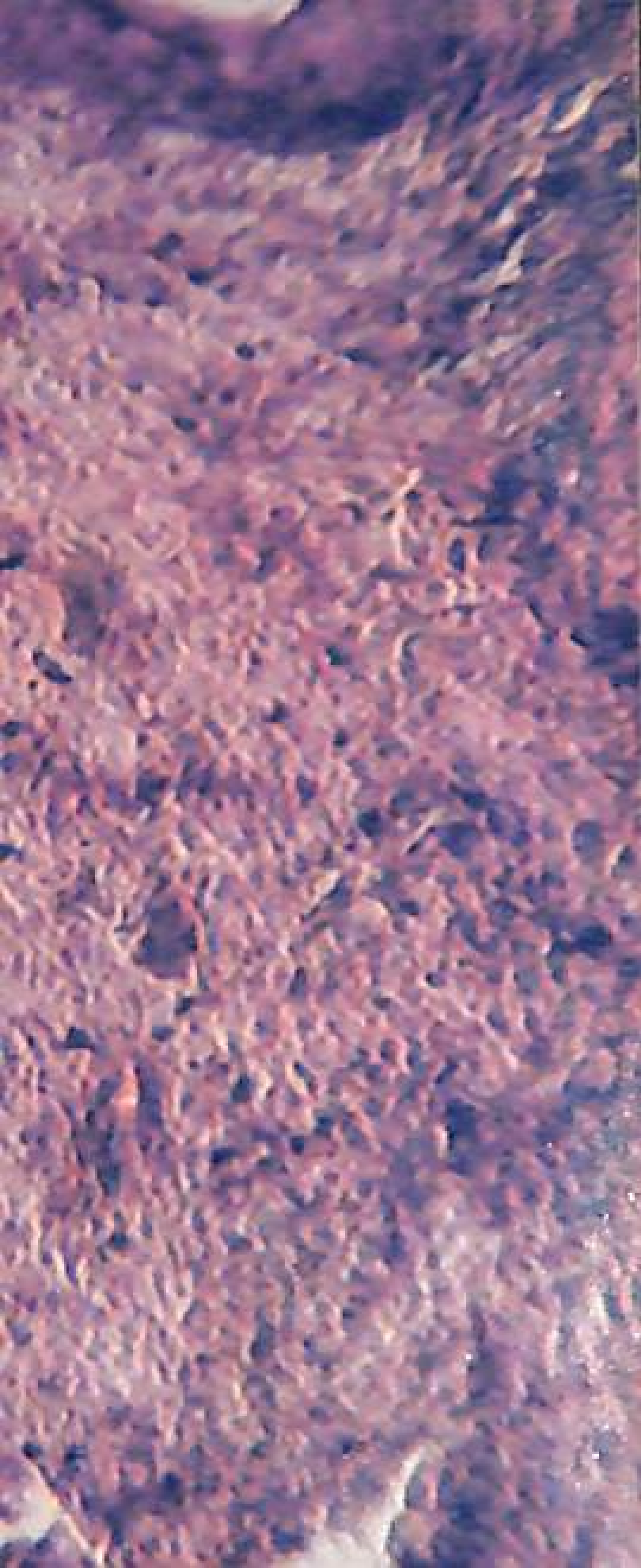

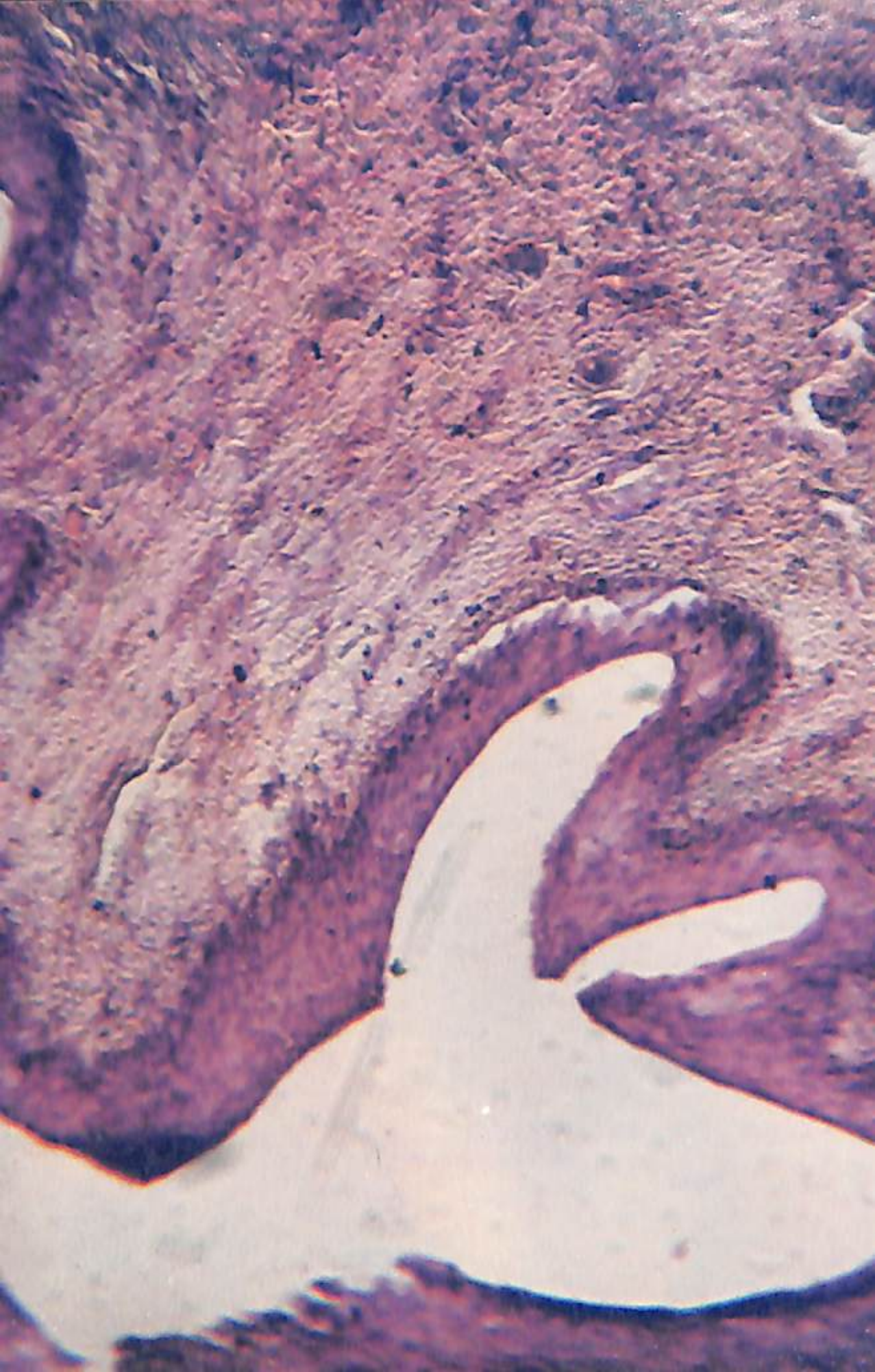

Chapter - 5

DISCUSSION

Discussion

The surgical management of unsutured urethrotomy wound is one of the challenging task because of various complications like subcutaneous infiltration of urine, formation of urinary fistula at the site of operation and development of stricture at the site of incised urethra. There are few reports available where suture material have been employed (absorbable and unabsorbable) for urethrotomy wound closure in bovine (Chatuphale, 1932; Barshikar, 1932-33; Kolhekar, 1960; Vig and Tyagi, 1970; Gera and Nigam 1979; Sharma and Agrawal, 1997 and Prakash *et al.,* 2000). However, these reports are lacking in clinical recommendation. The present study was thus undertaken to assess the feasibility of suturing of urethrotomy incision with different suturing materials both absorbable and nonabsorbable in early healing and management of urethrotomy wounds. There is difference of opinion whether the incised urethra should be sutured or not. It is generally feared that the suturing may leads to stricture formation at the incision site. To prevent subcutaneous infiltration of urine in the dependent area of the abdomen. It is advocated to fix a snugly fitted indwelling polythene catheter in urethra for 10 – 12 days (Prasad *et al.,* 1978). It has been seen clinically that indwelling catheter is not properly fitted to urethral lumen, it can invariably lead to urethral fistulation resulting in subcutaneous infiltration and may cause severe myositis and subsequently necrosis.

