

13

# Difficulties in Laparoscopic Renal Cyst Removal and Giant Hydronephrosis Nephrectomy

Hamdy M. Ibrahim, Ahmed Al-Kandari, Hani S. Shaaban, and Inderbir S. Gill

# Introduction

A giant kidney is a massively enlarged kidney having more than 1 L of fluid within the collecting system or is one that is the seat of large multiple cysts. It is defined radiographically when the enlarged kidney meets or crosses the midline, or spans more than five vertebral lengths [1]. The majority of giant hydronephrotic kidneys are nonfunctional and symptomatic, making nephrectomy the procedure of choice [2]. Because of the massive size and altered anatomic relationships of these kidneys, surgical excision is challenging. The other common cause of giant hydronephrotic kidney is the presence of large renal cysts, which may be simple or associated with other pathologies. Renal cystic disease is a common incidental, radiographic, and postmortem finding. It is estimated that evidence of renal cysts exists in 50% of the adult population [3]. The increased use of imaging modalities, such as ultrasonography and computed tomography (CT), has produced a corresponding increase in the detection of renal cystic disease. Simple renal cysts occur with an incidence of at least 20% by age 40 and 33% at age 60 [4]. Most of these lesions are asymptomatic. At times, the lesions may be associated with dull renal angle pain, flank pain, hypertension, a palpable mass, hematuria, infection, and collecting system obstruction [5, 6]. Certain renal cysts can be associated with other pathologic

H.M. Ibrahim, MD

Department of Urology, Fayoum University, Fayoum, Egypt

Ahmed Al-Kandari, BM BCh FRCS(C) (🖂)

Department of Surgery (Urology), Faculty of Medicine, Kuwait University, Jabriyah, Kuwait e-mail: drakandari@hotmail.com

H.S. Shaaban, MD, MSc Department of Urology, Ahmadi Hospital, Ahmadi, Kuwait

I.S. Gill, MD USC Institute of Urology, Keck School of Medicine, University of Southern California, Los Angeles, CA, USA

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conditions, such as autosomal dominant polycystic kidney disease (ADPKD) and acquired cystic disease in chronic dialysis. ADPKD is an important cause of renal failure, accounting for approximately 9–10% of patients on chronic hemodialysis. Other clinical manifestations include hypertension, chronic pain, hematuria, urinary tract infection, stone formation, and cyst infection. Symptomatic renal cysts can be treated by percutaneous aspiration with or without injection of sclerosants [7], percutaneous marsupialization, open surgery, and, currently, by laparoscopic surgery by transperitoneal and retroperitoneal access with similar efficacy to open surgery and less morbidity [5, 7, 8]. The excellent results achieved with laparoscopic decortication of symptomatic simple renal cysts, first described by Hulbert et al. in 1992, have prompted its acceptance [9]. In this chapter, the authors present the laparoscopic approaches for giant kidneys of varied pathology with an emphasis on the surgical options used in their management.

## **Evaluation**

## **Evaluation of Renal Cystic Disease**

Renal cystic disease can be adequately evaluated with ultrasonographic techniques. For more complicated lesions, renal CT is the cornerstone of diagnosis. Based on CT scan, renal cysts have been classified into four categories by Bosniak [10]:

*Type I*: This simple cyst is the most common and generally requires no treatment. A large simple cyst will rarely cause symptoms or obstruction, which would require intervention.

*Type II*: This homogenous, hyperdense cyst has thin septations and fine calcifications that do not enhance after intravenous injection of contrast medium. Cysts that contain multiple hairline thin septa and/or walls or smooth, minimally thicker septa and/or walls may contain perceived enhancement and/or coarser calcification but no measurable enhancement. Also included are uniformly high attenuation lesions greater than 3 cm that may be totally intrarenal [11].

*Type III*: This multiloculated cystic mass or cyst has thick, irregular calcifications, a thick wall, or nodularity.

*Type IV*: A cyst associated with a solid component; such lesions are considered to be cystic malignancies and should be managed with radical nephrectomy.

