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Gastric Adenocarcinoma: Reduction of Perioperative Mortality by Avoidance of Nontherapeutic Laparotomy

Brian R. Smith · Bruce E. Stabile

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Abstract National trends indicate a longstanding decline in gastric adenocarcinoma due presumably to a decreasing prevalence of *Helicobacter pylori* infection. Nonetheless, surgical outcomes continue to include relatively high morbidity and mortality rates owing to the advanced stage of disease encountered. We hypothesize that recent immigration patterns are responsible for a leveling off or even reversal of the declining incidence of gastric cancer associated with *H. pylori* infection. Furthermore, advances in preoperative tumor staging and nonoperative palliation currently permit better patient selection for operation with lower perioperative morbidity and mortality. A retrospective review of a consecutive case series at a public teaching hospital located in an area of high immigration was conducted that included all patients presenting, from 1995 through 2004, with gastric adenocarcinoma. For time comparison purposes, patients were divided into early (1995–1999) and recent (2000–2004) periods. There was no decline in the frequency of gastric adenocarcinoma among the study population over the 10 years. A total of 260 patients were treated of whom 137 (53%) underwent operation. The operation rate decreased and the gastric resection rate increased from the early period to the recent period as fewer incurable advanced stage (M1) patients underwent exploratory laparotomy and were palliated by nonoperative means. Perioperative morbidity and mortality rates also declined over time. Of the four total perioperative deaths, two followed 11 nontherapeutic laparotomies (18% mortality), whereas the only two additional deaths followed 122 curative or palliative laparotomies (2% mortality) (p=0.034). We conclude that in an area of high immigration there has been no decline in gastric adenocarcinoma rates over the past decade, and the marked reduction in perioperative mortality was due to near elimination of nontherapeutic laparotomy.

Keywords Gastric adenocarcinoma · Outcomes · Nontherapeutic laparotomy

Introduction

Worldwide, gastric adenocarcinoma ranks second only to lung cancer as the leading cause of cancer-related mortality. In the United States and other developed countries, both the incidence and mortality rates for gastric cancer have declined over the past more than half century. The most plausible explanation for these phenomena is the decreasing prevalence of *Helicobacter pylori* infection, a factor well established to be causative in the majority of cases.[1–6] In some areas of the United States, however, immigration of large populations harboring *H. pylori* may be reversing this trend and contributing to a resurgence of gastric cancer that will become more evident in the future. Despite the promise of gastric cancer prevention through the implementation of a widespread *H. pylori* eradication program, current management continues to rely primarily on effective surgical resection. Unfortunately, poor clinical outcomes because of typically advanced stage disease at presentation remain the rule. Aggressive gastrectomy with en bloc lymphadenectomy is most often required because of large primary tumors and associated lymph node metastases. The morbidity and mortality that follow operation remain substantial although recent refinements in preoperative staging, surgical techniques, perioperative intensive care,

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and nonoperative palliation have all served to improve clinical outcomes.

A relative paucity of scientific data has not diminished the widely held surgical opinion that nontherapeutic laparotomy is attended by inordinately high postoperative morbidity and mortality.[7] Avoidance of operations that fail to remove all grossly apparent tumor is of little therapeutic benefit except in instances where tumor complications such as bleeding, obstruction, perforation, or otherwise unmanageable pain are present. Therefore, it is important that patient selection for operation be optimized to minimize the avoidable adverse outcomes associated with nontherapeutic laparotomy.

In recent years, endoscopic ultrasonography (EUS) and laparoscopy have been employed in the preoperative staging of gastric adenocarcinoma. Both techniques have proven useful in improving the accuracy of staging and in the avoidance of unnecessary operation. Similarly, the availability of newer endoscopic interventions, including stent placement or laser ablation to relieve obstruction and argon beam photocoagulation to control bleeding, now obviate the need for many palliative gastrectomies.

The purposes of the present study were to verify the hypotheses that (1) in an area of high immigration gastric cancer is no longer declining in incidence; (2) nontherapeutic laparotomy is responsible for a disproportionate contribution to the morbidity and mortality attending operation for gastric cancer; and (3) advances in preoperative tumor staging and nonoperative palliation currently allow better patient selection for operation, a reduction in nontherapeutic laparotomies, and thus reductions in overall perioperative complication and death rates.

Materials and Methods

A 10-year retrospective review of all patients presenting to Harbor-UCLA Medical Center with gastric adenocarcinoma between 1995 and 2004 was conducted. The medical center is a large acute care urban public teaching hospital located in an area of high immigration in the southwest portion of Los Angeles County. To evaluate changes in demographics, surgical care, and outcomes over time, patients were grouped according to date of presentation. The early period patients included those who presented during the 5 years between January 1, 1995 and December 31, 1999, and the recent period patients included those who presented between January 1, 2000 and December 31, 2004.

Epidemiologic and clinical data were retrieved from the medical center cancer registry and individual patient medical records. Data recorded and analyzed included patient demographics, *H. pylori* testing results, perioperative tumor staging results, types of interventions employed, and early clinical outcomes. For the purposes of this study, the term "nontherapeutic laparotomy" refers to only those open abdominal operations that involved inspection and biopsy only, and excludes all those that involved tumor debulking, gastrointestinal bypass or decompression, management of perforation or bleeding, or other palliative procedures. The study was approved by the medical center's Institutional Review Board. Statistical analysis was performed using Student's *t* test, chi-square test, or Fisher's exact test as appropriate, and odds ratios with 95% confidence intervals were determined. Values of $p<0.05$ were considered statistically significant.

Results

A total of 260 patients were treated over 10 years. The number of patient presentations remained essentially constant over the early and recent periods (Table 1). There was a substantial decrease in the male to female ratio of patients such that the gender distribution became virtually equal during the recent period. Hispanic patients predominated and their proportion increased over the successive time periods. The proportions of Asian patients remained constant whereas those of African American and white patients were small and each decreased by over one-third. The average patient age increased significantly over the decade; however, fully 13% of patients were only 40 years of age or younger. Of the 76 (29%) patients tested for *H.*

Table 1 Patient Demographics

	Early Period 1995–1999	Recent Period 2000–2004	Odds Ratio (95% CI)	*p* Value
Patients	128	132	–	–
Male/Female	1.6/1	1.1/1	0.68 (0.42–1.12)	0.135
Hispanic, *n* (%)	62/128 (48)	76/132 (58)	1.44 (0.89–2.36)	0.172
Asian, *n* (%)	37/128 (29)	38/132 (29)	0.99 (0.58–1.70)	1.000
African American, *n* (%)	14/128 (11)	8/132 (6)	0.53 (0.21–1.30)	0.185
White, *n* (%)	15/128 (12)	10/132 (8)	0.62 (0.27–1.43)	0.297
H. pylori Infected, *n* (%)	31/38 (82)	30/38 (79)	0.85 (0.27–2.63)	1.000
Mean age (years)	54.2±2.56	58.7±2.55	–	0.018

pylori infection, the rate of positivity remained constant at approximately 80%.

Of the 128 total patients who presented in the early period, 76 (59%) underwent operation compared to only 61 of 132 (46%) in the recent period (Table 2). This decrease was because of improvements in patient selection for laparotomy afforded by more accurate tumor staging and by greater use of nonoperative palliation. The use of staging laparoscopy increased from 20 to 25% among patients deemed potentially curable based on preoperative physical examination, computed tomography (CT) scanning, and EUS results. Endoscopic palliation techniques, including stenting for obstruction and argon beam photocoagulation for bleeding, were utilized in 35% of eligible patients in the recent period compared to 21% in the early period. As a result, the operation rate for patients with distant metastatic (M1) disease decreased from 34 to 20% over the 10 years. In concert with these advances, the operative gastric resection rate and the frequency of complete (R_0) resection both increased, whereas the frequencies of microscopically and macroscopically incomplete (R_1 and R_2) resections decreased. Furthermore, the nontherapeutic laparotomy rate decreased from 11 to 5%. The overall postoperative morbidity rate declined from 29 to 23% and the mortality rate fell from 5 to 0%.

All of the four total perioperative deaths that occurred during the early period (Table 2). Two followed 11 nontherapeutic laparotomies (18% mortality), whereas the two other deaths attended 122 therapeutic laparotomies (2% mortality). The difference was statistically significant (Table 3). Thus, fully 50% of the perioperative mortality was related to nontherapeutic laparotomy. The morbidity rate after nontherapeutic laparotomy was 36%, compared to 26% after therapeutic laparotomy.

Discussion

The causal role of *H. pylori* infection in gastric carcinogenesis has been convincingly established by both prospective and multiple retrospective studies involving a number of large populations from around the world.[5,8] The dramatic declines in incidence and mortality rates for gastric adenocarcinoma that have been observed in many Westernized countries are now widely believed to be related to decreases in *H. pylori* infection rates over time.[1,2,9] In the United States, the frequency of gastric cancer appears to have nearly leveled off over the past 20 years, presumably because of the influx of large numbers of immigrants from countries where *H. pylori* infection is endemic.[10] Thus, both the tumor and its antecedent precancerous mucosal alterations are being imported to an extent that the incidence may at present, or in the near future, actually be increasing rather than decreasing. The data reported in this study is derived from an area of intense immigration by peoples from throughout the Pacific Rim and supports the notion that the incidence of gastric cancer may actually be rising in such areas at this time. At the very least, it is not falling. In the predominantly Hispanic and Asian population treated, the frequency of *H. pylori* infection among those tested was approximately 80%, a figure far greater than the less than 50% frequency found in the United States population as a whole in which the prevalence of gastric cancer remains very low.[6] It is yet to be determined whether the recent wave of increased immigration into the United States will result in an increased incidence of the disease nationwide.

In concert with the sustained frequency of presentation of gastric cancer patients in this study was an unexpected decline in the historic male predominance. The reason for

Table 2 Operations, Staging Results, and Operative Outcomes

	Early Period 1995–1999 (%)	Recent Period 2000–2004 (%)	Odds Ratio (95% CI)	*p* Value
Patients	128	132	–	–
Total operations	76/128 (59)	61/132 (46)	0.59 (0.36–0.96)	0.036
Staging laparoscopies	15/76 (20)	15/61 (25)	1.33 (0.59–2.99)	0.537
Endoscopic palliations	10/52 (21)	25/71 (35)	2.28 (0.98–5.31)	0.069
M1 cases operated	28/82 (34)	18/90 (20)	0.48 (0.24–0.96)	0.040
Total resections	61/76 (81)	57/61 (93)	3.50 (1.10–11.19)	0.044
R_0 resections	37/76 (49)	38/61 (62)	1.74 (0.88–3.46)	0.123
R_1 resections	10/76 (13)	4/61 (7)	0.46 (0.14–1.56)	0.262
R_2 resections	29/76 (38)	19/61 (31)	0.73 (0.36–1.50)	0.472
Nontherapeutic laparotomies	8/74 (11)	3/58 (5)	0.45 (0.11–1.78)	0.346
Perioperative complications	22/76 (29)	14/61 (23)	0.73 (0.34–1.59)	0.443
Perioperative deaths	4/76 (5)	0/61 (0)	0.00 (0.01–2.48)	0.129

Table 3 Comparison of Therapeutic and Nontherapeutic Laparotomy Results

	Therapeutic Laparotomy (%)	Nontherapeutic Laparotomy (%)	*p* Value
Patients	122	11	–
Resections	113 (93)	0 (0)	0.001
Nonresective palliations	9 (7)	0 (0)	1.000
Perioperative complications	32 (26)	4 (36)	0.488
Perioperative deaths	2 (2)	2 (18)	0.034

this change is unclear, but the observation is consistent with a similar gender equalization recently noted among patients with peptic ulcer disease treated at our institution.[11] This may represent simply a recent relative increase in female immigration into the area or an increased utilization of the public health care services by female immigrants. Regardless of the cause, it is noteworthy that both gastric adenocarcinoma and peptic ulcer are predominantly *H. pylori*-mediated diseases.

Because the majority of gastric cancer patients present with advanced stage disease, only a minority are curable.[12] Whenever possible, the surgical goal is complete extirpation of all macroscopic and microscopic tumor. Anything less is attended by a poor long-term prognosis as adjuvant therapies have generally been ineffective.[9,12] When curative resection is not possible, palliative gastrectomy or bypass may be indicated in selected patients. However, newer endoscopic techniques such as stenting, laser ablation, and argon beam photocoagulation are likely eliminating the need for laparotomy in many such cases. Equally important at present is the capability for more accurate preoperative staging afforded by minimally invasive techniques that obviate nontherapeutic laparotomy when complete resection is not possible and palliative laparotomy is not needed.[13,14] Whereas perioperative serious morbidity and mortality rates associated with therapeutic laparotomy for gastric cancer are approximately 20 and 3%, respectively, those for nontherapeutic laparotomy for unresectable disease are substantially higher at about 35 and 20%, respectively.[9,12,15,16] Whenever possible, avoidance of nontherapeutic laparotomy in favor of nonoperative primary therapy such as chemoradiation in combination with endoscopic palliation is clearly desirable. Nonresective palliative operations and exploratory laparotomy for disease staging with the unlikely hope of curative resection are now largely contraindicated. In addition to the morbidity and mortality, pain and suffering, and costs of nontherapeutic laparotomy, the postoperative recovery time often delays the initiation of palliative therapy.[17–19]

The results from the present study indicate that 13% fewer patients underwent operation during the latter half of the decade. The decrease occurred in the context of increased use of staging laparoscopy and endoscopic palliation. These findings suggest that the goal of improving patient selection for laparotomy and maximizing the use of endoscopic palliative techniques did allow avoidance of noncurative abdominal operations. This is evidenced by a 13% increase in curative (R_0) operations from the early period to the recent period whereas the operation rate for patients with metastatic (M1) disease decreased by 14% and the nontherapeutic laparotomy rate decreased from 11 to 5%.

The reduction in nontherapeutic laparotomies during the recent period did have a salutary effect on overall perioperative morbidity and mortality, as predicted. The morbidity rate fell by 6% and mortality disappeared with no deaths among the 61 operated patients during the recent period. The importance of the contribution of nontherapeutic laparotomy to the poor outcomes associated with gastric cancer surgery is underscored by the 36% morbidity and 18% mortality that attended 11 nontherapeutic operations. The latter figure was statistically significantly lower than the 2% death rate for 122 therapeutic laparotomies.

The avoidance of nontherapeutic laparotomy has derived largely from the improved preoperative staging allowed by EUS and diagnostic laparoscopy. Whereas the accuracy of CT scanning for tumor (T) staging has been only 20–30%, that for EUS has been 80–90%.[20] For nodal (N) staging, the

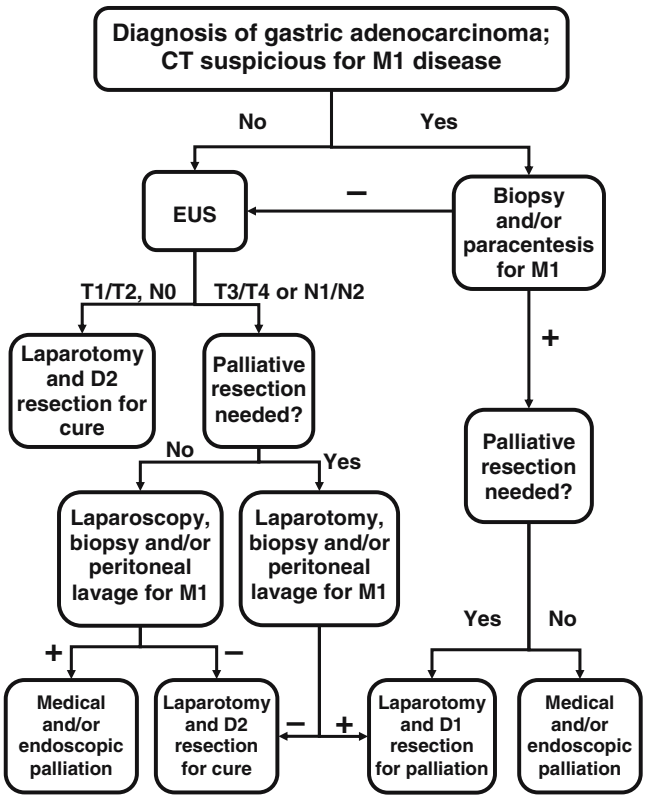

Figure 1 Algorithm for management of biopsy-proven gastric adenocarcinoma.

figures have been 30–40% and 70–80% for CT and EUS, respectively.[21] Small-volume malignant ascites and occult left lobe liver metastases (M1 disease) not appreciated by CT have also been detected by EUS. In addition, EUS-guided fine needle aspiration has been used to obtain cytologic confirmation of malignant cells from lymph nodes, liver, and ascitic fluid, thus obviating the need for more invasive means to establish incurability.[22]

As there is at present no indirect imaging technique sensitive enough to detect occult peritoneal carcinomatosis, laparoscopy before laparotomy permits minimally invasive visualization and biopsy of minute metastases and aspiration of small volume ascites or peritoneal lavage fluid for immediate cytologic examination.[23] Laparoscopic ultrasonography can further enhance staging by detection of liver metastases less than 1 cm and nonresectable distant lymph node metastases.[14] The collective experience suggests that laparoscopic staging can alter the clinical stage in up to one-third of patients. Use of these techniques has permitted operative intervention for gastric cancer to become more precise, with specific selection of those patients more likely to benefit from laparotomy and aggressive gastric resection with curative intent.