The most common site of lodgement of urolith in cattle is an area distal to sigmoid flexure adjacent to the attachment of retractor penis muscles (Gulati *et al.*, 1966; Gera and Nigam, 1979; Singh and Singh 1990; Tyagi and Singh, 1993). Gera and Nigam (1979) reported that post-scrotal urethrotomy to be the most common surgical procedure of choice in

obstructive urolithiasis. Keeping in view the common site of urolith lodgement, post-scrotal urethrotomy was performed and urethral incision was given near the attachment of retractor penis muscles towards ischial arch. The animals were easily controllable in lateral recumbency and local infiltration of 2% lignocaine hydrochloride injection provided satisfactory analgesia.

In present study all the groups of animals had normal general activity like hydration status, mucous membranes of eye, feeding, defecation and urination till the end of experiment. Only two animals of group A and one animal of group B had less feed intake because of severe subcutaneous infiltration of urine and pain. Draining of subcutaneous infiltrated urine resulted in decreased swelling and these animals started taking normal feed on third post operative day. Two animals out of six in group A (Control) had problem of severe urine infiltration on the dependent subcutaneous area and seepage of urine from operative site resulting in gaping of wound after removal of skin sutures on 10^{th} post-operative day. Sharma *et al.* (1983) and Tyagi and Singh (1993) also reported that wound dehiscence may occur due to infection. Though the polythene catheters were selected keeping in view diameter of urethra. These might not be properly fitted to the urethral lumen of these animals. Prakash *et al.* (1999) also observed urine infiltration and gaping of wound in case of catheterization unsutured urethrotomy wound in male calves. This might be due to indwelling catheter not snugly fitted because of in male bovine, the calibre of urethra diminishes from behind forward (Kolhkar, 1960) or pressure of urine flow due to high slope of urethra in the dorsal region resulted into seepage of urine from urethrotomy site (Tyagi and Singh, 1993). The continuous dropping of urine from wound

site resulted in delayed healing of wound and gaping was observed at the time of removal of skin sutures. Timely stabbing of the infiltrated subcutaneous area prevented myositis and cutaneous wound healed by second intention (Sharma *et al.*, 1983). The two animals, which had moderate subcutaneous infiltration of urine after 24 hours of surgery did not show any wound gaping and infiltration of urine completely subsided on 6th post-operative day. Polythene catheter acted as a splint for regeneration of mucosa, where it was not sutured (Weaver and Schulte, 1962). Rest two animals had no post-operative complications. It is clear that snugly fitted indwelling catheter may not allow leakage of urine at urethrotomy site or dependent area (Tyagi and Singh 1993). At the same time it should not be too tight that it may damage the urethral mucosa. O'conner (1950), Anjaria (1969) and Bozrab *et al.* (1983) advocated the unsuturing of urethral incision and skin incision after urethrotomy to avoid the subcutaneous stagnation of urine in bovine and canine, which also resulted in faster healing. But Mohindra *et al.* (1996) observed marked inflammation of urethrotomy site in dog due to subcutaneous infiltration of urine in unsutured urethrotomy wound.

In group B urethral incision was sutured with 3-0 chromic catgut in closely placed interrupted suture. The penile incised sheath and facia were also sutured with chromic catgut. Where as Prakash *et al.* (1999) sutured the urethral mucosa along with cavernous spongiosum and tunica albuginea, but facia left unsutured after performing urethrotomy. Vig and Tyagi (1970) and Gera and Nigam (1979) sutured the urethrotomy incision in three and two tier respectively.

Four animals of this group showed no infiltration and seepage of urine

and wound healed normally. This is in accordance with Vig and Tyagi (1970), Shastry and Rao (1981), Tiwari (1990), Ashturkar (1994), Prakash *et al.* (1999) in male bovine. Waldron *et al.* (1985), Weber *et al.* (1985) and Mohindra *et al.* (1996) observed that suturing of urethral incision may obviate urine leakage from surgical site into subcutaneous tissue in canine. They also observed haematuria as post-operative complication in unsutured urethrotomy wound in dog. This might be due to bleeding from corpus spongiosum, which was left unsutured. Haemorrhage after urination, excitement or sexual arousal has been reported as a frequent compliction of allowing the urethral incision to heal by second intention in dogs (Everingham *et al.*, 1973; Brown, 1975 and Winghfield and Rawling, 1979). Suturing of urethral incision may prevents bleeding from corpus spongiosum (Brown, 1975; Stone, 1984 and Mohindra *et al.* 1996). Two animals of this group showed seepage of urine from operation site though in very small amount. Although wound healed and skin sutures were removed on 10^{th} post operative day, very small pin head fistula persisted till the end of the experiment. Vig and Tyagi (1970) reported leakage of urine in one out of six animals in which urethra was sutured with catgut. Whereas, Prakash *et al.*(1999) observed pin head permanent fistula at the urethral incision which was sutured with chromic catgut. Seepage of urine might be due to loosing of suture or infection at the operation site. But infiltration of urine was not enough to cause wound dehiscenece and sloughing of abdominal muscles.

