Gastric neobladder: an experimental study in dog

M. M Seif 1*, M. S. Aimen1, H. H. Kamel2

1 Department of Surgery, Anaesthesiology and Radiology, Faculty of Veterinary Medicine
Beni-Suef University

2 Department of Clinical Pathology, Faculty of Veterinary Medicine Beni-Suef University

The urinary bladder of 15 clinically normal dogs was excised and the ureters were implanted into an isolated, vagotomized gastric segment derived from the fundic region of the stomach. The gastric segment was closed to form a neobladder. Continence was maintained with a "nipple valve" created at the tubularized end of isolated segment of stomach. Clinical, radiological, ultrasonographical, urine and blood analysis and histopathological examination were carried out for assessment of the technique. Eleven cases showed an apparently normal bladder function. Two cases suffered from renal hydronephrosis and other two suffered from incontinence. It was concluded that gastric neobladder urinary diversion is satisfactory for clinical use in dogs.

Reconstruction of the lower urinary tract using small or large intestine has been widely used in many different configurations to achieve a capacious, non-refluxing, low-pressure reservoir for urine storage (Nguyen and Mitchell, 1991). A variety of detubularized bowel segments have been used to construct continent urinary reservoirs with the stoma on the abdominal wall called (stomal urinary reservoirs) or orthotopically to the urethra (orthotopic neobladders) (Shamsa 1998). Gastric segments have been studied for bladder augmentation and continent gastric conduits as alternatives to the use of intestinal segments. (Piser et al., 1987)

Formation of an artificial urinary bladder from an isolated gastric pouch was first performed experimentally in dogs in 1956 (Sinaiko, 1956). Then Leong (1978) described the use of stomach for such purposes in humans and Adams et al. (1988) used gastrocystoplasty in the pediatric population.

The objectives of this study were to determine if gastric neobladder urinary diversion could preserve normal renal function and structure after total cystectomy.

Materials and Methods

Fifteen apparently healthy adult mongrel dogs (5 males and 10 females) were used in this study. Their ages ranged from 1.5-2.5 years and their weight from 20-30 kg. The animals were kept in separate kennels and put under observations and examination, one week before the experiment. The urinary system proved to be normal according to biochemical, radiological and ultrasonographic examination.

Surgical technique. The animals were prepared for aseptic surgery as usual. They were premedicated with atropine sulfate in a dose of 0.04mg/kg subcutaneous and promazine HCl in a dose of 2-3mg/kg intra-muscular. Anesthesia was induced and maintained by intravenous injection of sodium thiopental in a dose of 20-30mg/kg.2.5%.

Each dog was placed in dorsal recumbency, and the abdomen was prepared for aseptic surgery. The abdominal cavity was exposed through a ventral midline celiotomy incision from xiphoid to pubis. Excision of a wedge-shaped portion of the gastric fundus measuring 5-7 cm, deriving its blood supply from both the short gastric and left gastroepiploic arteries was performed (Fig.1). The right gastroepiploic artery was ligated and divided at the distal end of the proposed gastric segment. The gastric which tissue isolated to form the gastric neobladder was not more than one-third of the stomach and did not include the pylorus or antrum. Care was taken to avoid trauma to the vasculature and vagal innervation of the lesser curvature of the stomach. The incised edges of the greater curvature were apposed with a two-layer

* Corresponding author. Tel.: +20 127456156; fax: +20 822327982.
E-mail address: seif_th2003@yahoo.com (M. M. Seif).
inverting suture of 3-0 polydioxanone. A wedge shaped gastric segment with intact vasa bervia from the gastroepiploic artery was then rotated down to the pelvis.

The ureters and their blood supply were resected from the bladder, and complete cystectomy was performed. The ureters were implanted 3 to 4 cm apart in the gastric segment by submucosal tunneling. The distal end of each ureter was spatulated by a 5 mm longitudinal incision, and the ureter was sutured to the gastric mucosa with simple interrupted sutures of 5-0 polydioxanone (Fig.2). The isolated gastric segment was closed with a two-layer inverting suture of 3- zero chromic gut to form a neobladder (Fig. 3).