Based on the experience reported in this study, an algorithm for the staging and treatment of gastric cancer patients has been devised (Fig. 1). The algorithm emphasizes the sequential use of EUS and laparoscopy for staging to determine the appropriateness of gastric resection with aggressive D2 lymphadenectomy for potentially curable tumors vs limited D1 lymphadenectomy for incurable cases that require palliative resection vs medical and/or endoscopic palliative interventions. Laparoscopy with biopsy and/or peritoneal lavage for cytology is recommended for patients found on EUS to have T3 and T4 primary tumors or positive lymph node involvement as the incidence of occult M1 disease is substantial. The algorithm is intended to minimize nontherapeutic laparotomy and the associated high perioperative morbidity and mortality demonstrated in this study.

Conclusions

This study examined the changing demographics of gastric cancer in an area of high immigration and evaluated the detrimental outcomes associated with nontherapeutic laparotomy. Our data indicate that (1) there has been no decline in the frequency of the disease over the past decade; (2) based on a limited sampling, the *H. pylori* infection rate appears to be very high; (3) the historic male predominance has virtually disappeared; (4) improvements in preoperative staging and nonoperative palliation have allowed for a substantial reduction in the nontherapeutic laparotomy rate; and (5) the marked decrease in overall perioperative mortality is primarily due to obviation of the excessive mortality attending nontherapeutic laparotomy.

References

1. Howson CP, Hiyama T, Wynder EL. The decline in gastric cancer: Epidemiology of an unplanned triumph. Epidemiol Rev 1986;8:1–27.
2. Sipponen P. *Helicobacter pylori* infection—a common worldwide environmental risk factor for gastric cancer? Endoscopy 1992;24:424–427.
3. Sipponen P. *Helicobacter pylori*: A cohort phenomenon. Am J Surg Pathol 1995;19(Suppl 1):S30–S36.
4. Imrie C, Rowland M, Bourke B, Drumm B. Is *Helicobacter pylori* infection in childhood a risk factor for gastric cancer? Pediatrics 2001;107:373–380.
5. Huang JQ, Sridhar S, Chen Y, Hunt RH. Meta-analysis of the relationship between *Helicobacter pylori* seropositivity and gastric cancer. Gastroenterology 1998;114:1169–1179.
6. Stabile BE, Smith BR, Weeks DL. *Helicobacter pylori* infection and surgical disease—Part II. Curr Probl Surg 2005;42:791–862.
7. Siewert JR, Böttcher K, Stein H, Roder JD. Relevant prognostic factors in gastric cancer. Ten-year results of the German Gastric Cancer Study. Ann Surg 1998;228:449–461.
8. Uemura N, Okamoto S, Yamamoto S, Matsumura N, Yamaguchi S, Yamakido M, et al. *Helicobacter pylori* infection and the development of gastric cancer. N Engl J Med 2001;345:784–789.
9. Hohenberger P, Gretschel S. Gastric cancer. Lancet 2003;362:305–315.
10. Stabile BE, Smith BR, Weeks DL. *Helicobacter pylori* infection and surgical disease—Part I. Curr Probl Surg 2005;42:743–790.
11. Smith BR, Stabile BE. Emerging trends in peptic ulcer disease and damage control surgery in the H. pylori era. The American Surgeon 2005;71:797–801.
12. Lim L, Michael M, Mann GB, Leong T. Adjuvant therapy in gastric cancer. J Clin Oncol 2005;25:6220–6232.
13. Lee YT, Ng EKW, Hung LCT, Chung SCS, Ching JYL, Chan WY, et al. Accuracy of endoscopic ultrasonography in diagnosing ascites and predicting peritoneal metastases in gastric cancer patients. Gut 2005;54:1541–1545.
14. Conlon KC, Karpeh MS Jr. Laparoscopy and laparoscopic ultrasound in the staging of gastric cancer. Semin Oncol 1996;23:347–351.
15. Asencio F, Aguilo J, Salvador JL, Villar A, De la Morena E, Ahamad M, et al. Video-laparoscopic staging of gastric cancer: A prospective multicenter comparison with noninvasive techniques. Surg Endosc 1997;11:1153–1158.
16. Jansen EPM, Boot H, Verheij M, van de Velde CJH. Optimal locoregional treatment in gastric cancer. J Clin Oncol 2005;23:4509–4517.
17. Valen B, Viste A, Haugstvedt T, Elide GE, Soreido O. Treatment of stomach cancer, a national experience. Br J Surg 1988;75:708–712.
18. Viste A, Haugstvedt T, Eide E, Soreide O. Postoperative complications and mortality after surgery for gastric cancer. Ann Surg 1988;207:7–213.
19. Tsioulias GJ, Wood TF, Chung MH, Morton DL, Bilchik A. Diagnostic laparoscopy and laparoscopic ultrasonography opti-

mize the staging and respectability of intraabdominal neoplasms. Surg Endosc 2001;15:1016–1019.
20. Pollack BJ, Chak A, Sivak MV Jr. Endoscopic ultrasonography. Semin Oncol 1996;23:336–346.
21. Caletti G, Ferrari A, Brocchi E, Barbara L. Accuracy of endoscopic ultrasonography in the diagnosis and staging of gastric cancer and lymphoma. Surgery 1993;113:14–27.
22. Chang KJ, Katz KD, Durbin TE, Erickson RA, Bulter JA, Lin F, et al. Endoscopic ultrasound-guided fine-needle aspiration. Gastrointest Endosc 1994;40:694–699.
23. Stell DA, Carter CR, Steward I, Anderson JR. Prospective comparison of laparoscopy, ultrasonography and computed tomography in the staging of gastric cancer. Br J Surg 1996; 83:1260–1262.

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What to Expect in the Excluded Stomach Mucosa after Vertical Banded Roux-en-Y Gastric Bypass for Morbid Obesity

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Abstract Mucosal alterations after vertical banded Roux-en-Y gastric bypass have not been clearly evaluated. The aim of this paper was to analyze the histological findings and the presence of *Helicobacter pylori* in the excluded stomach. Forty consecutive patients who underwent Roux-en-Y gastric bypass longer than 36 months were selected for double-balloon enteroscopy. The excluded stomach was reached in 35/40 patients (88%). Morphological alterations were analyzed through hematoxilin and eosin and the presence of *H. pylori* was confirmed with Giemsa staining. Thirty patients (86%) were female, and the mean age was 43 years old. The mean postoperative time was 78 months (36–110 months). Histologically, all patients had chronic gastritis in the bypassed stomach, with pangastritis in 33/35 (94%). Five cases (5/35, 14%) presented atrophy and four of them also had intestinal metaplasia. *Helicobacter pylori* was detected in 7/35 (20%) of the excluded stomach and in 12/35 (34%) of the functional pouch. All patients positive for *H. pylori* in the excluded stomach were also positive in the functional pouch, $p=0.0005$. *Helicobacter pylori* is still present in the excluded stomach after Roux-en-Y gastric bypass and might be considered for treatment. Histological findings indicated high prevalence of atrophy and intestinal metaplasia in this selected population.

Keywords Vertical banded Roux-en-Y gastric bypass · *Helicobacter pylori* · Double-balloon enteroscopy

Introduction

Morbid obesity has increased in developed countries and achieved epidemic public health status.[1] The National Institutes of Health consensus conference in 1991 concluded that "surgical therapy offers the best long-term approach to treat morbid obesity, and is probably the most effective therapy to cure type 2 diabetes".[2] Surgical treatments are now largely performed on these patients, and vertical banded Roux-en-Y gastric bypass using a long limb has become a common and gold standard procedure.

Mucosal alterations after vertical banded Roux-en-Y gastric bypass have not been clearly evaluated because the excluded stomach is not easily reached by conventional endoscopy. However, gastritis, gastric ulcer, intestinal metaplasia, and few cases of gastric cancer developed in the bypassed stomach have been described.[3–8]

Helicobacter pylori colonization in the excluded stomach and its influence has not been evaluated after surgical

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treatment in obese patients. Endoscopic assessment of the excluded stomach may be performed safely utilizing a double-balloon enteroscope, as previously reported.[9] Thus, the aim of this investigation was to analyze the mucosal alterations and the presence of *H. pylori* in the excluded stomach and in the gastric stump epithelium (functional pouch) after gastric bypass for morbid obesity.

Materials and Methods

Clinical Material

Informed consent was obtained from all patients. The study was approved by the Hospital Ethics Committee of the University of São Paulo School of Medicine, Brazil. Double-balloon enteroscopy was performed in 40 consecutive patients who underwent vertical banded Roux-en-Y gastric bypass longer than 36 months (Fig. 1). The surgical technique incorporates a small pouch along the lesser curvature of the stomach, an outlet restricted by a non-distensible band and a Roux-en-Y gastric bypass.[10] To reach the excluded stomach, the enteroscope had to be advanced through the esophagus (40 cm), proximal gastric pouch (5 cm), Roux-en-Y jejunum loop (usually with 100 to 150 cm), biliopancreatic jejunal limb (50 to 75 cm from the Treitz' ligament), and duodenum (20 cm).

The patients underwent endoscopic examination with multiple biopsies of the gastric stump mucosa (four) and of the excluded stomach mucosa (four at the body and four at the antrum) during surveillance endoscopy. Four duodenal biopsies were also performed. The excluded stomach of five patients who underwent double-balloon enteroscopy could not be achieved by this method; therefore, the analysis was done in 35 patients who completed the examination.

Histopathologic Evaluation

Endoscopic biopsies were cut from tissue blocks fixed in formalin and embedded in paraffin. Sections 4-μm thick were stained by hematoxylin and eosin and modified Giemsa staining.

The grade of inflammation in the epithelium of the specimens obtained through endoscopic biopsies was also categorized into four levels (0=none; 1=mild; 2=moderate, and 3=severe). The presence or absence of atrophy and/or intestinal metaplasia in the gastric mucosa was also noted. The colonization of the tissues by *H. pylori* was detected using a modified Giemsa staining. *Helicobacter pylori* colonization was recorded solely as present or absent. All sections were examined independently by two authors in a blind fashion.

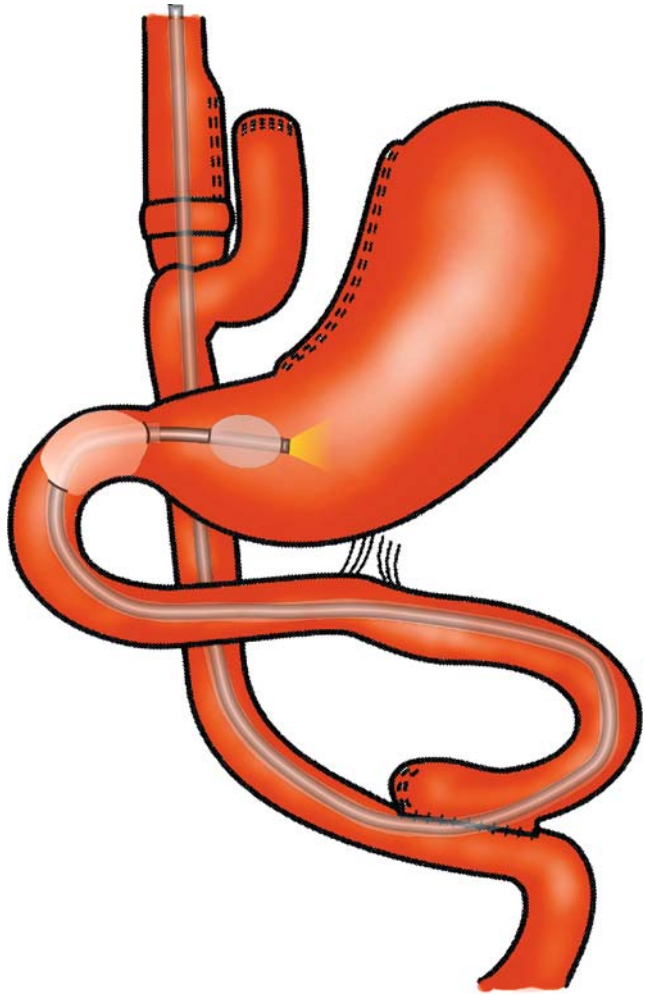

Figure 1 Schematic representation of vertical banded Roux-en-Y gastric bypass.

Statistical Analysis

Data are expressed as mean ± standard deviation. The data were analyzed using unpaired Student *t* test, Chi-square, and Fisher's exact test, with two-tailed *p* value <0.05 considered significant.

Results

Thirty patients (85.7%) were female and the mean age was 43.4 years old (22–61 years-old). The mean postoperative time was 77.6 months (range 36–110 months). Eight cases (8/35, 22.8%) presented endoscopically normal bypassed stomach. According to Sydney's classification, 4/35 (11.4%) patients had body or antrum gastritis, including three atrophic and one erythematous. Twenty-three patients (23/35, 65.7%) had pangastritis, including nine erythematous, nine flat erosive, and five atrophic. Two patients 2/35

(5.7%) also had endoscopic suspicious areas of intestinal metaplasia, which were histologically confirmed.

Histologic Alterations

All patients had chronic gastritis in the bypassed stomach, with pangastritis in 33 out of 35 (94.3%). Five cases (5/35, 14.3%) presented atrophy (Fig. 2) and four of them also had intestinal metaplasia (Fig. 3). Mild gastritis was detected in 23 out of 35 (65.7%) and moderate gastritis in 12 out of 35 (34.3%). No severe gastritis was found.

Helicobacter pylori Colonization

Helicobacter pylori was detected in 7 out of 35 (20%) cases of the excluded stomach, and was positive in the antrum in all of them and in the body in four patients. The microorganism was seen within the mucus layer overlying the mucosa or along the luminal surfaces of the mucosal epithelial cells, the mucous neck cells, and the epithelial cells in the most preserved gastric pits (Fig. 4). Moreover, *H. pylori* was positive in the gastric stump in 12 out of 35 (34.3%). All patients that were positive for *H. pylori* in the excluded stomach were also positive in the gastric functional stump, $p=0.0005$ (Table 1).

Helicobacter pylori and Inflammation

Severity of gastritis of the excluded stomach was associated to the presence of *H. pylori*, $p=0.02$ (Table 1). The pattern of inflammation was usually in the form of active chronic gastritis characterized by the presence of acute and chronic inflammatory cells in the lamina propria, commonly accompanied by intraepithelial infiltration of eosinophils and lymphocytes (Fig. 5).

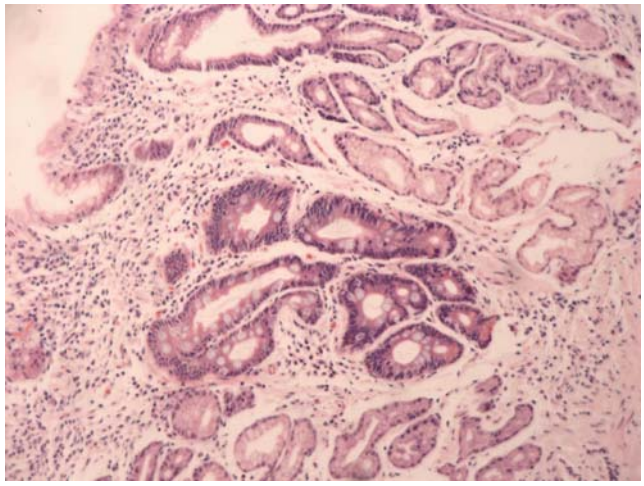

Figure 3 Foci of intestinal metaplasia can be noted in the excluded stomach (hematoxilin and eosin, ×20).