In group C urethral wound was sutured with 3-0 polyglactin – 910. No post – operative complication was observed and skin sutures were removed on 10^{th} post-operative day. Weber *et al.* (1985) compared healing of non-sutured pre-scrotal urethral incision with that of sutured with 5-0 polyglactin

-910 and 5-0 polydioxanone in dog and observed that non-sutured urethrotomy wound had more post operative complication than sutured ones. There was little difference between incision sutured with polyglactin-910 and polydioxanone. Mohindra *et al.* (1996) and Prakash *et al.* (2000) observed no post-operative complication in urethral incision sutured with polyglactin-910. But Stewart *et al.* (1990) recommended that polydioxanone, Polyglactin-910 and chromic catgut would be equally valuable in bladder surgery because none of these predisposed to infection and these were no difference in their calculogenic potency. But according to Gourley and Vasseur (1985) polyglactin-910 is one of most recommended suture material as it has maximum tensile strength and minimum tissue reaction. The suturing with polyglactin-910 have minimum tissue reaction and might be resulted in complete sealing of urethral incision. This did not allow leakage of urine at operation site resulted in faster healing of the tissue.

In group D urethral incision was sutured with 3-0 black braided silk. All animals of this group showed normal general activity. One out of six animals of this group showed moderate infiltration of urine at the site of operation, which subsided on 5^{th}-6^{th} post-operative days. Rest five animals had no post-operative complication. This is in accordance with Vig and Tyagi (1970), Datt *et al.* (1973), Gera *et al.* (1973), Sharma and Khan (1978), Larson (1996) and Sharma and Agrawal, (1997). This might be due to suturing of urethra may obviate the risk of infiltration of urine and promote early healing. According to Gera *et al.* (1973) repair of catheterized urethra with an atraumatic needle in two layers prevent seepage of urine.

One animal on 16^{th} post operative day in group A, two animal on 10^{th} and 15^{th} post-operative days in group C and one animal on 15^{th} post

operative day in group D dropped the polythene catheter, respectively. This might be due to accidental stampede and pulling of protruding part of catheter by animal itself or by other animals. In spite of removal of polythene catheter no complication was observed in voiding the urine in these animals.

The retention of catheter for a period of three weeks was found essential for protection against physiological stress of urination. Regeneration of entire urethra including corpus spongiosum was possible within three to four weeks. However, the mucosa reformed within seven days (Weaver and Schulte, 1962). Noordsy and Trotter (1963) and Hastings (1965) retained indwelled rubber catheter in steers for a period of three to four weeks. Whereas, Prasad *et al.* (1978) advocated to fix a snugly fitted polythene catheter in urethra for 10-12 days may prevent subcutaneous infiltration of urine in the dependent area of abdomen.

Sulphadiazine and trimethoprim combination being maximally secreted through urinary system (Jones, 1977) was administered prophylactically to prevent post-operative and ascending urinary tract infection. Regular dressing with povidone iodine minimised external wound contamination.

It is explicit that the clinical manifestation of the increase in body temperature, respiration rate, pulse rate, heart rate on 2^{nd} and 3^{rd} post operative days as compared to zero day were almost similar in all the four groups and statistically non-significant. The alteration in the body temperature, pulse rate, respiration rate and heart rate were only marginal. These results were in accordance with that of Samanta (1971), Sharma and Khan (1978), Lavina and Angelo (1987), Prakash *et al.* (1999) and Prakash

et al. (2000). The marginal changes in these parameters might be due to the surgical stress. This was suggested that chromic catgut, polyglactin – 910 and black braided silk did not behave differently in urethrotomy wound healing. From 7th day onwards uniformity of body temperature, respiration rate, pulse rate and heart rate was indicative of normal physiological status of animals.