The end of the stomach segment was sutured in a tube-shaped manner and positioned to form a nipple valve. By telescoping, 3 to 5 cm of the tubularized gastric segment end upon itself the valve was created. The serosal surface of the tubularized gastric segment end was scarified with electrocautery to cause adhesions, and two rows of 2-0 chronic gut seromuscular sutures preplaced to help create and maintain the intussusception (Fig.4). A Foley catheter was passed from the neobladder to the outside through the urethra and fixed to skin by suture. It left in situ for 7 days for drainage of residual urine and for flushing of bladder with mild antiseptic povidone iodine twice daily.

Each animal received kanamycine (10 mg /kg body weight) and penicillin streptomycin combination (10.000 I U/ kg + 10 mg/ kg streptomycin) for 5 days via intramuscular injection. Fluids were given I/V for 5 days in a dose of 50 ml (kg/day in the form of glucose 5 % and Ringer's solution). Diluted milk and water were given for 2 days, and then ordinary food was given. The skin stitches were removed after eight days.

**Radiographic examination.** Excretory urography and ascending cystography were done.

**Excretory urography (I.V.U.).** Radiographic films were taken at 5, 15 and 20 minutes after completion of the injection of contrast material (Sodium diatrizoate, 76 % concentration) diluted to about 20%. The technique was performed before the surgical interference and at intervals of one, 4 and 8 weeks postoperatively to follow up function of the kidney and continence of the gastric neobladder. Radiographs were taken at potential of 65Kvp, 25 mAs and 100cm ffs.

**Ascending cystography.** It was performed after euthanasia; the kidney ureter and bladder specimens were taken and ascending cystogram with contrast substance was made for detection of the vesicoureteral reflux and presence of hydronephrosis.

**Ultrasonography of the urinary tract.** Ultrasonographic examination was performed using 240 PARUS VET Pie Medical Equipment (B.V. Philips weg 16227 Maastricht) and 3.5/5.0 MHz curved array electronic transducer (R40-401665) and 6.0/8.0 MHz linear array probe (6 CM-401663).

**Ultrasonographic examination of the urinary bladder.** The animal was prepared prior to the examination by injection of furosemide sodium (Laxis) in a dose of 1 - 2 ml / kg/body weight and normal saline in a dose of 20 ml/kg body weight to achieve filling of the bladder with urine 10 - 15 minute before examination. By using the suprapubic technique the animal was placed in dorsal recumbency. The linear transducer was placed just superior to the pubic symphesis and angled caudally. Various degrees of angulation were required to evaluate the base, body and dome of the bladder. Scans were obtained in longitudinal and transverse orientation. The length, width and depth of the bladder were measured electronically by means of 2 cursors. The ultrasonography device automatically computed the bladder volumes. The ultrasonographic examination was performed before the operation and at intervals of 1, 4, and 8, weeks postoperatively.

**Urine analysis.** Urine sample was taken by using catheterization, before the operation and at intervals of 3, 7, 15, 30 and 60 days postoperatively. By using test strip (Comber 10 test, manufactured by Boehringer Mannheim, GmbH, Mannheim, Germany) the urine analysis was performed. Specific gravity, pH, leucocytes, nitrite, protein, glucose, ketone bodies, urobilinogen, bilirubin and blood were determined.

**Determination of certain blood parameters.** Serum samples were taken, before the operation and at intervals of 3 days, 1, 2, 4, and 8 weeks postoperation. Urea, createnin, sodium,
Table (1): Ultrasonographic measurements of gastric neobladder:

<table>
<thead>
<tr>
<th></th>
<th>Preoperation</th>
<th>1(^{st}) week post operation</th>
<th>4(^{th}) week</th>
<th>8(^{th}) week</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume (cm(^3))</td>
<td>152.3±13.5</td>
<td>65.9±10.7</td>
<td>89.7±8.7</td>
<td>123.5±6.7</td>
</tr>
<tr>
<td>Length (cm)</td>
<td>13.2±1.3</td>
<td>9.5±0.9</td>
<td>11.2±0.98</td>
<td>12.3±7.5</td>
</tr>
<tr>
<td>Width (cm)</td>
<td>6.01±0.45</td>
<td>4.6±0.23</td>
<td>5.1±0.88</td>
<td>5.8±0.97</td>
</tr>
<tr>
<td>Depth (cm)</td>
<td>5.4±0.63</td>
<td>3.9±0.76</td>
<td>4.6±0.86</td>
<td>5.1±0.87</td>
</tr>
</tbody>
</table>

Table (2): Biochemical blood parameters.