Ultrasound is the recommended imaging modality for reliable identification and follow-up of cystic lesions. Adding the IIF category increases the accuracy and, thus, the clinical impact of the categorization system, as evidenced by a low rate of progression in category IIF cysts and an increased malignancy rate in surgically treated category III lesions. Progression in complexity but not in size appears to be the most important indication of malignancy. The high malignancy rate suggests that all category III cysts should undergo definitive treatment when clinically feasible [12]. For complicated cysts that are more likely benign (category II), but have some worrisome findings, follow-up CT should be performed to establish the benign behavior of the lesion (category IIF). An initial follow-up CT is performed at 6

months and 1 year, and then again after one more year. If the lesion has not changed in appearance during this period, a benign nature is established [13]. Equivocal cysts raising the possibility of malignancy can be further evaluated with CT, but, if any doubts remain regarding the nature of the lesion, percutaneous aspiration or biopsy is advocated to provide a definitive answer. The sensitivity for the detection of malignancy using percutaneous techniques is 90%. The specificity for the determination of benign disease is 92% [14].

## Indications

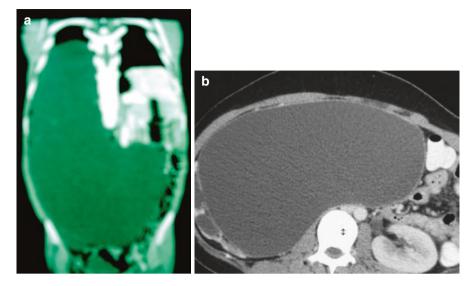
Depending on the symptoms, size, site, locations, number, presence of infection, suspicion of malignancy, and other associated pathologic conditions, the following renal cystic disorders can be managed by laparoscopic intervention:

- Bosniak type I and II renal cysts large symptomatic renal cysts (≥10 cm in size) may be directly taken up for laparoscopic management. Smaller symptomatic renal cysts may be treated by percutaneous aspiration alone or with a sclerosing agent, and, failing that, a laparoscopic resection can be undertaken [8].
- 2. Renal cystic masses with malignancy should be treated by laparoscopic radical nephrectomy [10].
- 3. Peripelvic or parapelvic cysts [15], ADPKD, renal hydatid cysts.

Laparoscopic evaluation, marsupialization, excision, and radical nephrectomy for a cystic mass harboring malignancy offer a definitive treatment for patients who have renal cystic diseases and can obviate open surgery [16].

## **Evaluation of Giant Hydronephrosis**

Giant hydronephrosis can be adequately evaluated with ultrasonographic scanning. It is very important to delineate the extent of the dilated hydronephrotic kidney and its relationship to surrounding organs. The addition of coronal views on a CT scan is very helpful (Fig. 13.1). Magnetic resonance imaging (MRI) is also useful in delineating the intraabdominal anatomy in these challenging cases. Renal dynamic scintigraphy is used to determine the split function (split GFR mL/min) of the hydronephrotic units. Kidneys with a split glomerular filtration rate (GFR) of less than 10 mL/min are considered unsalvageable. Most giant hydronephrotic kidneys are almost nonfunctioning and any indication for retrograde ureteropyelography is rare. Routine blood work ups, including complete blood cell count (CBC), renal function tests, and urine tests, should be performed. If a patient is having respiratory difficulty due to the large hydronephrotic kidney compressing the diaphragm, nephrostomy drainage may be required for 3 days or longer until the patient's condition stabilizes. In case of significant laparoscopic difficulties, the patient should consent to a possible open conversion.



**Fig. 13.1** (a) Preoperative CT with IV contrast showing right-sided giant hydronephrosis occupying the whole abdomen and displacing abdominal contents. (b) Right-sided giant hydronephrosis with no contrast excretion. Notice the normal average-sized left kidney

# **Operative Techniques**

The authors have reported single stage decompression of giant hydronephrotic kidney. This is achieved under the same anesthesia with a percutaneous nephrostomy insertion and decompression until no further drainage is noted. Then transperitoneal trocars are inserted (as routine) and the procedure is continued [17]. Others have suggested a staged approach because of the possible effects of sudden abdominal decompression that results in a change in the hemodynamic balance. The hydronephrotic kidney is first slowly decompressed with drainage, and nephrectomy is then performed after the patient's cardiorespiratory and alimentary systems have been stabilized. Therefore, a two-stage procedure with slow decompression by percutaneous nephrostomy before the nephrectomy is preferred in the compromised patient [15, 17–19]. Recently Harper et al. claimed that, prior to laparoscopic nephrectomy, renal decompression via ureteral catheters is helpful to achieve adequate working space [19]. Retrograde ureteral decompression is an important consideration to reduce spillage of potentially infected urine into the peritoneum or retroperitoneum. Also, partial decompression of the kidney to 25–50% turgidity allowed adequate space for dissection and visualization while preserving enough turgidity to allow easier identification of surgical structures.