The increase in total leucocytes count was observed on 2nd and 3rd post operative days in each group as compared to zero day and then started to decline from 7th day onwards. However, this increase was statistically non-significant. This is in accordance to Samanta (1971), Mohindra (1993), Prakash *et al.* (1999) and Prakash *et al.* (2000). The increase in total number of leucocytes count might be due to concurrent inflammatory process and surgical stress (Willard *et al.*, 1989).

Increase in neutrophil count and decrease in lymphocyte count was observed on 2nd and 3rd day after performing urethrotomy as compared to zero day in all the groups. However, this increase in neutrophils and decrease in lymphocyte count is statistically non- significant. This is in accordance to Samanta (1971), Lavina and Angelo (1987) and Mohindra (1993). The non-significant neutrophilia and lymphopenia is indicative of concurrent inflammatory process and stress (Willard *et al.*, 1989). From 7th day onwards uniformity of total leucocyte count, neutrophil percent and lymphocyte percent showed a progress towards normalcy.

There was non-significant change of eosinophil count and monocyte count was observed in any group of animals, which indicative of normal physiological status of animal. This is in accordance with Levina and Angelo (1987), Mohindra (1993) and Prakash *et al.* (1999) and Prakash *et al.* (2000).

There was non- significant changes in haemoglobin percent in all the groups of animals which is in accordance with Samanta (1971), Levina and Angelo (1987), Mohindra. (1993), Prakash *et al.* (1999) and Prakash *et al.* (2000). However, Gera and Nigam (1980), Jadon *et al.* (1987) and Gangwar *et al.* (1990) observed gradual increasing trend after urethral obstruction in experimental bovines. Kumar *et al.* (1991) observed increased haemoglobin percent in obstructive urolithiasis in bullocks which declined following operation for cure. This increase in haemoglobin percent might be due to dehydration. But the present experimental investigation does not come under this clinical situation. In the present experimental endevour free flow of urine was provided by indwelling polythene catheter.

Blood urea nitrogen (BUN) and serum creatinine observed to be almost near normal in all the four groups. There was slight rise in the level on 2^{nd} and 3^{rd} post-operative days as compared to zero day which was within normal range and statistically non-significant. This is in accordance to Rajamani and Ganpaty (1965), Samanta (1971), Sharma (1973) and Sharma and Khan (1978). However, Pandey *et al.* (1986), Barua, *et al.* (1990) and Kumar *et al.* (1991) observed increased level of BUN and serum creatinine in clinical cases of obstructive urolithiasis in bovine, which decreased post-operatively. This increase in BUN and serum creatinine might be due to decreased glomerular filteration rate. But in the present experimental endeavour excess urine was not allowed to be accumulated in urinary bladder and free flow of urine was provided by indwelling polythene catheter. Subcutaneous infiltrated urine was also drained out by timely stabbing.

During post-operative period, clinical observations and BUN and

serum creatinine level evidenced that there was no obstruction to the clearance of urine through indwelling polythene catheter in any group of animals. This also suggested that chromic catgut, polyglactin-910 and black braided silk did not behave differently in urethrotomy wound healing as per biochemical parameters.

Micrometric results of group A revealed slight increase in luminal diameter at operation site as compared to tissue section at about 1 cm. anterior and posterior to the operated site. However, increased was statistically non-significant. Whereas, in group B, C and D slight constriction were observed in urethral lumen of operation site as compared to anterior and posterior portion of urethra. But this constriction was also non-significant. Weber *et al.* (1985) also measured the urethral circumference in dogs at three locations (surgical site, 1 cm. cranial and 1 cm. caudal to surgical site) and did not observe any post-operative urethral stricture in any dog in which urethra was either left unsutured or sutured with polyglactin-910 or polydioxanone. Whereas, Mohindra *et al.* (1996), Prakash *et al.* (1998), Prakash *et al.* (1999) and Prakash *et al.* (2000) observed increase in luminal diameter of urethra at operation site in case of catheterised unsutured urethrotomy wound which might be due to accumulation of granulation tissues at the site of urethrotomy. Weaver and Schult (1962) also reported that the traumatized canine urethra could refrom over a splinting catheter without stricture formation, when mucosal continuity was present. Waldron *et al.* (1985), Mohindra *et al.* (1996), Prakash *et al.* (1998), Prakash *et al.* (1999) and Prakash *et al.* (2000) did not observe stricture in the lumen of urethra in sutured urethrotomy wound. Stricture formation as a post operative complication after closure of incised urethra was reported by

Metcalf (1965), Brown (1975), Bozrab *et al.* (1983) and Stone (1984).