Helicobacter pylori and Histologic Alterations

Helicobacter pylori was not detected directly in the areas of the slides showing intestinal metaplasia. However, the microorganisms could be seen in areas immediately adjacent, areas showing gastric type columnar cells and surface mucus cells. There was a reduction in the severity of the infection with the progression of gastric damage.

Discussion

Bariatric surgery is the most successful means of salvage currently available for morbidly obese patients. It dramatically improves the quality-of-life expectancy in adults, as well as in children and adolescents, and may now be considered as a pediatric maneuver in some countries.[1]

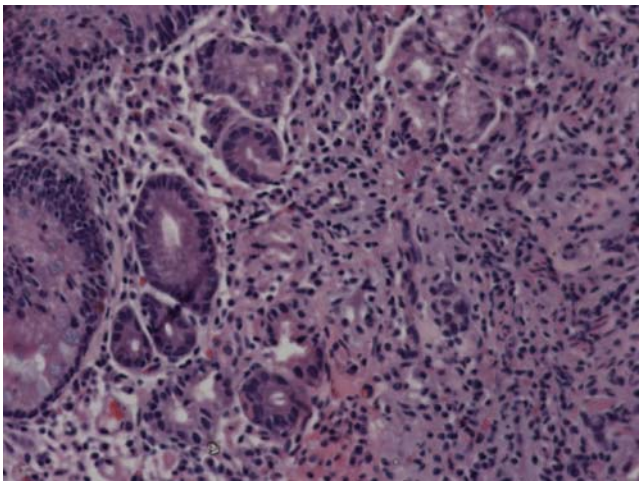

Figure 2 Atrophy of the epithelial glands can be detected in the excluded stomach (hematoxilin and eosin, ×20).

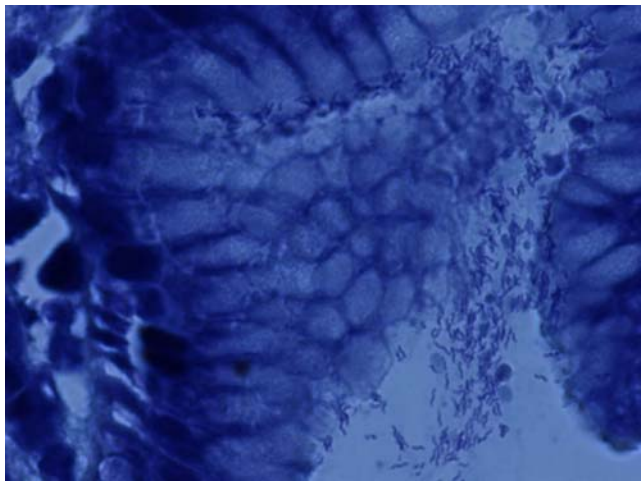

Figure 4 *Helicobacter pylori* colonization in the gastric pits of the excluded stomach (modified Giemsa, ×100).

Table 1 Distribution of *Helicobacter pylori* in the Excluded Stomach of 35 Patients Who Underwent Roux-en-Y Gastric Bypass for Morbid Obesity

Helicobacter pylori				
		Positive (%)	Negative (%)	*p* Value
Gastritis—antrum				0.01
	Mild	2 (5.7)	22 (62.9)	
	Moderate	5 (14.3)	6 (17.1)	
Gastritis—body				0.003
	Mild	0	23 (65.7)	
	Moderate	4 (11.4)	8 (22.9)	
Gastritis—functional pouch				0.02
	Mild	2 (5.7)	21 (60)	
	Moderate	5 (14.3)	7 (20)	
Helicobacter pylori—Functional pouch				0.0005
	Positive	7 (20)	5 (14.3)	
	Negative	0	23 (65.7)	

Fisher's exact test

Therefore, it is noteworthy to study the epithelium of the excluded stomach for the presence of mucosal changes and for the presence of *H. pylori* because these patients will remain with their stomachs *in locu*, but excluded of the food transit for decades.

Few cases of gastric cancer in the bypassed stomach have been described after Roux-en-Y gastric bypass for morbid obesity. Some authors even recommend distal subtotal gastrectomy when a suspicious lesion is present preoperatively, due to the difficulty of endoscopic gastric surveillance. [11,12]

Intraoperative endoscopy, endoscopy through ultrasound-guided percutaneous gastrostomy, and virtual CT gastroscopy have been performed as alternatives to diagnose lesions of the excluded stomach[13–15] because conventional endoscopy does not normally reach the excluded stomach. The new technique of push-and-pull enteroscopy using the double-balloon enteroscope described by Yamamoto et al. in 2001[16] permits deep intubation of the small intestine. The endoscope can be advanced through the small bowel by pushing it through the overtube and the loops can be easily reduced by gentle withdrawal of the endoscope while the balloons are inflated.[17]

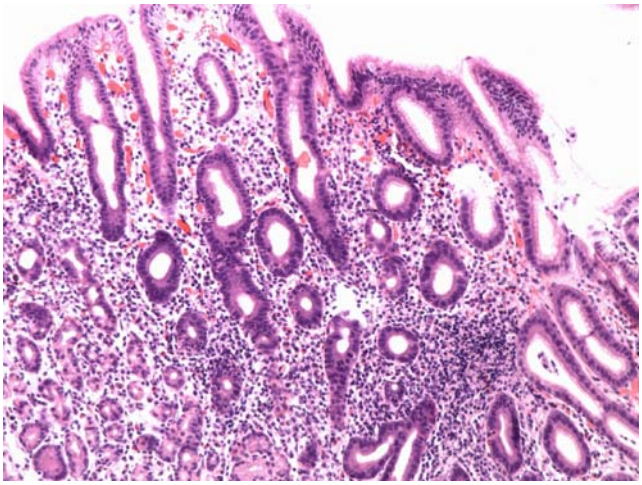

Figure 5 Chronic inflammatory infiltrate in the epithelium of the excluded stomach (hematoxilin and eosin, ×20).

Sakai et al., in 2005, [9] first described the double-balloon enteroscopy in the evaluation of the excluded stomach. With double-balloon enteroscopy, the excluded stomach could be reached and visualized endoscopically. Biopsies could also be performed.

Sundbom et al., in 2002,[18] found scintigraphic evidence of duodenogastric bile reflux in 36% of the patients who underwent Roux-en-Y gastric bypass, meaning that more than one third of such patients have their excluded stomach exposed to the potential deleterious effects of bile.

This investigation showed clearly that some patients may present with mucosal alterations, such as chronic atrophic gastritis and intestinal metaplasia. These data have to be interpreted with caution, especially because the number of young patients who have undergone surgical treatment has increased rapidly. The gastric bypassed stomach with duodenal reflux of bile and pancreatic secretions, without any buffering for the food intake, may work as in postgastrectomy patients,[19] and they will have time enough to damage the gastric epithelium, probable augmenting the gastric cancer risk.

Our results demonstrated that *H. pylori* is still indeed present in the excluded stomach many years after gastric bypass surgery. It is most commonly identified in biopsies from the antrum, but also occurred in the body region, and its presence was associated with an increase in inflammation. All positive *H. pylori* patients in the excluded stomach were also positive in the gastric functional stump. The concurrence between *H. pylori* infection in the functional pouch and in the excluded stomach indicates that there is no need for *H. pylori* detection in the excluded stomach.

Recolonization of the excluded gastric remnant may occur from recrudescence of infection, suppressed by the presence of bile in the stomach, with the same strain of *H. pylori*, as occurs after unsuccessful antibiotic therapy.[20] The colonization of the functional pouch may occur as mentioned above or, alternatively, by reinfection with another strain. The likelihood that this bacterium could not be adequately treated before the gastric bypass, and may persist for years following surgery in the excluded stomach, suggests that it could play a major role in the progression of mucosal changes following gastric bypass.

The finding that *H. pylori* colonization of the resected stomach has a synergistic effect on gastric mucosal cell proliferation is of particular interest.[21] The combination of

H. pylori and bile reflux seems to engender greater epithelial damage with an increased proliferative response.[22–24] The changes are not restricted to patients who have had surgery to their stomachs.[22] The presence of these agents together in the intact stomach has also been reported to have a synergistic effect on the development of intestinal metaplasia.[22] Thus, *H. pylori* may increase even more the gastric cancer risk in postgastrectomy patients, who commonly have a high prevalence of precursor lesions such as chronic atrophic gastritis, intestinal metaplasia, and dysplasia.[24]

Therefore, *H. pylori* is still present in the excluded stomach after vertical banded Roux-en-Y gastric bypass and might be considered for treatment. Gastric bypassed patients may benefit from receiving bacterial eradication therapy if they are *H. pylori*-positive in the endoscopic biopsies of the functional pouch because all positive cases of the excluded stomach were also positive in the functional pouch. Histological findings indicated high prevalence of atrophy and intestinal metaplasia in this selected population.

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References

1. Weber M, Clavien PA. Bariatric surgery—a successful way to battle the weight crisis. Br J Surg 2006;93:259–260.
2. Consensus Development Conference Panel. NIH Conference. Gastrointestinal surgery for severe obesity. Ann Intern Med 1991; 115, 956–961.
3. Raijman I, Strother SV, Donegan WL. Gastric cancer after gastric bypass for obesity. Case report. J Clin Gastroenterol 1991;13 (2):191–194.
4. Lord RV, Edwards PD, Coleman MJ. Gastric cancer in the bypassed segment after operation for morbid obesity. The Australian and New Zealand Journal of Surgery 1997;67 (8):580–582.
5. Voellinger DC, Inabnet WB. Laparoscopic Roux-en-Y gastric bypass with remnant gastrectomy for focal intestinal metaplasia of the gastric antrum. Obes Surg 2002;12(5):695–698.
6. Khitin L, Roses RE, Birkett DH. Cancer in the gastric remnant after gastric bypass: A case report. Curr Surg 2003;60(5):521–523.
7. Escalona A, Guzman S, Ibanez L, Meneses L, Huete A, Solar A. Gastric cancer after Roux-en-Y gastric bypass. Obes Surg 2005;15 (3):423–427.
8. Trincado MT, del Olmo JC, Garcia Castano J, Cuesta C, Blanco JI, Awad S, Carbajo MA. Gastric pouch carcinoma after gastric bypass for morbid obesity. Obes Surg 2005;15(8):1215–1217.
9. Sakai P, Kuga R, Safatle-Ribeiro AV, Faintuch J, Gama-Rodrigues JJ, Ishida RK, Furuya Jr CK, Yamamoto H, Ishioka S. Is it feasible to reach the bypassed stomach after Roux-en-Y gastric bypass for morbid obesity? The use of the double-balloon enteroscope. Endoscopy 2005;37:566–569.
10. Capella JF, Capella RF. An assessment of vertical banded gastroplasty-Roux-en-Y gastric bypass for the treatment of morbid obesity. Am J Surg 2002;183(2):117–123.
11. Csendes A, Burdiles P, Papapietro K, Diaz JC, Maluenda F, Burgos A, Rojas J. Results of gastric bypass plus resection of the distal excluded gastric segment in patients with morbid obesity. J Gastrointest Surg 2005;9(1):121–131.
12. Madan AK, Lanier BJ, Tichansky DS, Ternovits CA. Laparoscopic Roux-en-Y gastric bypass with subtotal gastrectomy. Obes Surg 2005;15(9):1332–1335.
13. Sundbom M, Nyman R, Hedenstrom H, Gustavsson S. Investigation of the excluded stomach after Roux-en-Y gastric bypass. Obes Surg 2001;11(1):25–27.
14. Braley SC, Nguyen NT, Wolfe BM. Late gastrointestinal hemorrhage after gastric bypass. Obes Surg 2002;12(3):404–407.
15. Silecchia G, Catalano C, Gentileschi P, Elmore U, Restuccia A, Gagner M, Basso N. Virtual gastroduodenoscopy: A new look at the bypassed stomach and duodenum after laparoscopic Roux-en-Y gastric bypass for morbid obesity. Obes Surg 2002;12(1): 39–48.
16. Yamamoto H, Sekine Y, Sato Y, Higashizawa T, Miyata T, Iino S, Ido K, Sugano K. Total enteroscopy with a nonsurgical steerable double-balloon method. Gastrointest Endosc 2001;53:216–220.
17. Yamamoto H, Kita H, Sunada K, Hayashi Y, Sato H, Yano T, Iwamoto M, Sekine Y, Miyata T, Kuno A, Ajibe H, Ido K, Sugano K. Clinical outcomes of double-balloon endoscopy for the diagnosis and treatment of small-intestinal diseases. Clin Gastroenterol Hepatol 2004;2:1010–1016.
18. Sundbom M, Hedenstrom H, Gustavsson S. Duodenogastric bile reflux after gastric bypass: A cholescintigraphic study. Dig Dis Sci 2002;47:1891–1896.
19. Safatle-Ribeiro AV, Ribeiro Jr. U, Reynolds JC. Gastric stump cancer: what is the risk? Dig Dis 1998;16:159–168.
20. Alam K, Schubert TT, Bologna SD, Ma CK. Increased density of *Helicobacter pylori* on antral biopsy is associated with severity of acute and chronic inflammation and likelihood of duodenal ulceration. Am J Gastroenterol 1992;87(4):424–428.
21. Lynch DAF, Mapstone NP, Clarke AM T, Jackson P, Dixon MF, Quirke P, Axon ATR. Cell proliferation in the gastric corpus in *Helicobacter pylori* associated gastritis and after gastric resection. Gut 1995;36:351–353.
22. Sobala GM, O'Connor HJ, Dewar EP, King RFG, Axon ATR, Dixon MF. Bile reflux and intestinal metaplasia in gastric mucosa. J Clin Pathol 1993;46:235–240.
23. Safatle-Ribeiro AV, Ribeiro Jr. U, Clarke MR, Sakai P, Gama-Rodrigues JJ, Safatle NF, Garrido Jr. AB, Reynolds JC. Relationship between persistence of *Helicobacter pylori* and dysplasia, intestinal metaplasia, atrophy, inflammation, and cell proliferation following partial gastrectomy. Dig Dis Sci 1999;44:243–252.
24. Correa P. Human gastric carcinogenesis: a multistep and multifactorial process—first American Cancer Society award lecture on cancer epidemiology and prevention. Cancer Res 1992;52:6735–6740.

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Clostridium difficile Enteritis: An Early Postoperative Complication in Inflammatory Bowel Disease Patients After Colectomy

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Emily T. Carman · William J. Peppard

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Abstract *Clostridium difficile*, the leading cause of hospital-acquired diarrhea, is known to cause severe colitis. *C. difficile* small bowel enteritis is rare (14 case reports) with mortality rates ranging from 60 to 83%. *C. difficile* has increased in incidence particularly among patients with inflammatory bowel disease. This case series of six patients from 2004 to 2006 is the largest in the literature. All patients received antibiotics before colectomies for ulcerative colitis and developed severe enteritis that was *C. difficile* toxin positive. Three patients underwent ileal pouch anal anastomosis and loop ileostomy. Four of the six patients had *C. difficile* colitis before colectomy. Presenting symptoms were high volume watery ileostomy output followed by ileus in five of six patients. Four of the six patients presented with fever and elevated WBC. Five of the six developed complications requiring further surgery or prolonged hospitalization. Patients were treated with intravenous hydration and metronidazole then converted to oral metronidazole and/or vancomycin. None of the patients died. A high suspicion of *C. difficile* enteritis in patients with inflammatory bowel disease and history of *C. difficile* colitis may lead to more rapid diagnosis, aggressive treatment, and improved outcomes for patients with *C. difficile* enteritis.