## **Retroperitoneal Approach**

#### Laparoscopic Nephrectomy Technique

General anesthesia was given via a cuffed endotracheal tube and the patient was catheterized preoperatively. The patient was placed in the kidney position with a minimal kidney bridge – enough to increase the space between the costal margin and iliac crest without compromising the retroperitoneal space. A 1.5-cm to 2-cm transverse skin incision was made 1–2 cm below and posterior to the tip of the 12th rib in the lumbar (Petit's) triangle between the 12th rib and the iliac crest, bounded by the lateral edges of the latissimus dorsum and external oblique muscles. A tunnel is created down to the retroperitoneal space by blunt dissection using Overhold or Kocher forceps. This tunnel is dilated until an index finger can be inserted to push the peritoneum forward, thus creating a retroperitoneal cavity. Three dissection techniques of the retroperitoneum are used. The first technique uses a modification of the Gaur balloon technique by ligating a latex balloon using the middle finger of a powder-free surgical glove on an 18 F Mercier catheter. The second technique employs a balloon trocar system consisting of a latex balloon ligated to an 11 mm metal trocar sheath. During instillation of 500–1000 cc warm normal saline through the insufflation channel of the trocar, dissection was monitored with the telescope inserted into the trocar sheath. In the last technique, dissection of the space between the lumbar aponeurosis and renal (Gerota's) fascia is performed exclusively with the index finger [20].

Two secondary trocars (ports II and III) were inserted to one side of an index finger introduced through the primary access site. To avoid injury to the finger, a track was dilated first, using forceps. The initial incision (port I) was closed around the port using a mattress suture to prevent gas leakage. After establishing pneumoretroperitoneum (maximum carbon dioxide pressure 15 mmHg), residual minor adhesions were lysed easily and the renal fascia was opened longitudinally for exposure of the psoas muscle, representing the most important anatomical landmark of retroperitoneoscopy. If necessary, an additional 5 mm trocar (port IV) was inserted under endoscopic view to retract the kidney during the dissection.

Independent of the individual retroperitoneoscopic procedure, it is important to incise Gerota's fascia completely. The wide longitudinal incision opens the retroperitoneal space, thereby adding to the effect of the carbon dioxide insufflation, allowing retraction of the peritoneum and exposing all further anatomical landmarks. The usual technique of identifying the upper ureter first and then tracing it up to the renal pelvis and vessels has been modified as the hugely dilated kidneys restrict exposure of the upper ureter. The kidney was dissected posterior and then along the lateral surface as far medial as possible, and along the upper and lower poles. Inadvertent injury to the renal sac with early deflation of the kidney before the initial mobilization causes considerable difficulty in the subsequent dissection as the sac collapses, and thus special care should be taken to prevent this complication [2].

Once vision and available space limit further dissection, the hydronephrotic sac was deflated by percutaneous aspiration using a Veress needle along the anterior axillary line, while simultaneously monitoring the insertion and deflation laparoscopically. The aspirated fluid was measured to determine the volume of the hydronephrotic kidney. Further posterior dissection was performed after retracting the kidney anterior using a tri-flange retractor.

Once adequately dissected, the collapsed renal sac was brought out through the anterior port (extracorporeal retraction) as intracorporeal retraction of the large renal sac does not provide adequate countertraction to aid dissection. This procedure stretched the hilar structures and further dissection around the hilar region was performed with relative ease, allowing for individual clipping of the renal vessels. The ureter was then clipped and divided between clips [2].

The kidney, partly freed extracorporeally with the rest in the retroperitoneal space, was delivered after suitably enlarging the anterior port used for extracorporeal retraction. The retroperitoneum was then irrigated with saline and inspected for hemostasis, and a drain was placed. Finally, the port sites were closed with muscle and skin sutures.

#### Laparoscopic Management of Renal Cysts

If the retroperitoneal space is created underneath Gerota's fascia, the kidney lies directly in front; otherwise, the overlying fascia of Gerota is usually thinned out and the blue-domed cysts are easily recognized. Gerota's fascia is divided in relation to the location of the cyst. If the cyst is located anteriorly, medially, or anteromedially, the kidney is mobilized on the lateral border and retracted posteriorly with the help of a triflanged retractor to delineate the anterior surface of kidney.

A combination of blunt and sharp dissection using a dissector and scissor permits rapid and complete mobilization of the cyst surface. If the cyst is very tense after initial exposure, it can be punctured and grasped with the help of an atraumatic dissector that provides countertraction for further dissection. The renal cyst can be further treated in the following ways.