In the present experiment no appreciable stricture formation was encountered. This might be due to use of indwelling catheter. Suturing of longitudinal urethral incision transversely minimize the possibility of stricture formation (Motta and Walmsley, 1954). In a clinical instance, urethral mucosa damaged due to calculi may increase the chances of stricture formation after surgical procedure (Weber *et al.*, 1985).

Macroscopically moderate adhesion of penis with adjoining tissue was observed in group A animals and only one animal in group B at the time of collection of tissues on 45th post-operative day. The degree of fibrous adhesions were comperatively negligible in group C and D. However, urethrotomy site was very faintly visible and difficult to recognised in all the animals of four groups. It might be due to trauma, inflammation or irritation of surrounding tissue by stiff knot of chromic catgut (Pareira, 1949 and Tyagi and Lumb, 1961). No pus pocket or necrotic tissue was observed that was indicative of normal healing of urethra. No much scar formation was observed in any group of animals. Vig and Tyagi (1970), observed palpable scar and adhesion of penis with adjoining tissue were less marked on 30th day than 10th day in case of catgut sutured urethrotomy wound. They also observed palpable scar and adhesions were less marked in non-absorbable suture material.

At the site of operation in group A (control) animals in which urethral incision was not sutured exhibited marked fibroblastic activity and necrosis of epithelium. The findings are in consonance with the observations of Weber *et al.* (1985), Waldron *et al.* (1985) and Mohindra *et al.* (1996). This might be due to urethral incision healed by second intention. Occurrence of

granulation and fibroblastic tissues at the site of operation is suggestive of normal wound healing. Absence of inflammatory reaction was indicative of normal healing of urethra. However, corpus cavernosum urethrae did not seen to be obliterated suggestive of almost normal periurethral tissue. The urethral lumen in the operated area seemed to be near normal. Although there was extensive infolding of lamina epithelialis particularly at one side of urethra. It acquired flattened shape in group A. However, urethral lumen did not seen to be obliterated rather it was more flattened as compared to normal urethral lumen. In pre-operative portion of urethra, it was almost rounded because of minor folding thrown into the lumen of urethra. In this group, urethral epithelium and propria-submucosa at the site of operation showed similar mild content of neutral mucopolysaccharide content as also observed in pre-operated part of urethra indicative of normal mucosal healing. A moderate acidic mucopolysaccharide in propria submucosa was also comparable with pre-operated portions suggestive of normal healing of periurethral tissue.

Suturing of urethral incision with chromic catgut in group B showed inflammatory cellular reaction at the site of sutures in lamina propria sub-mucosa.. The urethritis was characterized by accumulation of polymorphonuclear leucocytes, plasma cells, lymphocytes and macrophages at the site of operation. It is in accordance with Vig and Tyagi (1970) and Mohindra *et al.* (1996). This might be due to cellular reaction or local infection but it did not seem to be any clinical significance since the wound healed satisfactorily. In this group fibroblastic activity in the lamina propria sub-mucosa caused obliteration of cavernous spaces which showed absorption and remodelling of wound. The epithelium lining of urethral

lumen showed necrosis at certain locations where as basal cells still appeared normal. Other areas of normal urethral lining seemed to have hyperplastic activity. Vig and Tyagi (1970) reported marked proliferation of urethral epithelium sutured with catgut and severe reaction incited by catgut resulted in obliteration of corpus cavernosum urethral.

The zone of reaction in case of urethral incision sutured with catgut in calves was represented by predominance of immature fibroblast at early stages (Lacalio *et al.,* 1943; Pareira, 1949; Tyagi and Lumb, 1961 and Vig and Tyagi, 1970. It was reported that catgut revealed comparatively less hyperplastic changes in epithelium and corpus cavernosum urethae were less obliterated on 30^{th} day of surgery as compared to 10^{th} post operative day (Vig and Tyagi, 1970).

In present study urethral lumen at operative site near normal to anterior tissue section of urethra in group B. Micrometry did not show significant change in lumen of urethra also on 45^{th} post operative day. Prakash *et al.* (1999) observed slight narrowing of urethral lumen in case of urethral incision sutured with 3-0 chronic catgut. A low content of neutral mucopolysacchride in propria submucosa revealed comparatively less metabolic activity as compared to normal urethra (anterior to operation site). However, moderate acidic mucopolysaccharide content indicative of advanced healing of urethral tissues and was comparable to the normal.