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Keywords *Clostridium difficile* · Ileal pouch anal anastomosis · Inflammatory bowel disease · Surgery · Enteritis · Colectomy

Introduction

Clostridium difficile is known to cause severe colitis and is the leading cause of hospital-acquired diarrhea in the United States. However, *C. difficile*-associated small bowel enteritis is a rare complication with only 14 case reports in the literature from 1980 to 2006. Twelve of the 14 reports were single cases and one report was a series of two patients. Published mortality rates range from 60 to 83%. *C. difficile* has increased in incidence particularly among patients with inflammatory bowel disease (IBD). We report the largest case series of six patients with *C. difficile* enteritis. All of these patients were treated at our tertiary IBD referral center from 2004–2006. *C. difficile* toxins were detected in the ileal effluent of all patients in the early postoperative time frame of <90 days. None of our patients had a colon. We report the demographics, presentation, management, and disposition in this cohort of patients.

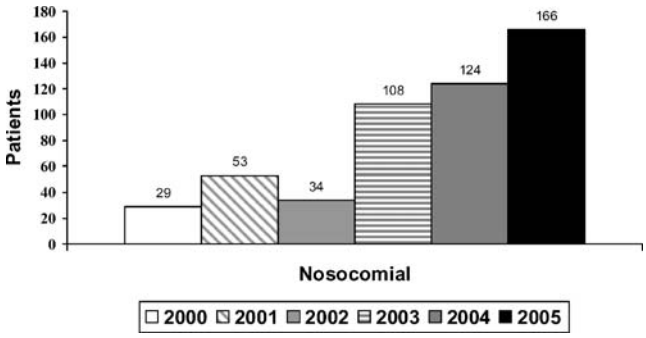

Figure 1 Positive toxin assays from inpatients are demonstrated in this bar graph. There was a marked increase in the number of positive assays over the last 6 years. A waterless hand-washing system was introduced into the hospital in 2003 and may account for some of the increase in positive toxin assays.

Materials and Methods

A review was performed of consecutive patients followed at a tertiary IBD Center between 2004 and 2006. Patients undergoing a colectomy were eligible for analysis. Fifty-one colectomies were performed during the study period at our hospital. Twenty-one of the 51 colectomies were performed for inflammatory bowel disease. Of these, twenty colectomies were performed for ulcerative colitis and one colectomy was performed for Crohn's colitis. All patients undergoing a colectomy for inflammatory bowel disease were on a single surgical team.

The cytotoxin assay is considered the gold standard. However, the 1- to 2-day turn around time was not practical for the inpatient setting.[1] In place of the cytotoxin assay, an ELISA test specific for toxin A was utilized before 2002 at our center due to its ease of use and rapid turn around time of only a few hours.[2] In 2002, the detection method of the ELISA test was changed to detect both toxins A and B.[3] This change in detection method was in response to evidence indicating that assays capable of detecting both toxins were more effective than assays capable of detecting only one toxin.

In 2003, a waterless, chlorhexidine and alcohol-based hand washing system was introduced into our hospital.

The Medical College of Wisconsin's human research review committee approved this study.

Results

Over the previous 5 years, the rate of *C. difficile* diagnosed at this center has increased dramatically (Fig. 1). These values represent all inpatients with positive toxin assays, which include toxin positive patients at hospital entry and hospitalized patients who convert to a positive toxin status.

After total abdominal colectomy and ileostomy, the six patients identified developed *C. difficile* enteritis. All received a dose of preoperative antibiotics. Three patients underwent ileal pouch anal anastomosis and loop ileostomy. All six patients had a final pathology of ulcerative colitis. Five of the six patients are female and one is male. Before colectomy, four of the six patients had *C. difficile* colitis superimposed upon ulcerative colitis. The average age of our patients was 35.3 years with an age range from 20 to 59 years. Demographics of our patients are presented in Table 1.

The presenting symptoms of *C. difficile* enteritis were high volume watery ileostomy output followed by ileus in five of six patients. Radiographic studies suggested large volumes of fluid within the small intestine even if the intestine was bypassed as in the case of an ileal pouch anal anastomosis (Fig. 2). Some patients experienced relief with placement of a Foley catheter (24 french) into the stoma with expulsion of malodorous, yellow effluent positive for *C. difficile* toxin.

Four of our six patients presented a fever and an elevated WBC that ranged from 15,000–35,000 cells/dl. One patient's WBC decreased to 2,800 cells/dl and presented in profound sepsis. Elevated platelet counts ranging from 374,000 to 1,162,000 platelets/dl were detected in five of the six patients.

Five of the six patients developed complications which resulted in prolonged hospitalization, readmission, and further surgery. Three of these patients were readmitted with fever and dehydration. One patient required a reoperation to bypass strictured small bowel and one patient was transferred to the ICU for the management of hemodynamic instability. The mean number of hospital days required for this series of IBD patients with postoperative *C. difficile* enteritis was 13.7±7.9 days with a hospital stay day range from 8–29 days. Readmission was

Table 1 Characteristics of Patients with Small Intestinal *C. difficile*

Patient	Age (year)	Prednisone (mg/day)	Immunosuppression	Acid Suppression	History of *C. difficile*
1	20	40	–	–	+
2	59	10	Azathioprine	H2 blocker	+
3	20	20	Azathioprine	–	+
4	52	0	–	–	–
5	22	20	–	–	+
6	40	20	–	–	–

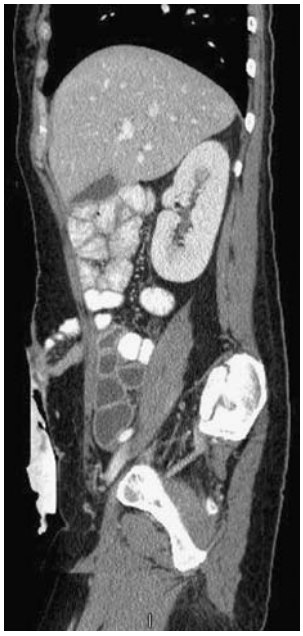

Figure 2 This is a sagittal CT reconstruction of a patient who had had an ileal pouch anal anastomosis with diverting loop ileostomy. The loops of bowel can be seen in the upper abdomen and are filled with contrast that was administered orally. The fluid filled loops of small bowel are the distal small intestine. Normally, these loops are collapsed but, because of the *C. difficile* enteritis, they are fluid filled. The patient was constantly passing fluid through her anus. The loop ostomy and appliance filled with contrast are visualized midabdomen.

required in half of the patients and the mean number of days required for the readmission was 7.9±0.6 days.

The single patient who underwent a repeat laparotomy had a stricture at the site of the closure of her diverting loop ileostomy closure. When the intestine was opened, sheets of dead mucosa with hemorrhaging small bowel were identified. Because the stricture was surrounded by dense adhesions, 10 cm of small bowel was bypassed.

Once the diagnosis was suspected, patients were treated with intravenous hydration and metronidazole until able to tolerate oral metronidazole and/or vancomycin. Combination therapy was utilized of luminal vancomycin (250–500 mg every 6 hour) and IV or oral metronidazole (500 mg every 8 hour). One patient was treated with vancomycin lavages (1 g vancomycin per liter of saline; 250 cc every 12 hour) via the distal loop of the ileostomy, due to poor toleration of oral metronidazole. None of the six patients died. All have resumed education or employment.

Discussion

Review of the literature revealed 14 prior case reports of *C. difficile* enteritis from 1980–2006 (Table 2). Three patients from the case reports never underwent a surgical procedure and *C. difficile* enteritis was found during surgery performed for the infection.[4–6] Eleven patients underwent gastrointestinal (GI) surgical procedures before the development of their *C. difficile* enteritis. Six of the 11 had remote procedures performed from 1 to 31 years before developing enteritis. One of these patients received no antibiotics before the episode of *C. difficile* enteritis.[12] Antibiotic therapy predisposed enteritis in five patients, three for non-GI surgical procedures,[8–10] and two for urinary tract infections.[7,11]

The final five patients had undergone recent gastrointestinal surgery. All five patients received preoperative antibiotics and developed *C. difficile* enteritis as an early postoperative complication in less than 90 days.[6,13–16] These five patients with recent gastrointestinal surgery group had a mortality rate of 100%; their mean age was 66.4 years, (range 53–80 years). Patients with remote surgery were younger with a mean age of 51.2 years and an age range from 23 to 71 years. They had slightly decreased mortality of 66%.

Four patients in the literature had ileostomies. Two of these patients had Crohn's disease and the other two patients had ulcerative colitis. This group most closely resembles our cohort with a mean age of 39.5 years and an

Table 2 Previously Reported Cases of Small Intestinal *C. difficile*

Procedure	Antibiotics	Age (year)	Gender	Diagnosis	Disposition
None	+	18	M	Tuberculosis	Resolution[4]
None	+	83	F	Urinary tract infection	Resolution[5]
None	+	83	F	Pneumonia	Resolution[6]
GI surgery—remote	+	56	F	Crohn's disease	Resolution[7]
GI surgery—remote	+	71	M	Multiple colon polyps	Death[8]
GI surgery—remote	+	65	M	CRF/dialysis	Death[9]
GI surgery—remote	+	26	M	Ulcerative colitis	Resolution[10]
GI surgery—remote	+	66	M	Colon CA	Death[11]
GI surgery—remote	−	23	F	Crohn's disease	Death[12]
GI surgery—recent	+	53	M	Ulcerative colitis	Death[13]
GI surgery—recent	+	80	M	Colorectal CA	Death[6]
GI surgery—recent	+	60	M	Adhesions	Death[14]
GI surgery—recent	+	69	M	Colon CA	Death[15]
GI surgery—recent	+	70	M	Bladder CA	Death[16]

age range of 23–56 years. Fifty percent of this group was female. One of these patients died due to the *C. difficile* enteritis.[7,10,12,13]

Despite the historical rarity of *C. difficile* enteritis, our experience suggests that the incidence is rising. The reasons for this increase are unclear. Factors which mediate overt disease produced by *C. difficile* include the dose and toxigenicity of the colonizing strain, the ability to adhere to epithelium, the concurrent presence of organisms that affect multiplication and toxin production or activity, as well as the susceptibility of the host. Recently, the disease has been reported in other species. These include horses, ostriches, companion animals, calves, and pigs.[17] Because *C. difficile* is a spore-forming organism, sporicidal temperatures and pressures are necessary to eliminate the potential for food supply contamination. Because this is rarely achieved during food preparation, we anticipate, for susceptible populations, that the development of *C. difficile* infections will remain a new threat.

Others have suggested that gastric acid suppressive agents increase the incidence of *C. difficile* infections (proton pump inhibitors, adjusted rate ratio of 2.9; H2 receptor antagonists, adjusted rate ratio of 2.0).[18] However, this was not observed in our patient population with only one in six patients on an H2 blocker. Five of our patients were on prednisone but this has not been reported as a risk factor for the development of *C. difficile* infections. Similarly, azathioprine has not been reported as a risk. However, immunosuppression associated with chemotherapy may predispose patients to *C. difficile*.

The widespread implementation of waterless hand cleansing solutions may also contribute to an increase in the frequency of the disease. Weber et al.[19] showed that hand hygiene agents such as hand washing with soap and water, 2% chlorhexidine gluconate, or chlorine-containing towels reduced the amount of spore contamination. Whereas the use of a waterless rub containing ethyl alcohol was not effective in removing spores. This knowledge has led to strict water-based hand cleansing standards for our patients in enteric isolation.

Clinicians should suspect *C. difficile* enteritis in patients with a history of IBD, *C. difficile* colitis before colectomy, and those who exhibit an abnormal postoperative course. Although the mortality rate in the literature is alarmingly high, there were no deaths in this series. Our favorable outcomes may be attributed in part to rapid diagnosis and aggressive treatment of *C. difficile* enteritis.

Conclusions

Small intestinal *C. difficile* can be seen more frequently now than previously reported in patients with inflammatory bowel disease whom have undergone total colectomy. Initially, *C. difficile* enteritis presents as diarrhea followed by ileus with fluid filled loops of small intestine. The sepsis and fluid shifts, which accompany this disease, can be life threatening but are treatable. Even immunosuppressed patients can be successfully treated provided the disorder is considered early and treated aggressively.

The increase in the number of these patients may reflect an increase in the number of patients with *C. difficile*, the virulence of the organism, the implementation of waterless hand wash solutions or the susceptibility of this patient population to the infection. Early recognition of small and large intestinal disease with appropriate therapy will prevent potential mortality in this vulnerable cohort of patients.

References

1. Schroeder, M. S. (2005). Clostridium difficile-associated diarrhea. *American Family Physician, 71*(5), 921–928.
2. bioMerieux, I. Immunoassay, vidas® assays, antigen detection. http://www.biomerieux-usa.com/clinical/immunoassay/vidas/vidas_assays.htm.
3. Meridian Bioscience, I. *Product catalog, c. Difficile, premier™-toxins A and B*.
4. Miller, D. L., Sedlack, J. D., & Holt, R. W. (1989). Perforation complicating rifampin-associated pseudomembranous enteritis. *Archives of Surgery, 124*(9), 1082.
5. Jacobs, A., Barnard, K., Fishel, R., & Gradon, J. D. (2001). Extracolonic manifestations of clostridium difficile infections. Presentation of 2 cases and review of the literature. *Medicine (Baltimore), 80*(2), 88–101.
6. Hayetian, F. D., Read, T. E., Brozovich, M., Garvin, R. P., & Caushaj, P. F. (2006). Ileal perforation secondary to clostridium difficile enteritis: Report of 2 cases. *Archives of Surgery, 141*(1), 97–99.
7. Vesoulis, Z., Williams, G., & Matthews, B. (2000). Pseudomembranous enteritis after proctocolectomy: Report of a case. *Diseases of the Colon & Rectum, 43*(4), 551–554.
8. Yee, H. F. Jr., Brown, R. S. Jr., & Ostroff, J. W. (1996). Fatal clostridium difficile enteritis after total abdominal colectomy. *Journal of Clinical Gastroenterology, 22*(1), 45–47.
9. Kralovich, K. A., Sacksner, J., Karmy-Jones, R. A., & Eggenberger, J. C. (1997). Pseudomembranous colitis with associated fulminant ileitis in the defunctionalized limb of a jejunal–ileal bypass. Report of a case. *Diseases of the Colon & Rectum, 40*(5), 622–624.
10. Freiler, J. F., Durning, S. J., & Ender, P. T. (2001). Clostridium difficile small bowel enteritis occurring after total colectomy. *Clinical Infectious Diseases, 33*(8), 1429–1431, (discussion 1432).
11. Tsutaoka, B., Hansen, J., Johnson, D., & Holodniy, M. (1994). Antibiotic-associated pseudomembranous enteritis due to clostridium difficile. *Clinical Infectious Diseases, 18*(6), 982–984.
12. LaMont, J. T., & Trnka, Y. M. (1980). Therapeutic implications of clostridium difficile toxin during relapse of chronic inflammatory bowel disease. *Lancet, 1*(8165), 381–383.
13. Kuntz, D. P., Shortsleeve, M. J., Kantrowitz, P. A., & Gauvin, G. P. (1993). Clostridium difficile enteritis. A cause of intramural gas. *Digestive Diseases and Sciences, 38*(10), 1942–1944.

14. Tjandra, J. J., Street, A., Thomas, R. J., Gibson, R., Eng, P., & Cade, J. (2001). Fatal clostridium difficile infection of the small bowel after complex colorectal surgery. *ANZ Journal of Surgery, 71*(8), 500–503.
15. Testore, G., Pantosti, A., Panichi, G. P. G. I. L., Nardi, F., & Contini, M. (1984). Pseudomembranous enteritis associated with clostridium difficile. *Italian Journal of Gastroenterology, 16*, 229–230.
16. Shortland, J. R., Spencer, R. C., & Williams, J. L. (1983). Pseudomembranous colitis associated with changes in an ileal conduit. *Journal of Clinical Pathology, 36*(10), 1184–1187.
17. Songer, J. G. (2004). The emergence of clostridium difficile as a pathogen of food animals. *Animal Health Research Reviews, 5*(2), 321–326.
18. Dial, S., Delaney, J. A., Barkun, A. N., & Suissa, S. (2005). Use of gastric acid-suppressive agents and the risk of community-acquired clostridium difficile-associated disease. *JAMA, 294*(23), 2989–2995.
19. Weber, D. J., Sickbert-Bennett, E., Gergen, M. F., & Rutala, W. A. (2003). Efficacy of selected hand hygiene agents used to remove bacillus atrophaeus (a surrogate of bacillus anthracis) from contaminated hands. *JAMA, 289*(10), 1274–1277.