- The renal cyst can be completely excised when a good plane of cleavage can be developed between the cyst and the underlying parenchyma.
- If the previous maneuver is not possible, near total excision of the cyst wall is performed to avoid damage to the underlying collecting system or renal parenchyma.
- The renal cyst can be marsupialized by excising the wall.

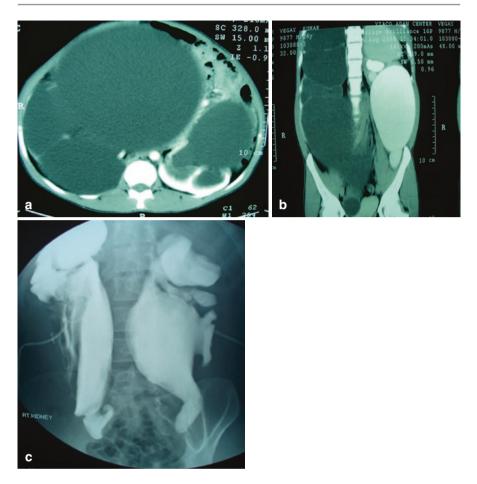
In cases of peripelvic cysts, the presence of a ureteral catheter is of great help in identifying the compressed and distorted collecting system. Because the cysts are deeply placed in the renal sinus, aggressive resection of the cyst wall should not be attempted; instead, marsupialization must be performed. At the end of the procedure, sterile methylene blue is injected through the preplaced ureteral catheter to identify any leak from the pelvicaliceal system.

When the cyst wall is excised, the edges and base of the residual cyst wall are fulgurated. The surgeon should be extremely cautious while fulgurating the base because it overlies renal parenchyma and is in close proximity to the pelvicaliceal system. To prevent recurrence, a pedicle of perirenal fat is placed in the residual cavity subsequent to excision or marsupialization and finally a drain is placed.

#### Transperitoneal Approach

#### Laparoscopic Nephrectomy Techniques

This approach was the preferred access for cases with unilateral giant kidney as it provides a better space and orientation, otherwise preoperative drainage of the hugely dilated kidney (Fig. 13.1) or intraoperative deflation using a percutaneous needle are needed to gain access to the retroperitoneum (Fig. 13.2) [17]. One needs



**Fig. 13.2** (a) CT with IV contrast showing bilateral giant hydronephrosis occupying the whole abdomen and displacing abdominal contents. (b, c) Antegrade pyelography showing arrest of the contrast at the pelviureteric junction (PUJ). Note the difference in the volume after insertion of the percutaneous nephrostomy (PCN). Right-sided nephrectomy was performed on the poorly functioning right kidney

to compress the flank until no more drainage noticed. The patient is placed in a shallow flank position (30° ipsilateral side up), and Veress needle insufflation is performed at the superior aspect of the umbilicus. A 12-mm laparoscopic port is placed at the umbilicus, and the remaining ports are placed under laparoscopic vision.

For the left side, a 10-mm to 12-mm port is placed just below the umbilicus along the midclavicular line, and a 5-mm port is placed in the midline between the xiphoid and the umbilicus. On the right side, a 12-mm upper midline and a 5-mm midclavicular line port are placed. An optional 5-mm port is placed along the anterior axillary line between the iliac crest and the costal margin to facilitate retraction of the liver or spleen.

Because of the large kidney underneath, care must be taken during trocar placement to prevent injury to the renal sac and other abdominal viscera. Once the colon



**Fig. 13.3** Gross specimen of a giant hydronephrotic kidney removed laparoscopically. Since the parenchyma was very thinned out, there was no need for morcellation as it was removed intact through one of the ports

is dissected off the kidney, the kidney is mobilized as far as possible before percutaneous deflation of the renal sac. During deflation, especially in a transperitoneal procedure, spillage of the renal contents into the peritoneum must be minimized. Traction on the renal sac extracorporeally facilitates further dissection at the renal hilum. In the transperitoneal approach, the renal sac is delivered through the lateral port near the more mobilized pole (generally the inferior pole). The remaining steps are similar to conventional transperitoneal laparoscopic nephrectomy. The kidney, once completely mobilized, can be delivered through the port used for retraction (Fig. 13.3) [2].