<table>
<thead>
<tr>
<th>Date Parameter</th>
<th>Preoperation</th>
<th>3days post operation</th>
<th>1(^{st}) week</th>
<th>2(^{nd}) week</th>
<th>4(^{th}) week</th>
<th>8(^{th}) week</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urea mg/dl</td>
<td>28±3</td>
<td>32.3±2.1</td>
<td>36.2±4.7</td>
<td>33.2±1.2</td>
<td>31.0±0.9</td>
<td>29.4±3.6</td>
</tr>
<tr>
<td>Creatinine mg/dl</td>
<td>0.9±0.11</td>
<td>1.3±0.15</td>
<td>1.2±0.65</td>
<td>1.1±0.29</td>
<td>1.0±0.33</td>
<td>0.9±0.13</td>
</tr>
<tr>
<td>Chloride mEq/L</td>
<td>117±5</td>
<td>108±6</td>
<td>108±2</td>
<td>108±3</td>
<td>109±2</td>
<td>111±6</td>
</tr>
<tr>
<td>Potassium mEq/L</td>
<td>5.2±0.58</td>
<td>6.65±0.98</td>
<td>6.49±0.95</td>
<td>6.47±0.89</td>
<td>6.43±0.99</td>
<td>5.7±0.95</td>
</tr>
<tr>
<td>Sodium mmol/L</td>
<td>152.2±7.7</td>
<td>158±5.3</td>
<td>156±6.2</td>
<td>155±5.3</td>
<td>154±4.3</td>
<td>153.1±2</td>
</tr>
</tbody>
</table>

Potassium and chloride were determined through specific kits. For determination of urea a urease modified Berthelot reaction (Bio Merieux Imprine en France, Res Lyon) was used. Creatinine kinetic was used for determination of creatinine without deproteinization (Bio Merieux Imprine en France, Res Lyon). Quantitative determination of potassium in serum was performed by using A turbidimeter method (Eagal diagnostic/Desoto, texas, U. S. A.). Chloride thiocyanate method was used for determination of chloride in serum (Quinuca clinica oplicada S. A. torrogona/Spain). Colorimetric determination of sodium in serum was achieved by using Magnesium uranyl acetate method (Quimica clinica oplicada S. A. torrogona /Spain).

**Histopathological examination.** The animals were sacrificed at two months after the operation. Kidneys, ureters, and gastric neobladder were dissected for histopathological examination. Small specimens were taken from bladder wall, ureter and kidney. These specimens were fixed in 10% neutral formaline, dehydrated in alcohol, cleaned in xylol and embedded in paraffin. The specimens were then cut with a microtome into 5-micrometer thick sections, stained with hemotoxylene and eosine (H & E).

**Statistical analysis.** Using T-test performed statistical analysis for blood parameters and bladder measurements.

**Results**

**Clinical findings.** All dogs showed signs of straining and arched back at the 2nd day after operation. These signs disappeared at the 3rd day. Eleven dogs recovered uneventfully. Four dogs showed some forms of complications, two of them sacrificed as the result of renal failure due to hydronephrosis at 7 and 9 days after operation. The others showed signs of incontinence at the 12 and 18 days after operation. The remained dogs stayed alive until it was sacrificed at the 2nd month post operation.

**Radiographic findings.** Preoperative and postoperative intravenous urography revealed normal pelvicalyceal system and ureters in eleven dogs among the used animals in the study. At one week postoperatively, the gastric neobladder showed irregular outline for four weeks and regressed gradually till the 8th week postoperatively. The rate of growth of the gastric neobladder increased till the end of the study (Figures 5,6 and7). The ascending urography of urinary system specimens of hydronephrotic cases revealed, unilateral and bilateral hydroureter and hydronephrosis (Fig. 8).
Ultrasonographic results. The shape of the bladder after operation resumed a pear shaped gastric neobladder with anechoic appearance. The dimensions of the gastric neobladder (Table 1) were increased by time till the end of the experiment (Fig. 9,10, 11). The kidneys and ureters showed no pathological changes in eleven dogs during the time of the experiment. The two-hydronephrotic cases showed increase in the length, width, depth, medullary thickness and diameter of the renal sinus with gradual decrease in the cortical thickness. The ureter was clearly evident dilated and could be differentiated from the renal blood vessels at the level of the hilus and the dilated renal pelvis appeared as a anechoic central cyst like formation. The dilated ureter appeared as an anechoic caudally coursing tubular structure originating from the renal pelvis (Fig.12).