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Radio Frequency Ablation for Hepatocellular Carcinoma in Cirrhotic Patients: Prognostic Factors for Survival

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Abstract
Background Radio frequency ablation (RFA) of hepatocellular carcinoma has proved to be useful in local control of tumor. A few data on survival after treatment are available in literature. The aim of the study was to evaluate factors related to survival and to identify different classes of risk after radio frequency ablation.
Methods Ninety-eight cirrhotic patients with 145 hepatocellular carcinomas were treated with radio frequency ablation from January 1998 to May 2004. In 55 patients, cirrhosis was in Child-Pugh class A, and in 43, in class B. Tumor was single in 60 and multiple in 38; mean tumor number was 1.5 (range 1–3). Tumor size ranged from 1.5 to 6.0 cm, mean 3.8 cm. Mean follow up period was 24.9 months. Radio frequency ablation was performed with expandable type needle with percutaneous approach under real-time ultrasound guidance. For statistical analysis, univariate and multivariate analysis were performed.
Results Complete ablation of the tumor was achieved in 85.5% of lesions. Survival, 1 and 3 years, was 76.7 and 36.6%, respectively. Univariate analysis showed that Cancer of the Liver Italian Program (CLIP) score, tumor growth pattern, α-fetoprotein level, and complete tumor necrosis, were factors significantly related to poor survival. Multivariate analysis identified that factors related to poor survival were α-fetoprotein level >100 ng/ml, Child-Pugh class B, and incomplete tumor necrosis with a hazard ratio of 4.0, 2.7, and 3.8, respectively. After complete ablation, median survival was 38 months in patients with Child-Pugh class A cirrhosis and α-fetoprotein level ≤100 ng/ml, 22 months for patient with Child-Pugh class B cirrhosis and α-fetoprotein ≤100 ng/ml, and 9 months for patient with Child-Pugh class A cirrhosis and α-fetoprotein >100 ng/ml ($P<0.01$).
Conclusions Complete necrosis and absence of residual tumor positively affect survival after RFA. In patients with Child-Pugh A cirrhosis and α-fetoprotein level ≤100 radio frequency, ablation have results, 55% after 3 years, that are comparable to those of surgical resection. Patients with Child-Pugh B cirrhosis and/or α-fetoprotein >100 ng/ml showed less satisfactory results, and in these patients, multimodality treatment or other treatments should be considered.

Keywords Radio frequency ablation · Hepatocellular carcinoma · Prognostic factors · Survival

Abbreviations
(HCC) Hepatocellular carcinoma
(RFA) Radio frequency ablation
(WHO) World Health Organization
(HRs) Hazard ratio estimates
(PH) Proportional hazard
(UICC) International Union Against Cancer

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Introduction

Hepatocellular carcinoma (HCC) is the fifth *most* common tumor in the world, with an estimated incidence of 500,000–1,000,000 new cases every year, and its incidence is still rising in Western Countries.[1–3]

HCC arise on cirrhosis in more than 80% of patients with a natural history that is extremely variable because is it strongly related to the severity of underling liver disease.[4] Liver transplantation and liver resection offer the best results in terms of survival, but only 20–30 % of patients can be submitted to these treatments.[5–7] Liver transplantation treats both cirrhosis and HCC, but it can be applied only in a small portion of patients because of organ shortage.

Liver resection in cirrhotic patients has good results for patients with Child-Pugh class A cirrhosis without portal hypertension and normal bilirubin level, but results are not satisfactory in patients with more severe cirrhosis. Moreover, recurrence rate after liver resection is high with an incidence greater than 70% after 5 years.[8]

During the last years, local ablation techniques (ethanol injection, microwave ablation, and radio frequency ablation) gained *consent* because of good results in local necrosis and low complication rates.

Many studies in literature reported efficacy of radio frequency ablation (RFA), but data about survival are limited to a small series with a short follow-up. Moreover, a few studies in literature analyze prognostic factor for survival after RFA in HCC.[9–11]

The aim of this study was to analyze prognostic factors for survival after RFA of HCC on cirrhosis and to identify different prognostic classes of patients.

Materials and Methods

Between January 1998 and May 2004, 98 cirrhotic patients with 145 HCC were treated with percutaneous RFA at the 1st Department of General Surgery of University of Verona. Among these, 79 were male and 19 were female. Mean age of patients was 67 years (range 41–88). Patient characteristics are summarized in Table 1. Patients were classified according to Child-Pugh classification and CLIP (Cancer of The Liver Italian Program) score.[12,13]

Diagnosis of HCC were based on accordance of two imaging techniques [computed tomography (CT) and magnetic resonance imaging (MRI)] showing an arterial hypervascularization in a focal lesion ≥2 cm or with a combined criteria of an imaging technique and serum α-fetoprotein level greater than 400 ng/ml, according to The European Association for Study of the Liver (EASL) consensus conference criteria.[14] In 25 patients with uncertain radiological findings, diagnosis was confirmed with fine needle biopsy. Exclusion criteria from the study were: Child-Pugh class C cirrhosis, severe ascites, tumor larger than 6 cm, more than four tumor nodules and extrahepatic disease.

All patients were submitted to RFA with percutaneous approach under real-time ultrasound guidance in the operating room with general anesthesia or conscious sedation. The expandable electrode was inserted into the tumor with ultrasonography guidance and prongs were deployed. During the study period, two different types of needle and RF generator were employed. From 1998 to June 2000, we utilized 15-gauge electrodes with four hooks connected to an RF generator with 50-W output (model 500, RITA Medical System, Mountain View, CA). From June 2000, a new expandable electrode (with seven to nine hooks) and a new RF generator with 150-W output was introduced (model 1500, RITA Medical System, Mountain View, CA). Real time monitoring of temperature of hooks was set to maintain mean temperature of 100°C for 12–20 min. For lesions larger than 5 cm, multiple overlapping ablations were applied.

After treatment, blood count and liver function tests were performed after 12 and 24 h. Patients were monitored in the hospital overnight and discharged the next day unless there was presence of complications. Evaluation of tumor response after RFA was based on World Health Organization (WHO) criteria in which complete tumor response is defined as the absence of arterial enhancement within or at

Table 1 Characteristics of Patients who Underwent Radiofrequency Ablation

Variable	Number (*n*)	Percentage (%)
Gender		
Male	79	80.6
Female	19	19.4
Age (year)		
<65	33	33.7
65–70	28	28.6
>70	37	31.4
Type of cirrhosis		
Viral	68	69.4
Not viral	30	30.6
Child-Pugh class		
A	55	56.1
B	43	43.9
CLIP score*		
0	27	29.0
1	40	43.0
>1	26	28.0
Number of tumors		
Single	60	61.2
Multiple	38	38.8
Tumor size (cm)		
≤3	53	36.5
3–5	76	52.4
>5	16	11.1
α-fetoprotein level (ng/ml)		
≤100	74	75.5
>100	17	24.5

*Cancer of the Liver Italian Program

the periphery of all treated tumors determined by two observations (CT or MRI) not less than 4 weeks apart.[15]

In our institution, we routinely perform dual phase contrast-enhanced CT scan. We utilize dynamic gadolinium enhanced MRI in patients with contraindications to CT (i.e., chronic renal failure, history of adverse reactions to iodine contrast agents). Patients with incomplete tumor response were evaluated for a new RFA treatment.

Local recurrence was defined as evidence of pathologic enhancement within or at the periphery of a tumor with previous complete response.

Distant intrahepatic recurrence was defined as appearance of new liver tumors in the liver distant from the ablated tumor.

Follow-up protocol consists of monitoring serum α-fetoprotein level every 3 months and evaluation of imaging, contrast CT, or MRI after 3 months, and every 6 months thereafter. Local recurrences or distant intrahepatic recurrences were reevaluated for new treatment in all patients with ethanol injection, RFA, or chemoembolization according to number and size of recurrence.

Statistical Analysis

Data were collected and analyzed by Stata version 8.2 (StataCorp, College Station, Texas).[16] Comparison between categorical variables was carried out with Pearson chi-square test. For univariate survival analysis, Cox regression model was utilized with log-rank test evaluation of statistical significance. The univariate survival analysis was reported for each of the observed variables and uncorrected (crude) hazard ratios together with 95% confidence interval and *P* value of the log-rank tests.

For univariate analysis, the following variables were analyzed: gender, age, type of cirrhosis, CLIP score, Child-Pugh class, tumor size, number of tumors, tumor location, type of growth of tumor, α-fetoprotein level, complete ablation of tumor after treatment.

For multivariate analysis, the bootstrap variable selection method proposed by Austin and Tu was utilized to identify predictive variables. Cox models were estimated on a set of 5,000 bootstrap samples after stepwise (forward and backward) selection, and the candidate variables were ranked according to the proportion of bootstrap samples in which they were identified as independent predictors.[17] Starting from the most frequently selected variables and sequentially adding variables until the predictive accuracy do not significantly increase, the final proportional hazard Cox models were estimated. The goodness-of-fit of the models was tested by the method proposed in May and Hosmer using 6, 8, and 10 quantiles of risk.[18] Moreover, after model estimation, the analysis of deviance and efficient score residuals were performed, and the proportional hazard assumption was tested based on Schoenfeld residuals.

Results

During the study period, 98 patients with 145 tumors were treated with RFA. Median follow up for surviving patients was 22 months (range 3–76). A single tumor was present in 60 patients and multiple tumors in 38 patients; mean tumor number was 1.5 (range 1–3). Mean tumor size was 3.8 cm (range 1.5–6). In 14 patients, minor complications occurred, and major complications occurred in 8 patients. No treatment-related deaths were observed. Detailed description of complications are reported in Table 2.

Complete tumor response was achieved after the first treatment in 47 (88.6%) lesions smaller than 3 cm, in 54 (71%) lesion larger than 3 cm and smaller than 5 cm, and in 6 (37.5%) lesions larger than 5 cm ($P<0.001$).

RFA treatment was repeated in 38 lesions with incomplete tumor response. After single or multiple RFA treatment, complete response was achieved in 98.1% (52 lesions) for HCCs smaller than 3 cm, in 81.5% (62 lesions) for HCCs larger than 3 cm and smaller than 5 cm, and in 62.5% (10 lesions) for lesions larger than 5 cm ($P<0.001$).

We analyzed the following tumor-related variables to identify factors related to complete response: size, number, type of growth, subcapsular location, location near major vessels, and α-fetoprotein level. We identify that complete tumor response was higher in patients with tumor smaller or equal to 3 cm (98.1% versus 78.7%, $P=0.005$), distant from major vessel (90.2% versus 73.3%, $P=0.02$), and with α-fetoprotein level lower or equal to 100 ng/dl (89.2% versus 58.8%, $P=0.002$). No patients with HCC smaller than 3 cm and α-fetoprotein level lower than 100 ng/dl had incomplete tumor response.

Table 2 Complications After Radiofrequency Ablation Sessions in 98 Patients

Complication	Number (*n*)	Percentage (%)
Major	8	(8.1)
Rapid neoplastic progression	4	
Needle track seeding	1	
Bacterial endocarditis	1	
Intraperitoneal bleeding*	1	
Hepatic decompensation	1	
Minor	14	(14.2)
Fever and pain†	11	
Pleural effusion	2	
Insertion needle site burn	1	

*Medical treatment
†Fever >38°C and pain for longer than 5 days

During follow-up, local recurrences were observed in 31 lesions (21.3%), whereas distant recurrences were detected in 36 patients (36.7%). During follow-up, 57 patients died. At the time of death, cause of death was related to tumor in 39 patients, whereas in 18 patients, no viable tumor was present and death was related to complications of cirrhosis (liver failure in 10, variceal bleeding in 7 and hepato-renal syndrome in 1).

With univariate analysis, we identified that CLIP score, tumor growth type, α-fetoprotein level, and complete response after treatment were significantly related to survival (Table 3). We identified that the best cutoff level for α-fetoprotein level in our sample is 100 ng/dl. We estimated a Cox multiple regression model with the following explanatory variables: gender, age, tumor ablation, Child-Pugh class, and α-fetoprotein. Age was added to the model after standardization (mean 67.2, SD 8.7).

Table 4 shows the hazard ratio estimates (HRs) of the model, together with their 95% confidence interval and the *P* values of the corresponding HR=1 hypothesis tests. The goodness-of-fit of the model was successfully tested within quantile *z*-scores, and *P* values showed that the estimated expected numbers of failures were not significantly different from the observed numbers of failures. Finally, the proportional hazard (PH) assumption was globally tested and cannot be rejected (χ^2=10.3, *P* value=0.115).

Another multivariate model was estimated with the following explanatory variables: gender, age, CLIP score,

Table 3 Univariate Survival Analysis

Variable	Number (*N*)	Median survival		Hazard Ratio (Not Corrected)		Log-Rank Test
		Median	95% CI	HR	95% CI	*P* value
Gender						
Female	19	39.1	22.2–45.9	Reference		0.151
Male	79	25.0	16.1–28.5	1.7	0.8–3.6	
Age (years)						
<65	33	35.3	16.1–42.9	Reference		0.641
65–72	36	25.0	15.0–36.9	1.3	0.7–2.5	
>72	29	22.2	11.9–28.3	1.3	0.7–2.5	
Type of cirrhosis						
Viral	30	28.0	15.1–48.9	Reference		0.390
Not viral	68	24.8	18.3–29.3	1.3	0.7–2.3	
CLIP score*						
0	27	39.0	25.0–50.3	Reference		0.020
1	40	22.2	15.0–27.9	2.4	1.2–5.0	
>1	26	15.1	06.4–42.9	2.8	1.3–6.2	
Child-Pugh class						
A	55	28.3	20.9–41.5	Reference		0.091
B	43	21.7	13.1–27.9	1.6	0.9–2.6	
Tumor size (cm)						
≤3	29	29.3	20.6–42.9	Reference		0.146
>3 and <5	45	25.0	15.0–35.3	1.3	0.7–2.5	
≥5	24	20.3	8.1–28.5	1.9	1.0–3.8	
Number of tumors						
Single	60	26.4	20.9–39.0	Reference		0.254
Multiple	38	24.8	15.0–28.5	1.4	0.8–2.3	
Subcapsular location						
No	51	29.3	21.7–41.5	Reference		0.152
Yes	41	20.9	14.1–26.4	1.5	0.9–2.6	
Growth type						
Expansive	83	28.0	21.7–39.1	Reference		0.028
Infiltrative	15	20.3	12.3–26.1	2.0	1.1–3.8	
α-fetoprotein (ng/ml)						
≤100	74	26.4	15.1–43.2	Reference		0.001
>100	17	8.4	6.4–20.9	3.1	1.5–6.4	
Tumor ablation						
Complete response	81	26.7	22.2–36.9	Reference		0.019
Partial response	17	9.7	04.0–13.4	2.2	1.1–4.5	

*Cancer of The Liver Italian Program

Table 4 Cox Regression Model With or Without CLIP Score

Variable	Hazard ratio			
	Coefficient	HR	95% CI	*P* value
Without CLIP Score*				
Gender				
Female		Ref.		0.004
Male	1.24	3.5	1.5–8.1	
Tumor ablation				
Complete response		Ref.		
Partial response	1.33	3.8	1.5–9.7	0.006
Age† (year)	0.51	1.7	1.2–2.3	0.003
Child-Pugh class				
A		Ref.		
B	1.00	2.7	1.6–4.8	<0.001
α-fetoprotein (ng/ml)				
≤100		Ref.		
>100	1.39	4.0	1.6–9.8	0.002
With CLIP Score				
Gender				
Female	Ref.			
Male	0.92	2.5	1.1–5.8	0.031
Tumor ablation				
Complete response	Ref.			
Partial response	1.26	3.5	1.4–9.0	0.008
CLIP score				
0	Ref.			
1	0.81	2.3	1.1–4.7	0.033
>1	1.23	3.4	1.5–8.0	0.005
α-fetoprotein (ng/ml)				
≤100	Ref.			
>100	0.91	2.5	1.0–6.1	0.044

*Cancer of The Liver Italian Program
†Standardized continuous variable

and α-fetoprotein. A complete description of the model is reported in Table 4. The goodness-of-fit of the model was successfully tested. Finally, the proportional hazard (PH) assumption was globally tested and should be rejected (*P* value=0.029).