#### **Transperitoneal Laparoscopic Renal Cyst Decortication**

Although the risk of collecting system injury is rare for peripheral cysts, peripelvic cysts and cysts associated with adult polycystic kidney disease may impinge on the collecting system, thereby increasing the risk for injury. The optional placement of a ureteral catheter provides a route for injection of indigo carmine or methylene blue to facilitate identification of inadvertent collecting system injury. If the injury is identified intraoperatively and is large, then laparoscopic repair is advised. If the injury is small, then the ureteral catheter can be converted to an internal ureteral stent after completion of the laparoscopic procedure [21]. Initially, the procedure was performed using four ports, although simple cysts can be treated using three ports. In cases of ADPKD, four ports are invariably needed. The technique is similar to that previously described.

After incising the line of Toldt from the iliac vessels to the splenic or hepatic flexure, the colon is mobilized medially and Gerota's fascia is opened. The kidney does not need to be mobilized in its entirety for unroofing of a solitary cyst. In contrast, for ADPKD, the kidney is mobilized completely and the hilum exposed to provide optimal access to the maximum number of cysts.

The surface of the kidney overlying the cyst is dissected until a rim of normal renal parenchyma surrounding the cyst is exposed. For large cysts, dissection may

be facilitated by partially decompressing the cyst using an 18-gauge spinal needle placed percutaneously and guided by laparoscopic vision. The cyst wall is grasped and excised using electrocoagulating scissors until it is flush with the renal capsule. The cyst is then sent for histopathologic evaluation. Other energy devices, like an ultrasonic activated device such as the Ultracision<sup>®</sup> Harmonic Scalpel<sup>®</sup> (Ethicon Endo-Surgery, Inc., Cincinnati, OH) or an electrocautery device like LigaSure<sup>TM</sup> (ValleyLab, Boulder, CO) can be used. The base of the cyst is inspected for suspicious nodules or irregularities that are excised for biopsy with cup biopsy forceps.

Hemostasis is secured at the biopsy site and along the incised cyst wall with cautious use of electrocautery or the argon beam coagulator. Routine coagulation of the base of the cyst is discouraged because of the risk of collecting system injury. Perirenal fat or a tongue of omentum may be mobilized and placed into the cyst cavity to act as a wick to divert cyst fluid and prevent reaccumulation. For large cysts, a 7-mm suction drain is placed through a lateral port and is left in place for 1-2 days. As an alternative to mobilizing the colon to expose the kidney, some authors describe accessing a right renal cyst directly through a window in the mesocolon [22]. This approach, however, risks ischemic bowel injury, and the authors advise caution to avoid injury to the ileocolic and right colic arteries.

For peripelvic cysts, the renal hilum and pelvis are exposed. Identification of the cyst is facilitated by injection of indigo carmine stained saline into the renal pelvis through a ureteral catheter, with alternating filling and draining of the pelvis to distinguish it from the cyst. The cyst wall is incised without the use of electrocautery because of the close proximity to the renal vessels and collecting system. The use of laparoscopic ultrasound may help distinguish the cyst from the renal vein [23]. It is essential to fill the residual cavity by fixing peripelvic cyst is considered a contraindication to percutaneous sclerosis. These cysts can be managed effectively, although the procedure is more complex than decortication of simple renal cysts. The peripelvic cyst located near the hilum and pelvis requires more complicated, skillful, and challenging dissection [16].

For ADPKD, the objective is to unroof as many cysts as possible, or to be aspirated if excision of the cyst wall is precluded by the presence of surrounding renal parenchyma. A 7 mm suction drain is placed through a lateral port for 1–2 days. Because of extensive mobilization of the kidney, Elashry et al. advocate performing a nephropexy at the conclusion of the procedure [24].

# Miscellaneous Issues Related to Giant Hydronephrosis Nephrectomy

In a recent study, Laparoendoscopic single site (LESS) retroperitoneoscopic nephrectomy was completed in all patients with non-functioning giant hydrone-phrosis without conversion to open surgery and the additional placement of ports. The mean operative time was 98 min (range 77–146), and estimated blood loss was 45 mL (range 20–120). No major intraoperative complications occurred. The post-operative period was uneventful in all patients and the mean hospital stay was 3.4

days (range 2–7).LESS retroperitoneoscopic nephrectomy for giant hydronephrosis is technically feasible and safe for selected patients. The combination of conventional and prebent laparoscopic instruments represents an attractive option for retroperitoneoscopic LESS.