Urine analysis. The urine appeared more viscid in consistency, dark yellow in color and turbid with suspended dark particles till the 7th day post operation. After that it returned to its normal color, viscosity and without turbidity till the end of the experiment. In eleven animals no changes were observed by using urine strips concerning protein, glucose, ketones, bilirubin, nitrites and urobilinogen after surgery. The pH changed
Fig (5): Gastric neobladder (GB) 1 week post operation

Fig (6): Gastric neobladder (GB) 2 weeks post operation

Fig (7): Gastric neobladder (GB) 8 weeks

Fig (8): A- Bilateral hydrenephrosis

Fig (9): Ultrasonographic appearance of gastric bladder (GB) after 1 week:
L- length of the bladder
D- depth of the bladder

Fig (10): Ultrasonographic appearance of gastric bladder (GB) after 4 weeks:
L- length of the bladder
D- depth of the bladder

Fig (11): Ultrasonographic appearance of gastric bladder (GB) after 8 weeks

Fig (12): Ultrasonographic appearance of hydronephrotic kidney:
M- medulla
C- cortex
RS- renal sinus
U- ureter
Fig. (17): Full thickness of gastric mucosa of gastric neobladder (2 months postoperation) (H&E 10x10).
Fig. (18): Ill-vascularized gastric neobladder with disrupted degenerated gastric glands (2 months post operation)(H&E 10x10).
Fig. (19): Gastric neobladder showed prominent chief and parietal cells (2 months post operation)(H&E 10x40).
Fig. (20): Ureter showed prominent transitional epithelium of normal structures (2 months postoperation) (H&E 10x20).
Fig. (21): Ureteral uroepithelium showed proliferative desquamatve (2 months post operation)(H&E 10x20).
Fig. (22): Kidney showed normal histopathological structures (2 months postoperation)(H&E 10x10).
Fig. (23):Kidney showed cystic dilated renal tubular hydronephrosis (2 months post operation)(H&E 10x10).
toward acidity till the 7th day, and then deviated towards alkalinity till the 15th day, and finally it returned to normal. Two cases suffered from bladder incontinence showed proteinurea, haematouria and presence of nitrite. The affected cases with hydrenephrosis present glucosuria, proteinuria and haematouria.

**Biochemical blood parameters.** The blood parameters and electrolytes measurements including urea, creatinene, chloride, sodium and potassium were presented (Table 2). The urea and creatinin values were increased significantly at the 1st week and 3rd day respectively post operation. These values began to regress gradually till the end of the study. The chloride values were decreased significantly at the 2nd week after operation, and then gradually increased till the 8th week. Potassium and sodium values were increased non significantly at the 4th week and the 3rd day respectively then it decreased gradually till the end of the study. The hydrenephrotic cases showed raised blood urea and createnin respectively from (36.5 mg/dl) and (1.6 mg/dl) at the 3rd day, reached to (255.3 mg/dl) and (9.3 mg/dl) before sacrification.

**Pathological findings.**

**Macroscopical findings.** The upper urinary tract and the gastric neobladder showed no evident gross pathological changes and no adhesions with the surrounding structures except two cases that revealed hydrenephrosis (Fig.13A, 13B, 14). The gastric neobladder was still intact with its vascular pedicle (Fig.15) and with the implanted ureters (Fig.16) without deterioration. The gastric neobladder of incontinent cases showed signs of inflammation.