Overall median survival after RFA was 26 months; 1- and 3-years survival was 76.7 and 36.6%, respectively. Survival for patients with Child-Pugh class A cirrhosis was 81.8 and 45.2% after 1 and 3 years, respectively, whereas it was 70.5 and 27% for patients with Child Pugh class B cirrhosis (*P*=0.09), respectively. We did not observe differences in cause of death between patients with Child-Pugh class A and B; deaths were tumor related in 70.4% and in 62.1%, respectively (*P*=0.5). Patients with complete tumor response after treatment had a significantly longer survival in comparison to patients with incomplete tumor necrosis: median survival of 27 months (95% CI 23–30) and 8 months (95% CI 5–11), respectively (*P*<0.01) (Fig. 1).

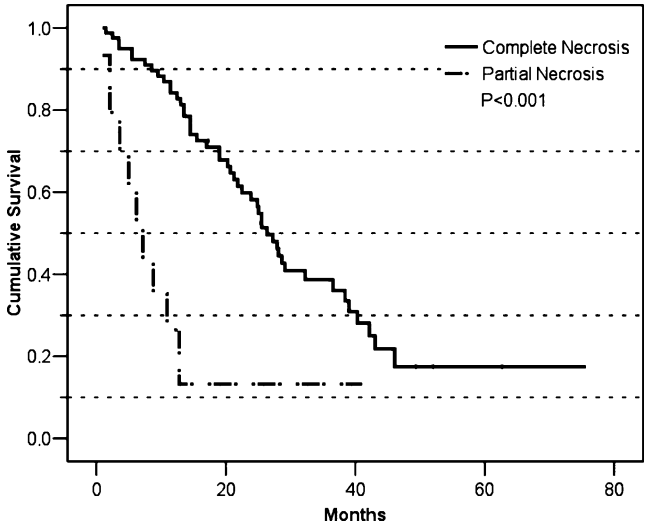

Figure 1 Survival curves according to complete or partial necrosis after RFA treatment (*P*<0.01).

After complete response of the tumor, median survival for patients with Child-Pugh class A cirrhosis and α-fetoprotein ≤100 ng/ml was 38 months (95% CI 23–53), 22 months (95% CI 12–31) for patients with Child-Pugh class B cirrhosis and α-fetoprotein ≤100 ng/ml, and 9 months (95% CI 7–11) for patients with Child-Pugh class A cirrhosis and α-fetoprotein >100 ng/ml (*p*<0.01, Fig. 2). Survival for patients with Child-Pugh class B cirrhosis and α-fetoprotein >100 ng/ml was not analyzed because of small sample size.

After complete ablation of the tumor, median survival for patients with Clip score equal to 0 and α-fetoprotein ≤100 ng/ml was 38 months (95% CI: 24–51), 26 months (95% CI: 20–32) for patients Clip score greater than 0 and α-fetoprotein ≤100 ng/ml and 9 months (95% CI 3–15) for

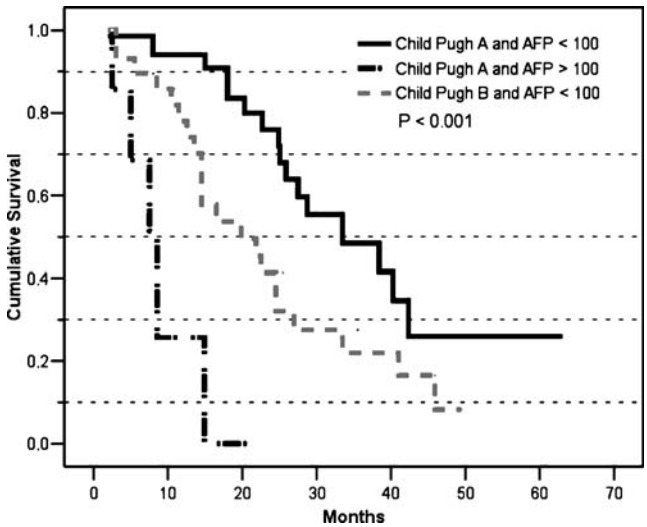

Figure 2 Survival curves after complete necrosis of tumor according to Child-Pugh class and α-fetoprotein level (*P*<0.01).

patients Clip score greater than 0 and α-fetoprotein >100 ng/ml ($P<0.01$). Survival for patients with Clip score equal to 0 and α-fetoprotein >100 ng/ml was not analyzed because no patient belonged to this group.

Discussion

RFA was recently introduced in clinical practice and is widely used in Europe and East countries to treat primary liver tumors with greater efficacy than ethanol injection in terms of tumor necrosis and number of sessions for complete response.[19,20] Studies on ablation efficacy showed that complete necrosis of the tumor can be achieved in more than 90% of HCCs smaller than 3 cm, in 60–93% for lesions between 3 and 5 cm, and in 24–93% for lesion between 5 and 8 cm.[21–23]

Data about survival after RFA of HCCs are limited to small series and short follow-up. However, survival for small HCCs are good and are comparable to those of surgical resection with a 3-year survival of 45–68%.[21,24] Few studies in literature analyze prognostic factors for survival after RFA, and they identify that factors related to survival are tumor size, α-fetoprotein level, complete tumor response after RFA, and albumin level.[9,10] Our study confirms data of literature, and we identified that factors related to survival were: Child-Pugh class, α-fetoprotein level, and complete tumor response after RFA.

We confirm, in multivariate analysis, the importance of severity of liver disease for survival of patients after treatment. Child-Pugh class B patients have a relative risk equal to 2.7 in comparison to Child-Pugh A patients. Survival after 1 and 3 years for Child-Pugh class A patients was 81.8 and 45.2%, respectively, whereas survival for Child-Pugh class B patients was 70.5 and 27% ($P=0.09$), respectively.

Our study confirms data of literature about the importance of complete response of the tumor.[9,25] Patients with incomplete tumor necrosis after single or multiple treatment have hazard risk for death of 3.8 (95% CI 1.5–9.7). Moreover, our study shows that complete response can be achieved with both single or multiple treatment without differences in terms of survival, with a relative risk of 0.7 (95% CI 0.3–1.3).

In our results, high α-fetoprotein level was the strongest prognostic factor for survival with a hazard ratio for death of 4.0 (95% CI 1.6–9.8). As reported in previous studies, α-fetoprotein reflect the biological behavior of a tumor, and higher levels of this marker are related to size, to number of neoplastic nodules, and to poor prognosis.[26]

Utilizing the multivariate model, we identify a group of patients with best prognosis (Child-Pugh class A cirrhosis and with α-fetoprotein level ≤100 ng/ml and complete response after RFA) that has a median survival of 38 months with a 3-year survival rate of 55% and that are comparable with surgical series.[27] The other group of patients (incomplete necrosis to treatment and α-fetoprotein level greater than 100 ng/ml) has worse prognosis with a median survival of 6 months (95% CI 6–7 months) and with no survivors after 3 years.

Staging system of HCC have a great importance in prognostic evaluation after surgical and nonsurgical interventions. Many staging systems were proposed, but complex relationship between cirrhosis, stage of tumor, and different types of treatment does not allow accurate stratification of different prognostic classes. Among different staging systems, results of those that consider only the severity of liver damage (Child-Pugh) or only the extension of tumor [International Union Against Cancer (UICC) TNM staging] does not allow a precise classification of risk.[28] More recently, other staging systems that combine evaluation of cirrhosis and extension of tumor were introduced [CLIP, Chinese University Prognostic Index (CUPI), Barcelona Clinic Liver Cancer (BCLC)].[28] The CLIP score, developed from a group of patients submitted to various types of treatment, showed good performances in surgical and nonsurgical patients in validation studies.[29–31]

With multivariate analysis, we identify that a combination of CLIP score and α-fetoprotein threshold level of 100 ng/ml have greater goodness-of-fit in comparison to CLIP score. This should be associated to α-fetoprotein threshold level (400 ng/ml) included in CLIP score that is observed in a few patients in our series (only 12 patients). In our opinion, a threshold level of 100 ng/ml better describe our study population.

This study confirms good results of RFA in a selected group of patients. Complete necrosis of the tumor after treatment have great value and is one of the most important prognostic factor. The best results were observed in patients with Child-Pugh class A cirrhosis and a low level of α-fetoprotein. In patients with Child B or with AFP level greater than 100 ng/ml, RFA treatment showed less satisfactory results, and in these patients, multimodality treatment or other treatments should be considered.

References

1. Bosch, F. X., Ribes, J., & Borras, J. (1999). Epidemiology of primary liver cancer. *Seminars in Liver Disease, 19*, 271–285.
2. Bosch, F. X., Ribes, J., Diaz, M., & Cleries, R. (2004). Primary liver cancer: Worldwide incidence and trends. *Gastroenterology, 127*(5 Suppl 1), S5–S16, Nov.
3. Yanaga, K. (2004). Current status of hepatic resection for hepatocellular carcinoma. *Journal of Gastroenterology, 39*(10), 919–926, Oct.
4. Carr, B. I. (2004). Hepatocellular carcinoma: Current management and future trends. *Gastroenterology, 127*(5 Suppl 1), S218–S224, Nov.

5. Fong, Y., Sun, R. L., Jarnagin, W., & Blumgart, L. H. (1999). An analysis of 412 cases of hepatocellular carcinoma at a Western center. *Annals of Surgery, 229*, 790–799.
6. Llovet, J. M., Fuster, J., & Bruix, J. (1999). Intention-to-treat analysis of surgical treatment for early hepatocellular carcinoma: Resection versus transplantation. *Hepatology, 30*, 1434–1440.
7. Poon, R. T., Fan, S. T., Lo, C. M., Ng, I. O., Liu, C. L., Lam, C. M., et al. (2001). Improving survival results after resection of hepatocellular carcinoma: A prospective study of 377 patients over 10 years. *Annals of Surgery, 234*, 63–70.
8. Bruix, J., & Llovet, J. M. (2002). Prognostic prediction and treatment strategy in hepatocellular carcinoma. *Hepatology, 35*(3), 519–524, Mar.
9. Camma, C., Di Marco, V., Orlando, A., Sandonato L., Casaril A., Parisi P., et al. (2005). Treatment of hepatocellular carcinoma in compensated cirrhosis with radio-frequency thermal ablation (RFTA): A prospective study. *Journal of Hepatology, 42*(4), 535–540, Apr.
10. Montorsi, M., Santambrogio, R., Bianchi, P., Donadon, M., Moroni, E., Spinelli, A., et al. (2005). Survival and recurrences after hepatic resection or radiofrequency for hepatocellular carcinoma in cirrhotic patients: A multivariate analysis. *Journal of Gastrointestinal Surgery, 9*(1), 62–67, Jan.
11. Vivarelli, M., Guglielmi, A., Ruzzenente, A., Cucchetti, A., Bellusci, R., Cordiano, C., et al. (2004). Surgical resection versus percutaneous radiofrequency ablation in the treatment of hepatocellular carcinoma on cirrhotic liver. *Annals of Surgery, 240*(1), 102–107, Jul.
12. Pugh, R. N. H., Murray-Lyon, I. M., & Dawson, J. L. (1973). Transection of oesophagus for bleeding oesophageal varices. *British Journal of Surgery, 60*, 646–664.
13. The Cancer of the Liver Italian Program (CLIP) Investigators. (1998). A new prognostic system for hepatocellular carcinoma: A retrospective study of 435 patients. *Hepatology, 28*, 751–755.
14. Bruix, J., Sherman, M., Llovet, J. M., Beaugrand, M., Lencioni, R., Burroughs, A. K., et al. (2001). EASL Panel of Experts on HCC. Clinical management of hepatocellular carcinoma. Conclusions of the Barcelona-2000 EASL conference. European Association for the Study of the Liver. *Journal of Hepatology*, *35* (3), 421–430, Sep.
15. Miller, A. B., Hoogstraten, B., Staquet, M., & Winkler, A. (1981). Reporting results of cancer treatment. *Cancer, 47*, 207–214.
16. Rabe-Hesketh, S., & Everitt, B. S. A. (2003). *Handbook of Statistical Analyses Using Stata* (3rd ed.). Boca Raton, FL: Chapman & Hall/CRC.
17. Austin, P. C., & Tu, J. V. (2004). Bootstrap methods for developing predictive models. *The American Statistician, 58*(2), 131–137.
18. May, S., & Hosmer, D. W. (1998). A simplified method of calculating an overall goodness-of-fit test for the Cox proportional hazards model. *Lifetime Data Analysis, 4*, 109–120.
19. Lencioni, R. A., Allgaier, H. P., Cioni, D., Olschewski, M., Deibert, P., Crocetti L., et al. (2003). Small hepatocellular carcinoma in cirrhosis: Randomized comparison of radio-frequency thermal ablation versus percutaneous ethanol injection. *Radiology, 228*, 235–240.
20. Livraghi, T., Goldberg, S. N., Lazzaroni, S., Meloni, F., Solbiati, L., & Gazelle, G. S. (1999). Small hepatocellular carcinoma: Treatment with radiofrequency ablation versus ethanol injection. *Radiology, 210*, 655–661.
21. Guglielmi, A., Ruzzenente, A., Battocchia, A., Tonon, A., Fracastoro, G., & Cordiano, C. (2003). Radiofrequency ablation of hepatocellular carcinoma in cirrhotic patients. *Hepatogastroenterology, 50*, 480–484.
22. Livraghi, T., Goldberg, S. N., Lazzaroni, S., Meloni F., Ierace T., Solbiati L., et al. (2000). Hepatocellular carcinoma: Radiofrequency ablation of medium and large lesions. *Radiology, 214* (3), 761–768, Mar.
23. Poon, R. T., Ng, K. K., Lam, C. M., Ai, V., Yuen, J., & Fan, S. T. (2004). Effectiveness of radiofrequency ablation for hepatocellular carcinomas larger than 3 cm in diameter. *Archives of Surgery, 139*(3), 281–287, Mar.
24. Rossi, S., Di Stasi, M., Buscarini, E., Quaretti P., Garbagnati F., Squassante L., et al. (1996). Percutaneous RF interstitial thermal ablation in the treatment of hepatic cancer. *American Journal of Roentgenology, 167*, 759–768.
25. Sala, M., Llovet, J. M., Vilana, R., Bianchi L., Sole M., Ayuso C., et al. (2004). Initial response to percutaneous ablation predicts survival in patients with hepatocellular carcinoma. *Hepatology, 40* (6), 1352–1360, Dec.
26. Tangkijvanich, P., Anukulkarnkusol, N., Suwangool, P., Lertmaharit S., Hanvivatvong O., Kullavanijaya P., et al. (2000). Clinical characteristics and prognosis of hepatocellular carcinoma: Analysis based on serum alpha-fetoprotein levels. *Journal of Gastrointestinal Surgery, 31*, 302–308.
27. Wayne, J. D., Lauwers, G. Y., Ikai, I., Doherty, D. A., Belghiti, J., Yamaoka Y., et al. (2002). Preoperative predictors of survival after resection of small hepatocellular carcinomas. *Annals of Surgery, 235*(5), 722–730, May.
28. Wildi, S., Pestalozzi, B. C., McCormack, L., & Clavien, P. A. (2004). Critical evaluation of the different staging systems for hepatocellular carcinoma. *British Journal of Surgery, 91*(4), 400–408, Apr.
29. The Cancer of the Liver Italian Program (CLIP) Investigators. (2000). Prospective validation of the CLIP score: A new prognostic system for patients with cirrhosis and hepatocellular carcinoma. *Hepatology, 31*, 840–845.
30. Ueno, S., Tanabe, G., Sako, K., Hiwaki T., Hokotate H., Fukukura Y., et al. (2001). Discrimination value of the new western prognostic system (CLIP score) for hepatocellular carcinoma in 662 Japanese patients. Cancer of the Liver Italian Program. *Hepatology, 34*, 529–534.
31. Zhao, W. H., Ma, Z. M., Zhou, X. R., Feng, Y. Z., & Fang, B. S. (2002). Prediction of recurrence and prognosis in patients with hepatocellular carcinoma after resection by use of CLIP score. *World Journal of Gastroenterology, 8*, 237–242.