#### Conclusion

Laparoscopic surgery has almost replaced open surgery in benign nephrectomy as in hydronephrotic kidneys. But this subset of unusually and massively enlarged hydronephrotic kidneys are really challenging cases that need laparoscopic experience. Typically, a transperitoneal approach is more efficient. The key to success is to create enough space by decompressing the massively dilated kidney and respecting other organ anatomy. In regards to laparoscopic cyst removal, the authors prefer the transperitoneal approach (although both approaches can be performed), and cyst wall excision with different energy devices is mostly curative.

## References

- 1. Crooks KK, Hendren WH, Pfister RC. Giant hydronephrosis in children. J Pediatr Surg. 1979;14:844–50.
- Hemal AK, Wadhwa SN, Kumar M, Gupta NP. Transperitoneal and retroperitoneal laparoscopic nephrectomy for giant hydronephrosis. J Urol. 1999;162:35–9.
- Kissane JM. The morphology of renal cystic disease. In: Gardner Jr KD, editor. Cystic diseases of the kidney. New York: Wiley; 1976. p. 31–63.
- 4. Laucks SP Jr, McLachlen MS. Aging and simple cysts of the kidney. Br J Radiol. 1981;54:12-4.
- Holmberg G, Hietala SO. Treatment of simple renal cysts by percutaneous puncture and instillation of bismuth-phosphate. Scand J Urol Nephrol. 1989;23:207–12.
- 6. Rubenstein CS, Hulbert JC, Pharand D, Schuessler WW, Vancaille TG, Kavoussi LR. Laparoscopic ablation of symptomatic renal cysts. J Urol. 1993;150:1103–6.
- 7. Fontana D, Porpiglia F, Morra I, Destefanis P. Treatment of simple renal cysts by percutaneous drainage with three repeated alcohol injections. Urology. 1999;53:904–7.
- Hemal AK, Aron M, Gupta NP, Seth A, Wadhwa SN. The role of retroperitoneoscopy in the management of renal and adrenal pathology. BJU Int. 1999;83:929–36.
- 9. Hulbert JC, Shepard TG, Evans RM. Laparoscopic surgery for renal cystic disease. J Urol. 1992;147(suppl):443; [abstract 882].
- 10. Bosniak MA. The current radiological approach to renal cysts. Radiology. 1986;158:1-10.
- 11. Israel GM, Bosniak MA. An update of the Bosniak renal cyst classification system. Urology. 2005;66:484.
- O'Malley RL, Godoy G, Hecht EM, Stifelman MD, Taneja SS. Bosniak category IIF designation and surgery for complex renal cysts. J Urol. 2009;182:1091–5.
- Bosniak MA. The use of the Bosniak classification system for renal cysts and cystic tumors. J Urol. 1997;157:1852.
- 14. Wolf JS. Evaluation and management of solid and cystic renal masses. J Urol. 1998;159:1120–33.
- 15. Talukder BC, Chatterjee SC, Agarwal TN, De PP. Giant hydronephrosis. Br J Urol. 1979;51(4):322–3.
- 16. Hemal AK. Laparoscopic management of renal cystic disease. Urol Clin North Am. 2001;28:115–26.
- Ibrahim HM, Al-Kandari AM, Taqi A, et al. Etiology and management of adult giant hydronephrosis. Arab J Urol. 2008;6(2):21–5.

- 18. Chiang PH, Chen MT, Chou YH, Chiang CP, Huang CH, Chien CH. Giant hydronephrosis: report of 4 cases with review of the literature. J Formos Med Assoc. 1990;89:811–7.
- Harper JD, Shah SK, Baldwin DD, Moorhead JD. Laparoscopic nephrectomy for pediatric giant hydronephrosis. Urology. 2007;70:153–6.
- Rassweiler JJ, Seemann O, Frede T, Henkel TO, Aiken P. Retroperitoneoscopy: experiences with 200 cases. J Urol. 1998;160:1265–9.
- Pearle MS, Traxer O, Cadeddu JA. Renal cystic disease: laparoscopic management. Urol Clin North Am. 2000;27:661–73.
- 22. Leveillee RJ, Amaral J, Stein BS. Laparoscopic unroofing of a renal cyst via a mesocolonic window: a different approach. J Laparoendosc Surg. 1994;4:227–32.
- Hoenig DM, McDougall EM, Shalhav AL, Elbahnasy AM, Clayman RV. Laparoscopic ablation of peripelvic renal cysts. J Urol. 1997;158:1345–8.
- Elashry OM, Nakada SY, Wolf JS Jr, McDougall EM, Clayman RV. Laparoscopy for adult polycystic kidney disease: a promising alternative. Am J Kidney Dis. 1996;27:224–33.