**Microscopical findings.** Changes in the gastric neobladder were variable and were related mainly to the degree of vascularization. In well-vascularized urinary bladder, the mucosal folds were clear during the experimental period (Fig. 17). The glandular epithelium of gastric tubules was intact and could be easily differentiated into parietal and central (Chief) types of cells. The parietal (oxyntic) cells were especially prominent having intense eosinophilic polyhedral cytoplasm and rounded nucleus (Fig. 19). The mucus - secreting chief cells having gastric tubules showed no great activity. These cells had a basophilic cytoplasm and flattened nucleus. However, some of the tubular glands were dilated and their lumen might contain zymogen granules. Blood capillaries embedded in the reticular tissue between gastric glands contained intact erythrocytes. The blood vessels of the plexus located in the muscularis and traversing the submucosa were patent. Infiltrating leukocytes in the gastric wall were few or absent. The submucosal connective tissue, plain muscles of the submucosal and muscularis, and serosa showed no abnormal changes. An individual number of mononuclear cells, including plasma cells, were distributed throughout the mucosa.

In badly vascularized cases, changes in the gastric mucosa were zonal. In severely affected areas the glandular epithelium underwent degenerative changes and necrosis (Fig. 18). The chief cells were more sensitive and many cells disappeared leaving tubules lined mainly with parietal oxynic cells that showed coagulative necrosis. The reticular tissue between the gastric glands and submucous connective tissue showed either degenerative changes or proliferation replacing distorted gastric glands. Many muscle fibers of the submucosa and muscularis had pyknotic nuclei. Blood capillaries of the superficial plexus between gastric glands were not seen.

The structure of the ureter in cases examined after 2 months postoperation was normal, consisting of mucosa, submucous muscle bundles and external connective tissue (Fig. 20). In one case examined after 2 months, the epithelium lining of the ureter showed degeneration and the cells were desquamated (Fig. 21).

Kidney of the animals revealed normal histological appearance (Fig. 22) while the hydrenephrotic kidney showed dilated renal tubules (Fig. 23).

**Discussion**

The search for an ideal material to augment or replace diseased bladders remains elusive for the reconstructive urologist. Although ileum and colon have been used for decades for such purposes, concerns have arisen regarding complications specific to their use (Hasegawa et al., 1989). Continuous search for an ideal tissue for reconstruction of the bladder showed the feasibility and advantages of gastric bladder reconstruction, because of the easy performance, availability of donor segment and limitation of postoperative morbidity (Kennedy, et al., 1988).

It was suggested that the gastric fundus might be the ideal urinary bladder replacement because the gastric mucosa was virtually nonabsorbing and could prevent metabolic alterations caused by the absorption of urine that occurred with other bowel conduits. The normal gastric secretions of acid and lysozymes could
create a bactericidal environment for urine collection and temporary storage, minimizing urosepsis and ascending infection. Because there is more gastric tissue than needed for normal gastrointestinal function, partial gastrectomy to create a gastric conduit is physiologically and anatomically possible, (Qiu, et al., 2003).

The stomach has been applied to patients in whom no other type of bowel was available for urinary reconstruction. This included those with previous radiation and those with absence of large bowel or reduced small bowel. The stomach also proved to have a merit in patients with renal compromise and metabolic acidosis, because of its ability to secrete chloride and hydrogen ions; in patients with chronic urinary infections, in patients with neurogenic low-compliance bladders and in patients with difficulty with catheterization because of excessive mucus (Sheldon et al., 1995; Leonard et al., 2000, and Baydar et al., 2005).

From the surgical point of view the use of the stomach for bladder reconstruction is clearly advantageous, as Stomach is an easy tissue with which to work and has a ready pedicle that allows an isolated segment to reach the pelvic region without difficulty. It also is a compliant tissue and will remain so when used in bladder reconstruction. In addition, the thick wall of the stomach easily accommodates submucosal tunnels to the reimplanted ureters. This agreed with Gosalbez et al. (1993) and, Koraitim et al. (1999). These results are in contrast to enterocystoplasty, as bladder augmentation with ileum or colon; need detubularization leading to poor bladder urodynamics (Atala et al., 1993). Also the ureteral reimplantation is usually resigned to the residual bladder plate or along the teniae of the colon (DeFoor et al., 2003).