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Laparoscopic Management of Rectal Prolapse

Conor P. Delaney

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Abstract Rectal prolapse can be a disabling condition for those affected. Treatment has historically been by transanal or abdominal approaches, with transanal approaches tending to have lower morbidity, and abdominal approaches having lower recurrence rates. With the advent of laparoscopy, many of the numerous described abdominal operations have been reported with a minimally invasive approach. Although few randomized data exist, laparoscopic operations appear to provide equal rectal fixation to open surgery, with less morbidity. Coexistent symtoms such as fecal incontinence and constipation must be evaluated before surgery, so that the operation can be tailored to the needs of the individual patient. Patients with severe constipation are often offered a concomitant sigmoid resection, although this does increase the potential for complications. Patients with incontinence, diarrhea, or otherwise normal function can be offered a rectopexy without resection.

Keywords Laparoscopy · Rectal prolapse

Introduction

More than 100 surgical operations were reported for the treatment of rectal prolapse, and these can be grouped into perineal and abdominal approaches. Choosing the optimal repair for an individual patient involves consideration of many factors, including overall health and preexisting bowel function relating to a history of constipation (present in 25–50%) or fecal incontinence (present in up to 75%).[1]

Rectal prolapse initially only occurs with defecation and straining. Later as the tissues become more lax, the rectum may prolapse with the mildest straining, an upright position, or even at rest. Tenesmus, bleeding, and mucus discharge are associated symptoms. Incontinence may range from mucus leakage to complete fecal incontinence. A history of bladder and gynecological dysfunction and prolapse should be sought in appropriate cases.

On physical examination, the anus may be patulous. Visualization of everted bowel with concentric folds allows definitive diagnosis. If prolapse is not evident, the patient should be examined while straining on the commode, as the left lateral or jacknife positions are frequently inadequate to reproduce the prolapse, and thus are inadequate to rule out a diagnosis of prolapse. If a small prolapse is difficult to distinguish from hemorrhoids, the index finger should be introduced to display the sulcus between the layers of prolapsed bowel and the anal sphincter. Female patients are examined for an anterior enterocele or rectocele. The sphincters are carefully examined. Proctosigmoidoscopy is the minimum requirement to look at the mucosa and evaluate for a lead point or other pathology. The majority of patients have already been examined by colonoscopy because of their age and the presence of rectal bleeding that is associated with their presentation.

Some authors advocate evaluating transit time in those with constipation. We selectively perform this in patients

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with a history of severe constipation and associated sphincter weakness, thus avoiding a colectomy in patients without definite slow transit.[1] Those with chronic straining at stool should have evaluation for paradoxical contraction of the anal sphincters or anismus so that biofeedback therapy may be instituted before repair of the prolapse. The clinical and cost benefit of routine preoperative studies, including anal manometry, pudendal nerve terminal motor latency, colonic transit studies, and defecography, is unclear.

Surgical Considerations

Altemeier popularized the procedure of perineal rectosigmoidectomy in the 1960s, and this operation remains the ideal option for patients presenting with an incarcerated, gangrenous prolapse. However, in most comparisons between abdominal and perineal repairs, recurrence rates were higher with perineal approaches, and complication rates may be significant.[2] A recent Cochrane review states that there is insufficient evidence to choose between surgical routes based on recurrence rates, although there is limited evidence available. Perineal surgery may increase incontinence rates, and laparoscopy may reduce complications.

Abdominal repairs involve mobilization of the rectum and fixation to the sacral promontory with suture, or a prosthetic material, or mesh. Abdominal repairs may be performed with or without a concomitant bowel resection. Resection rectopexy incorporates resection of the sigmoid and upper rectum, with recurrence rates in the order of 2 to 8%, but with the added morbidity of a colorectal anastomosis. Although recurrence rates are generally less than 10%, anterior wraps may be complicated by stenosis and obstruction. Posterior fixation, as per Wells, avoids stenosis and may reduce constipation. The rectum is mobilized by dissecting posteriorly in the presacral space down to the pelvic floor, and there is some discussion as to whether the lateral ligaments should be divided.

The Laparoscopic Approach

Laparoscopy reduces postoperative pain, allows earlier introduction and tolerance of diet, and can shorten the length of hospital stay, while being cost-efficient.[3,4] A recent meta-analysis has shown that complications are significantly less frequent with laparoscopic colon surgery, especially so for wound complications. As the wound is the major physiological insult in a rectal prolapse repair, laparoscopy is particularly suitable for these procedures. In fact, a nonresectional rectopexy becomes analogous to a Nissen's repair in general surgery, where the wound is also the primary physiological insult for the patients.

The surgical approach is identical to that used in open surgery.[5] The presacral space is entered and a posterior rectal mobilization is performed to the level of the pelvic floor. The "lateral ligaments" are not routinely divided. In a Wells rectopexy, a precut mesh is passed down a port and tacked or sutured to the sacral promontory in the midline.[6] The edges are then sutured to the lateral mesorectal tissue to maintain rectal support. In patients having a resection, the upper rectum is transected with an endoscopic stapler and passed out through a 4-cm left lower quadrant muscle splitting incision. The proctosigmoidectomy is completed and the anvil of a circular stapler is inserted in the proximal bowel before it is returned to the abdomen. The anastomosis to the rectal stump is completed before suturing the lateral mesorectal tissue to the sacral promontory for additional support.

We recently presented a series of 38 laparoscopic rectopexy repairs, using the Wells or resection rectopexy, depending on a symptom based algorithm.[1,5] Median hospital stay was 2.3 days for Wells and 3.6 days for resection rectopexy patients, with no recurrences at that time. This series was recently updated in a case-matched fashion with patients undergoing surgery by an open approach. For the 109 laparoscopic repairs, hospital stay was 3 days (compared to 6 for open surgery), and recurrence rates were 8% for laparoscopic surgery vs 5% for open (p=0.37).[7]

Algorithm for Management

Clearly, there are many options for repair of rectal prolapse. A major review by Kim et al.[2] over a 19-year period studied 188 perineal rectosigmoidectomies and 160 abdominal resection rectopexy patients. Although the morbidity was lower for perineal repairs, recurrence rates were increased from 5 to 16%. In our opinion, laparoscopy helps reduce postoperative morbidity rates, allowing for a safe "abdominal" repair in more patients. This allows the reduced recurrence rates of abdominal surgery to be offered to the older patient who would traditionally be offered a perineal repair.

Thus, patients who present to these authors are managed by laparoscopic Wells rectopexy if they have no constipation, or in the presence of diarrhea or incontinence. Those with constipation are managed by laparoscopic resection rectopexy. Perineal approaches are reserved for those who are medically very unfit, and Delorme and Altemeier approaches are used, with a preference for the Delorme approach in patients with poor continence.[1,5,7]

Late Complications and Recurrence

Mucosal prolapse may occur in 5–10% of cases, and is not considered to be a true recurrence. This is treated with elastic banding, or excision under local anesthesia. Patients with incontinence should be observed for improvement for up to 6 to 12 months, unless symptoms are extremely severe and warrant earlier operative sphincter repair. Many improve after the repair, and the anus is not being dilated by the prolapse.

Although solitary rectal ulcer, present in approximately 12% of prolapse patients, is often considered as a complicating issue, it should probably be treated separately. If the ulcer is associated with prolapse, then repair the prolapse. If not, then initial treatment of the ulcer involves correction of straining and defecation habit.

Internal intussusception, diagnosed by barium studies or defecating proctography, is a diagnosis to be wary of as an indication for surgical repair. In fact, many asymptomatic patients may have an internal intussusception on defecating proctography. These patients should be fully evaluated for other possible causes of their symptoms, including an evaluation for anismus, and pelvic floor dyssynergia. Surgical repair is often avoidable. Rectal prolapse in conjunction with urogenital prolapse or other pelvic floor disorders mandates a combined approach by colorectal, gynecological, and urological surgeons.[8]

Recurrent rectal prolapse generally occurs 18 to 24 months after the index operation. A repeat repair usually provides an excellent outcome for treatment of the prolapse; however, there is little improvement in other functional problems such as constipation and incontinence.[9] These patients should probably be extensively investigated before repeat repair to elucidate factors that might predispose to recurrence, such as slow-transit constipation and anismus. If a resection is performed for recurrence, any prior anastomoses must be resected to avoid leaving an ischemic segment. Some authors would suggest a perineal repair after a failed abdominal repair and vice versa. In fact, both types of repair are feasible and there is inadequate evidence in the literature to determine strategy. Our preference would be to perform repeat abdominal repair except in the most unfit patient, reserving laparoscopy for those with a failed perineal approach.

References

1. Delaney CP, Senagore AJ. Rectal prolapse. In Fazio VW, Church JM, Delaney CP eds. Current Therapy in Colon and Rectal Surgery, 2nd ed. Philadelphia, PA: Elsevier, Mosby Inc., 2005, pp 131–134
2. Kim DS, Tsang CB, Wong WD, Lowry AC, Goldberg SM, Madoff RD. Complete rectal prolapse: evolution of management and results. Dis Colon Rectum 1999;42:460–466.
3. Delaney CP, Kiran RP, Senagore AJ, Brady K, Fazio VW. Case matched comparison of clinical and financial outcome after laparoscopic or open colectomy. Ann Surg 2003;238:67–72.
4. Senagore AJ, Delaney CP. A critical analysis of laparoscopic colectomy at a single institution: lessons learned after 1000 cases. Am J Surg 2006;191(3):377–380.
5. Madbouly K, Senagore AJ, Delaney CP, Duepree HJ, Brady KM, Fazio VW. Clinically based management of rectal prolapse: a comparison of laparoscopic Well's procedure versus resection rectopexy. Surg Endosc 2003;17:99–103.
6. Himpens J, Cadiere GB, Bruyns J, Vertruyen M. Laparoscopic rectopexy according to Wells. Surg Endosc 1999;13:139–141.
7. Kariv Y, Delaney CP, Casillas S, Hammel J, Nocero J, Bast J, Brady K, Fazio VW, Senagore AJ. Long-term outcome after laparoscopic and open surgery for rectal prolapse. Surg Endosc 2006;20:35–42.
8. Sullivan ES, Longaker CJ, Lee PY. Total pelvic mesh repair: a ten-year experience. Dis Colon Rectum 2001;44:857–863.
9. Hool GR, Hull TR, Fazio VW. Surgical treatment of recurrent complete rectal prolapse: a thirty-year experience. Dis Colon Rectum 1997;40:270–272.

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Stapled Transanal Rectal Resection (STARR) for Rectocele

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Abstract Patients with obstructed defecation complain of an inability to initiate rectal emptying, incomplete evacuation, pelvic pressure or excessive straining at stool. The pathophysiologic features of obstructed defecation include an increased anterior-posterior diameter of the rectum, decreased rectal compliance and an increased sensory threshold volume. Recently, there has been interest in the transanal resection of the rectum for obstructed defecation with the developement of endoanal staplers and techniques specifically for these purpose. Stapled transanal rectal resection (STARR), in the only large series reported, decreased the anterior-posterior diameter of the rectum, restored rectal compliance and decreased the rectal sensory threshold with an associated improvement in incomplete evacuation in 81.1%, digital assistance to defecate in 83.4%, pelvic pain in 43.3%, and the need for laxatives 43.3% of patients. Risks of the procedure included stenosis in 3.3%, urgency in 1.1% and incontinence of flatus in 1.1% of patients. These data suggest that the STARR procedure is an effective management option for obstructed defecation with an acceptable risk of complications.

Keywords Stapled transanal rectal resection · Rectocele · Obstructed defecation

Introduction

Obstructed defecation is a broad term used to describe the condition of patients with defecatory dysfunction and constipation. While patients will frequently complain of constipation, a condition medically defined as less than three stools per week, they will describe symptoms of an inability to initiate rectal emptying, incomplete evacuation, pelvic pressure, or excessive straining at stool. Common causes of obstructed defecation include pelvic dyssynergy, rectocele, rectal intussusception, and enterocele. The pathophysiologic features of obstructed defecation include an increased anterior–posterior diameter of the rectum, decreased rectal compliance, and an increased sensory threshold volume.

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Rectocele

A rectocele is a herniation of the anterior rectal wall into the posterior vagina. Surgical repair has been recommended when the rectocele is greater than 3 cm in depth, if there is significant barium trapping on defecography, or if digital assistance of defecation is frequently necessary for satisfactory emptying. Once the decision for surgery has been made, rectoceles are most commonly addressed via a transvaginal or a transrectal approach. [1]

Transvaginal rectocele repair has been reported to result in a decrease in the need for digital assistance of defecation and an improvement in the symptom of constipation in 37–43% and 17–66% of patients, respectively. However, 18–37% of patients will complain of postoperative dyspareunia and as many as 36% will report a problem with fecal incontinence.

Transrectal repair of rectoceles has been reported to result in a decrease in the need for digital assistance of defecation and an improvement in the symptom of constipation in 54–100% and 48–71% of patients, respectively. [1] Randomized studies comparing transvaginal to transrectal rectocele repair have found comparable relief of symptoms, but the incidence of postoperative dyspareunia to be significantly less with the latter technique. [2]

Table 1 Results After Stapled Transanal Rectal Resection for Rectocele

	Preop (%)	3 mo (%)	6 mo (%)	12 mo (%)
Incomplete evacuation	98.9	24.4	21.1	18.9
Digital assistance to defecate	87.8	5.6	4.4	4.4
Pelvic pain	63.3	25.6	21.1	20
Laxatives	52.2	13.3	10	10
Enema	44.4	4.4	2.2	2.2
Abdominal pain	28.2	16.7	13.4	12.2
Rectal bleeding	17.8	4.4	2.2	2.2

Recently, there has been interest in the transanal resection of rectoceles with the development of endoanal staplers and techniques specifically for this purpose. Stapled transanal resection of the rectum in the only large series reported, decreased the anterior–posterior diameter of the rectum, restored rectal compliance and decreased the rectal sensory threshold. [3] The improvement in symptoms with this technique is shown in Table 1. Complications at 1 year of follow-up included stenosis in 3.3%, urgency in 1.1%, and incontinence of flatus in 1.1% of patients.

Rectal Intussusception

Rectal intussusception, also called internal procidentia and incomplete or occult rectal prolapse, is more likely a consequence of excessive straining to defecate rather than a cause of obstructed defecation. In addition to the usual symptoms associated with obstructed defecation, 50% of patients will complain of fecal incontinence. On proctoscopic examination, 49% of patients with rectal intussusception will be found to have either hyperemia and edema of the anterior rectal wall, colitis cystica profunda, or a solitary rectal ulcer. Surgical therapy is considered for patients with an intussusception, which extends distal to the puborectalis sling and is associated with either fecal incontinence or the proctoscopic findings described above. Both abdominal and perineal approaches have been used to manage rectal intussusception, but the paucity of good comparative studies precludes definite recommendations about the optimal approach. Recently, a series of 10 patients with rectal intussusception managed by stapled transanal resection of the prolapsing segment and a mean follow-up of 19+9 months have been reported. [4] Results included a decrease in the mean defecation scores from 13+3 preoperatively to 4+3 postoperatively ($p<0.05$). Complications in this series included one patient each with prolonged pelvic pain and recurrent symptoms.

Reference

1. Zbar AP, Lienemann A, Fritsch H, Beer-Gabel M, Pescatori M. Rectocele: pathogenesis and management. Int J Colorectal Dis 2003;18:369–384.
2. Kahn MA, Stanton SL, Kumar DA. Randomized prospective trial of posterior colporrhaphy vs transanal repair of rectocele: preliminary findings. Int Urogynecol J 1997;8:246–247.
3. Boccasanta P, Venturi M, Stuto A, Bottini C, Cavigilia A, Carriero A, Mascagni D, Mauri R, Sofo L, Landolfi V. Stapled transrectal rectal resection for outlet obstruction: a prospective multicenter trial. Dis Colon Rectum 2004;47:1285–1297.
4. Ommer A, Albrecht K, Wenger F, Walz MK. Stapled transanal rectal resection (STARR): a new option in the treatment of obstructive defecation syndrome. Langenbecks Arch Surg 2006;391:32–37.

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Transanal Endoscopic Microsurgery (TEM) Resection of Rectal Tumors

Mark H. Whiteford

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Abstract Local excision of rectal cancer is an attractive alternative to avoid the morbidity associated with radical rectal surgery. Oncologic concerns, specifically the inability to fully assess the status of the perirectal lymph nodes and the risk of local recurrence after local excision remain significant barriers to widespread adoption of this technique. Transanal endoscopic microsurgery is an alternative minimally invasive technique used for transanal excision of rectal polyps and tumors. It offers the advantage of better exposure, magnified stereoscopic view, and greater reach into the middle and upper rectum. This technique, combined with careful patient selection, has demonstrated optimistic results compared to standard transanal techniques and even total mesorectal excision when utilized for certain early rectal cancers.