In group C mild fibroblastic activity observed at the sutured urethral area with slight inflammatory cellular reaction. The fibroblastic activity and cellular reaction was less than the group B. This might be due to polyglactin -910 elicit minimum tissue reaction (Craig, 1975; Bojrab, 1983; Conn and Beal, 1980). The urethral lumen at the site of operation was not showing any

stricture as compared to anterior part of urethral section. Micrometry of urethral lumen also revealed non-significant change. The sutured site in this group showed better demonstrable neutral mucopolysaccharides and acidic mucopolysaccharides reflecting the advance healing of sutured area. Mohindra *et al.* (1996) observed urethritis on 10^{th} post operative day in dogs in which incision was sutured with chromic catgut and polyglactine – 910 showed accumulation of polymorphonuclear leucocytes, plasma cells, lymphocytes and macrophages in mucosa and submucosa. This might be due to the presence of intraluminal suture.

In group D, urethral incision was sutured with 3 – 0 black braided silk represent, the zone of reaction, which extended upto the periurethral tissue. The fragmented suture material was found to be encapsulated by immature and mature fibroblast, polymorph and lymphocytes. But inflammatory reaction and fibroblastic activity was less severe as compared to chromic catgut. This is in agreement with the findings of Meade and Ochsner (1940); Armistead (1952), Vig and Tyagi (1970) and Verma *et al.*, (1981). The hyperplastic reaction was observed in the luminal epithelium. Vig and Tyagi (1970) observed very mild proliferation of luminal epithelium on 10^{th} post-operative day in case of cotton sutured urethrotomy incision, whereas linen and silk revealed moderate proliferation of the epithelium. They also observed proliferation of epithelium was nearly the same on 10^{th} and 30^{th} post-operative day in case of silk. The lumen of operated part of urethra was slightly diminished. Micrometry of lumen also revealed non-significant constriction of lumen of urethra. High content of neutral mucopolysaccharide at suture site and more acid mucopolysacchride at fibroblastic tissue indicative of advanced healing of sutured area.

Thus, it is concluded that the urethral lumen did not show any appreciable stricture in any of the group of animals till 45th post operative day. The degree of cellular reaction was maximum in group B in which urethral incision was sutured with chromic catgut. Histological and histochemical findings showed satisfactory urethrotomy wound healing in all the groups. But clinical, macroscopic and microscopic observations proved group C to be the best among the four groups studied. However, long term studies are required to confirm the development of stricture in these animals.

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Chapter - 6

SUMMARY & CONCLUSIONS

Summary and Conclusion

The present study was conducted on twenty four clinically healthy male buffalo calves about one year of age. The animals were kept under uniform managemental conditions during the entire period of observation and randomly divided into four groups of six animals each. Snugly fitted polythene catheter was indewelled up to the ischial arch into the urethra in all the animals after performing post scrotal urethrotomy. Different suturing materials were used in this investigation to evaluate their effect on urethral wound healing. Group A served as control in which urethral incision was left unsutured, whereas, in group B, C and D urethral incisions were sutured with 3-0 chromic catgut, 3-0 polyglactin-910 and 3-0 black braided silk respectively in closely placed simple interrupted pattern. Cutaneous wound was sutured by Halsted stich with silk in all animals.

The parameters studied were clinical observations, haematological observations, biochemical observations, micrometry of lumen and gross & microscopic examinations of the urethral tissues. The clinical parameters included rectal temperature, heart rate, respiration rate, general activity feeding, hydration status, colour of mucous membranes, urination, defecation and seepage of urine. Rectal temperature, pulse rate, heart rate and respiration rate were studied on zero (day before operation) 1^{st}, 2^{nd}, 3^{rd}, 7^{th}, 14^{th}, 21^{st} and 28^{th} post-operative days. Other clinical symptoms were observed routinely on upto 45^{th} post-operative day. There was insignificant rise in temperature, pulse rate, heart beat and respiration rate on 2^{nd} and 3^{rd} day after urethrotomy but these clinical features manifested declining trend on 7^{th} day onwards in all the animals. These animals showed normal general activity, hydration status, colour of mucous membrane, defecation and urination throughout the period of observation. Two animals out of six in

group A showed severe urine infiltration on the dependent subcutaneous abdominal area.