In the present study a modification was performed in the surgical technique described by Mitchell et al. (1992), where a nipple valve was performed at the tubularized end of the gastric segment acting as urinary sphincter. On contrary to Lin et al. (2000) who anastomosed directly the posterior leaflet of gastric segment to the remaining membranous urethra leading to high incontinence rates. Previous studies have described placement of an artificial urinary sphincter and simultaneous augmentation cystoplasty with a segment of bowel (Ligt et al 1995). The artificial urinary sphincter is one of the many treatment options for urinary incontinence. Its efficacy in treating incontinence and safety profile has been previously reported (Simeoni et al., 1999; Kryger, et al., 1999).

Complete urinary continence was achieved in 11 cases. This was attributed to competent urinary sphincter and clean intermittent catheterization for bladder drainage that improved the continence rate and preserved upper urinary tracts (El- Ghoneimi et al., 1998). Also due to better bladder emptying as a result of inherent fibromuscular properties of gastric patch that contributed to the force of urination. The etiology of incontinence cases in this study could be attributed to decreased bladder capacity, decreased compliance, overactivity or other discoordinated voiding patterns. This was agreed with mentioned by Bogaert et al. (1994).

Hydronephrosis was observed in two cases in the present study may occur due to high intravesical pressure spikes resulting from decreased bladder capacity and decreased compliance. This clearly affects the upper urinary tract in a deleterious manner leading to hydronephrosis and worsening renal function, as explained by (Holmes et al., 2001).

Urine analysis showed physical changes in color and aspects of the urine till the 7th day postoperation, which may be related to the mucous and acid secretion. The changes of pH of urine toward the acid side till the 7th day may be attributed to the HCl secreted by the stomach graft, after that, the pH of urine changed toward alkalinity till the 15th day after operation, and this may be due to the delayed storage of urine in the bladder, incomplete emptying of urine and infrequent urination. Mevorach et al. (1995) proved that urinary pH does not reflect true pH of the bladder mucosa as measured by a probe because of the buffering effect of urine on gastric acid secretion. Bogaert et al. (1995) and Plawker et al. (1995) reported that in the fasting state pH was neutral, there was no titratable acid in the urine and serum gastrin level was normal in all cases. After a meal urinary acid secretion and serum gastrin level increased markedly. Bladder distention did not result in urinary acid secretion or gastrin secretion.

The picture of urine analysis showed that eleven cases had no signs of urinary infection. This may be due to aciduria and decreased mucus production. The same was reported by (Lewis et al.,1995; Plaire et al., 2000; Burgu et al., 2007). However, some investigaor reported that their patients had positive urine culture and episodes of cystitis relieved by antibiotics Kurzrock et al. (1998). According to
interpretation of Kaneko (1989) the urine analysis of the cases suffered from incontinence, indicating presence of infection due to presence of proteinuria, haematuria and nitrite. In cases suffered from hydroureter nephrosis the urine picture showed presence of glucosuria, proteinuria and haematuria as the affected tubules unable to reabsorb the glucose, leading to its descend in urine while the leakage of protein and blood was due to affection of glomeruli.

The results of blood analysis showed that, there were no significant changes of sodium, and potassium while urea and createnine increased significantly at the third day after operation. The chloride value was decreased significantly till the fourth week after operation and then slightly raised, but not reached mean value as before operation. This may be attributed to the chloride excreted by the stomach graft, a fact that coincides with that stated by, (Filho et al., 2001). The net acid-secreting properties of the stomach deserve special attention because they can promote resolution of metabolic acidosis in patients with end stage renal disease and may obviate use of bicarbonate supplementation (Kurzrock, 1998) However, acid hypersecretion can also lead to serious adverse effects such as systemic metabolic alkalosis (DeFoor et al., 2003).

An increase in bladder volume was recorded. This is in agreement with previous reports of Mingin et al. (1999), Bleustein et al. (2000) and Pareek, et al. (2001). A gastric segment used for bladder replacement can undergo considerable changes over time, due to direct contact with urine (Campodonico et al., 2002). The histopathological findings showed no pathological changes in the case of well-vascularized graft. In the case of poorly vascularized graft, different pathological changes were seen. This bad vascularization may be attributed to the tension lying on the long vascular pedicle of the gastric graft, by the surrounding organs. This was illustrated by Vajda et al. (2005) who stated that the stomach graft has some potential disadvantages comprising a long vascular pedicle, acid secretion and theoretically Vitamin B12 malabsorption.

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References