The blood samples were collected at different intervals on zero day (day before surgery) 1st, 2nd, 3rd, 7th, 14th, 21st and 28th post-operative days to examine the total leucocyte count, differential leucocyte count and haemoglobin percentage. These parameters did not show any significant variation in any group of animals. Slight rise in TLC and neutrophil count was recorded on 2nd and 3rd post-operative days in most of the animals.

Biochemical examination including estimation of blood urea nitrogen and serum creatinine were carried out at different interval on zero day (day before surgery) 1st, 2nd, 3rd, 7th, 14th 21st and 28th post-operative days. Blood urea nitrogen and serum creatinine evidenced almost near normal in all the groups of animals.

Macroscopically moderate adhesion of penis with adjoining tissue was observed in group A at the time of collection of tissue on 45th post-operative day. The degree of fibrous adhesions were comparatively negligible in group C and group D. Urethrotomy site was very faintly visible in all the animals. Micrometry of tissue sections revealed non-significant variation in urethral diameter in all the animals at the site of urethrotomy when compared with posterior and anterior urethral section.

Microscopically the urethral lumen in the operated area seemed to be near normal in group A animals. The marked fibroblastic activity and necrosis of epithelium observed at the operated site. Urethral epithelium and propria-submucosa showed low content of neutral mucopolysaccharide at the site of operation as seen in normal urethral tissue. Moderate content of acidic mucopolysaccharide in properia-submucosa was also comparable with normal tissue section.

In group B, urethral incision was sutured with catgut incited mild inflammatory cellular reaction at the site of sutures. Fibroblastic activity in propria-submucosa caused obliteration of cavernous spaces which showed absorption and remodelling. The epithelial lining of urethral lumen showed necrosis but basal cells appeared normal. There was mild cellular reaction seen at the site of operation in propria submucosa. Suture material appeared to be absorbed but zone of reaction was represented by polymorphonuclear leucocytes plasma cells lymphocyte and macrophages. However, urethral lumen at the site of urethrotomy was comparable to near normal. Histochemically, the neutral mucopolysaccharide content in luminal epithelium and popria-submucosa was low at operation site, but acidic mucopolysaccharide content was moderate.

In group C, urethral lumen did not show any stricture but slight cellular reaction was observed at the site of operation. Neutral mucopolysaccharide content was high at suture site but adjoining fibroblastic tissue reacted more intensely for acidic mucopolysaccharide.

The hyperplastic reaction was observed in the luminal epithelium of group D. The zone of reaction was extended up to the periurethral tissue, where fragment of suture material was encapsulated by immature and mature fibroblasts, polymorphoneuclear leucocytes, plasma cells and lymphocytes. However, fibroblastic activity was less severe as compared to catgut but more then polyglactin-910. Neutral mucopolysaccharide content was high at suture site but adjoining fibroblastic tissue showed more acidic mucopolysaccharide. The following conclusions could be drawn from this study.

1) Unsutured urethra showed maximum post-operative complications like infiltration of urine and fistula formation.

2) The post-operative complications like urine infiltration, fistula formation and dehiscence were minimum with 3-0 polyglactin-910 followed by 3-0 black braided silk, 3-0 chromic catgut as compared to unsutured urethrotomy wound.

3) Urethral lumen did not show appreciable constriction in any of the group till the 45th post-operative day.

4) Histomorphological and histochemical results showed satisfactory healing in all the groups.

5) On the basis of clinical observations macroscopic and microscopic findings of urethra, 3-0 polyglactin-910 proved to be the best suture material as compared to 3-0 black braided silk and 3-0 chromic catgut.

6) Suturing of urethral wound with 3-0 polyglactin-910 showed least fibrous tissue reaction at sutured area as compared to 3-0 chronic catgut and 3-0 black braided silk.

7) Inflammatory cellular reaction was least observed in tissue section of urethra in which urethral incision was sutured with 3-0 polyglactin- 910 followed by 3-0 black braided silk and 3-0 chromic catgut.

8) There were no significant haematological changes observed during urethrotomy wound healing in unsutured and sutured urethrotomy wound.

9) Blood urea nitrogen and serum creatinine level remained near normal during urethrotomy wound healing in all groups.

10) Long term studies are required to confirm the development of stricture.

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Chapter - 7

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