Keywords Proctectomy · Rectal tumor · Transanal endoscopic microsurgery

Introduction

Standard surgical therapy for rectal cancer is a radical proctectomy encompassing a total mesorectal excision with either an abdominoperineal resection or low anterior resection. Whereas this technique optimizes oncologic outcomes, it does not come without significant morbidity. These morbidities include defecatory dysfunction, fecal incontinence, urinary and sexual dysfunction, postoperative convalescence, and often the need for temporary or permanent fecal diversion.

In an effort to reduce the morbidity and mortality of rectal cancer therapy, many local options have been explored, including fulguration, endocavitary radiation, transsphincteric or transsacral approaches, transanal full-thickness excision, and transanal endoscopic microsurgery (TEM). These latter two options have gained the most favor because of their low morbidity and ability to retrieve a complete specimen for histologic evaluation. In the United States, transanal excision of rectal neoplasms is the most commonly performed local surgical therapy for rectal neoplasms. One of the biggest shortcomings of this technique is that it does not include a formal lymphadenectomy and thereby leaves the status of the mesorectal lymph nodes uncertain. Because local excision can only cure tumors confined to the rectal wall, appropriate patient selection is critical to successful treatment. The risk of lymph node metastasis increases with depth of tumor invasion and the presence of unfavorable histology such as lymphovascular invasion and a high-grade differentiation. Therefore, current recommendations are to limit transanal excision to palliation and low-risk superficial cancers, i.e., T1 cancers with no unfavorable histologic features and T2 cancers augmented by chemoradiotherapy.

Methods

Transanal endoscopic microsurgery is an alternative to transanal excision of rectal tumors. The limited reach of instruments and poor visibility afforded through retractors during standard transanal excision led Dr. Gerhard Buess to devise the TEM equipment (Richard Wolf Medical Instru-

This study was presented at the Society for Surgery of the Alimentary Tract meeting, May 23, 2006, Los Angeles, CA, USA

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Figure 1 Transanal endoscopic microsurgery operating telescope and instruments.

ments Corporation, Knittlingen, Germany) and procedure in 1984. This device includes a 12 or 20 cm operating proctoscope with an airtight faceplate and operating portals. This allows for pneumoinsufflation of the rectum and passage of an optical telescope and modified laparoscopic instruments (Fig. 1). It provides access to lesions up to 25 cm from the anal verge with markedly improved visualization over transanal excision. This improved exposure and visibility means that lesions larger than 3 cm and circumferential full thickness sleeve resections are technically feasible in experienced hands.

The operative technique for TEM surgery follows the principals of standard transanal excision. The patient is positioned with the lesion in the dependent portion of the surgical field. The dissection margins are scored 1 cm away from the gross lesion using electrocautery. Epinephrine solution is injected underneath the lesion to assist with hemostasis. The lesion is resected with the inclusion of mesorectal tissue if desired (Fig. 2). The resultant defect is closed using running absorbable sutures. As a substitute for tying knots, the end of the suture is secured using a specialized metallic bead clip.

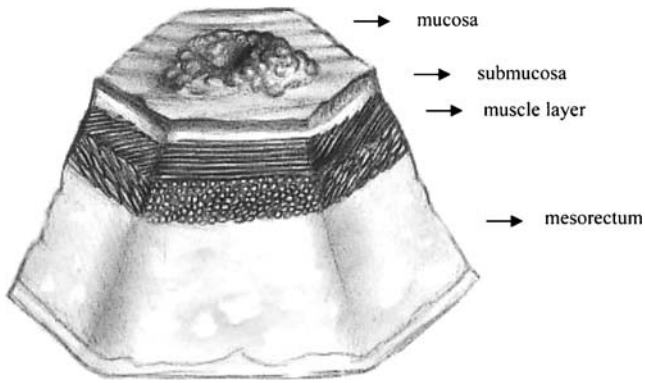

Figure 2 Schematic representation of the removed specimen by TEM (taken from[2]).

Results and Discussion

Recurrence After Local Excision of Adenoma Transanal endoscopic microsurgery is a highly effective technique for local excision of rectal adenomas. In 1996, Smith et al.[1] reported the pooled data of the initial United States experience, which reviewed the resection of 82 adenomas of a mean size of 4 cm. Recurrence rate after TEM was 11%. The same year, Winde et al.[2] published a randomized trial comparing 98 patients undergoing TEM to 90 patients undergoing the transanal excision. The complication rates were similar; however, the recurrence rate was 6 vs 22%, respectively, in favor of TEM.

Recurrence After Local Excision for T1 Cancers Numerous large case series have recently drawn attention to a higher than previously reported rate of local recurrence after transanal excision of favorable T1N0 rectal cancers (Table 1). The recurrence rates ranged from 7–18%, with overall survival 10–20% lower than with radical surgery. In the absence of randomization, these series may have been subject to selection bias whereby radical oncologic surgery was offered to healthy patients and transanal excision to the less fit individuals. This may account for at least some of the differences in overall survival. In studies reporting cancer-specific survival following the transanal excision, groups report 5-year survival is approximately 90%.

Floyd and Saclarides[3] in 2006 presented the largest case series in the United States reporting 53 patients with a pathologic T1 rectal cancer excised using TEM. With a mean follow-up of 2.8 years, they reported 7.5% local recurrence rate with no cancer-related deaths. Winde et al.[2] in 1996 reported the only randomized trial for uT1N0 rectal cancer in which 50 patients were randomized to TEM vs

Table 1 Local Recurrence and 5-year Survival Rates for T1N0 Rectal Cancer Treated by Transanal Excision and Oncologic Resection

	Local Recurrence		5-year Survival	
	TAE (%)	Resection (%)	TAE (%)	Resection (%)
Mellgren 2000	18	4	72	80
Paty 2002	14	ns	92	ns
Nascimbeni 2004	7	3	72	90
Madbouly 2005	17	ns	75	ns

TAE=transanal excision, Resection=oncologic resection, ns=not stated

low anterior resection. After a mean follow-up of 41–46 months, there was no difference in local recurrence between TEM (4%) and low anterior resection (0%). Survival was 96% in each group.

Recurrence After Local Excision for T2 Cancers Rectal cancers that have invaded into the muscularis propria (T2 tumors) have a high incidence of lymph node metastasis and local excision alone results in a high rate local recurrence. For this reason supplemental chemoradiation is recommended for T2 rectal cancers following local excision for patients unwilling or unable to undergo radical surgery. Because of the enhanced cytoreductive activity of combined chemo-radiation in tumors that maintain intact blood supply, there has been a recent trend towards preoperative chemoradiation. Read et al.[4] in 2004 reported that histologic response to neoadjuvant radiotherapy correlated with nodal status in patients undergoing radical rectal cancer surgery. Specifically, the patients post radiation ypT0-1 tumors had a 2–4% incidence of harboring metastatic lymph nodes compared to a 23% lymph-node-positive rate in patients who had post radiation therapy ypT2 staging.

This concept was recently subjected to a randomized trial by Lezoche et al.[5] Three-year follow-up data on the initial 40 patients were published in 2005. These patients were treated with neoadjuvant chemoradiation then randomized to either TEM or laparoscopic low anterior resection with total mesorectal. Local recurrence was 5% in each group and survival was 95 vs 83%, favoring TEM. Five-year follow-up data was presented at the Society for Surgery of the Alimentary Tract meeting in 2006. The study had now accrued 70 patients with a 3% local recurrence in each arm and a survival probability of 0.971 vs 0.943. This data suggests that there may be no oncologic advantage to radical surgery following this protocol.

Most of the recurrences after a transanal excision of a rectal cancer occur locally. Proposed mechanisms for this are progression of occult mesorectal or nodal metastasis, or perioperative wound implantation of viable tumor cells. Transanal endoscopic microsurgery may improve upon these two issues. TEM allows for the potential of a deeper en bloc resection of the mesorectum, which will permit improved sampling of the peritumoral tissue and lymph nodes. In addition, better visualization and more precise instrumentation may facilitate less traumatic tissue handling and more room to operate around a bulky tumor.

Conclusion

Transanal excision for rectal cancer can be performed with low morbidity and mortality. The risk of incontinence, sexual dysfunction, urinary dysfunction, and the need for a colostomy is quite low. Unfortunately, the status of the mesorectal lymph nodes is not known and poor oncologic results can occur if patients are not carefully selected.

From this data, it appears that TEM may benefit patients with early rectal neoplasia with reduced local recurrence and improved survival rates compared to transanal excision. TEM with curative intent should be limited to adenomas and favorable T1 cancers as well as T2 cancers after neoadjuvant chemoradiation.

References

1. Smith LE, Ko ST, Sachlarides T, Caushaj P, Orkin BA, Khanduja KS. Transanal endoscopic microsurgery. Initial registry results. Dis Colon Rectum 1996;39(suppl):S79-84.
2. Winde G, Nottberg H, Keller R, Schmid KW, Bünte H. Surgical cure for early rectal carcinomas (T1): Transanal endoscopic microsurgery vs. anterior resection. Dis Colon Rectum 1996;39:969-976.
3. Floyd ND, Saclarides TJ. Transanal endoscopic microsurgical resection of pT1 rectal tumors. Dis Colon Rectum 2006;49:164–168.
4. Read TE, Andujar JE, Caushaj PF, Johnston DR, Deitz DW, Myerson RJ, Fleshman JW, Birnbaum EH, Mutch MG, Kodner IJ. Neoadjuvant theraphy for rectal cancer:histologic response of the primary tumor predicts nodal status. Dis Colon Rectum 2004;47:825–831.
5. Lezoche E, Guerrieri M, Paganini AM, D'Ambrosio G, Baldarelli M, Lezoche G, Feliciotti F, DeSanctis A. Transanal endoscopic vs total mesorectal laparoscopic resections of T2-N0 low rectal cancers after neoadjuvant treatment: A prospective randomized trial with a 3-year minimum follow-up period. Surg Endosc 2005;19:751-56.

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Lymph Node Dissection Impact on Staging and Survival of Extrahepatic Cholangiocarcinomas, Based on U.S. Population Data

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Abstract Cholangiocarcinomas (CC) frequently demonstrate lymphatic spread. We investigated lymph node (LN) counts after resection of extrahepatic CC and survival based on the SEER 1973–2004 database. Out of 20,068 CC patients, 1,518 individuals were selected based on M0 stage and at least one LN examined. Primary cancer sites included gallbladder (29%), extrahepatic bile ducts (26%), and intrapancreatic/ampullary bile ducts (45%); 42% of patients were LN-positive. The median number of LNs examined was four (range 1–39). Median survival was 37 months for LN-negative and 16 months for LN-positive cancers. Multivariate prognostic variables were the number of positive LNs, primary site, age (all at $p<0.0001$), gender ($p=0.002$), size ($p=0.005$), T category ($p=0.009$), and total LN count (or number of negative LNs obtained, $p=0.01$). The impact of total LN counts was seen in LN-negative (median survival, 1 vs 10 or more LNs examined: 27 vs 51 months, $p=0.002$) and LN-positive disease (10 vs 22 months, $p<0.0001$). Survival prediction of extrahepatic CCs is strongly influenced by total LN counts and numbers of negative LNs obtained. Although the resulting incremental benefit is small, dissection and examination of 10 or more LNs should be considered for curative intent resections.

Keywords Extrahepatic cholangiocarcinoma · Lymph node count · Gallbladder cancer · Ampullary cancer · Lymphadenectomy · Survival

Background

Cancers of the biliary tract and gallbladder are among the most aggressive human malignancies. According to the American Cancer Society, 8,570 new cases of extrahepatic cholangiocarcinomas (CC) and 3,260 deaths are expected to occur in 2006[1]. Many patients present with advanced disease, and lymphatic metastatic involvement is common[2,3]. Operative therapy of biliary cancer remains the only potentially curative treatment modality, but is associated with high recurrence rates even after complete resection[2,4]. Adjuvant treatment strategies that could improve survival after resection of CC have not been firmly established. To optimize locoregional tumor control and subsequent survival, attempts have been made to enhance the radicality of surgical resection. Several retrospective series have reported single-institution experiences with extended lymph node dissection (ELND), primarily in Eastern patients[5–9]. While the resulting morbidity and mortality have been acceptable, it is not possible to evaluate survival improvements due to uncertain factors of patient selection and due to a lack of randomized trials. Only one prospective randomized trial of radical pancreatoduodenectomy with extended lymphadenectomy includes a smaller cohort of patients with periampullary CC[10]. This trial has failed to demonstrate an obvious survival advantage as a result of more extensive regional dissections; actuarial overall survival (OS) at five

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years was similar between the dissection groups, both for ampullary cancers (56% after standard vs 60% extended dissection) and bile duct cancers (23 vs 11%, respectively)[10]. Because of the relatively low prevalence of resectable CC and the lack of a uniform surgical approach to this disease, it does not appear to be feasible that potential benefits to ELND will be proven through an appropriate randomized trial with sufficient power. Despite this challenge, the rationale for removing regional lymph nodes (LNs) at risk for metastatic disease is rather clear. Examining a sufficient number of LNs improves staging accuracy and may aid therapeutic benefits by removing tissues at risk for recurrence. Improved survival after ELND can be postulated for those individuals with occult or obvious nodal disease, but without any other sites of metastatic tumor involvement. Similar concepts have been investigated for other cancers of the gastrointestinal system. We have shown earlier a strong relationship between total LN counts and survival of gastric and pancreatic cancers[11,12]. To test a similar concept in this different intra-abdominal malignancy, we investigated the relationship between LN numbers examined after resection of extrahepatic CC and survival by utilizing information from a large U.S. population-based database.

Patients and Methods

An extrahepatic CC data set was created from the public release version of the surveillance, epidemiology, and end result (SEER) database, covering the years 1973 to 2004[13]. Within the SEER program, clinical data are collected from 14 cancer registries throughout the U.S. Included into this analysis of extrahepatic CC were non-neuroendocrine cancers of gallbladder, extrahepatic bile duct, intrapancreatic bile duct, and ampulla. Stage information for these carcinomas was created according to the American Joint Committee on Cancer tumor, node, metastases criteria, 6th edition[14]. Out of a patient cohort with an extrahepatic CC diagnosis (n=20,068), individual patients were selected based on sufficient staging information, completely resectable pathologic stage (<T4, M0), with at least one LN examined, and sufficient information on treatment by resection and survival outcome. Individuals who had received adjuvant therapy by radiation and/or chemotherapy were kept in the analysis; those patients for whom margin positivity was documented (R1, R2 status) were excluded from the analysis. The stepwise extraction process of information from the original SEER database is shown in Fig. 1. Relationships between the number of LNs examined and OS were analyzed for the entire cohort, by diagnosis, and based on pathologic LN status. Differences in LN count between patient groups were calculated by the Wilcoxon–Mann–Whitney test. OS time, as tabulated by SEER in monthly increments, was the time from diagnosis until last contact, date of death, or the date used as a cutoff for the SEER database. Survival analyses included multivariate, univariate, and model projection calculations as shown earlier[11,12]. Cox regression was used for multivariate analyses, employing a backward elimination model for all covariates[15]. The threshold for keeping a variable in this regression model under backward elimination was p=0.05. Independent variables entered into the Cox multivariate model were primary diagnosis (gallbladder, bile duct, ampullary cancer), T stage category, tumor size (as continuous or categorical variable), grade, number of LNs examined (or number of negative LNs obtained), number of positive LNs, age at diagnosis, gender, ethnicity, year of diagnosis, and radiation or chemotherapy administered. Actuarial survival was calculated with the Kaplan–Meier method[16], and univariate comparison between groups was performed using the log-rank test[17]. In addition, we chose a linear regression model to correlate LN counts with the OS at 5 years based on Kaplan–Meier estimates for each LN count interval. The independent variable was constructed using the LN count interval midpoints. To link survival outcomes with LN count categories, we used Monte Carlo simulation for exact p values of the Kruskal–Wallis test. Additional nonparametric group comparisons of continuous variables were performed with Wilcoxon ANOVA. All calculations were performed using the SAS 8.2 statistical software package (SAS, Cary, NC, USA). Significance of differences was accepted at p<0.05.

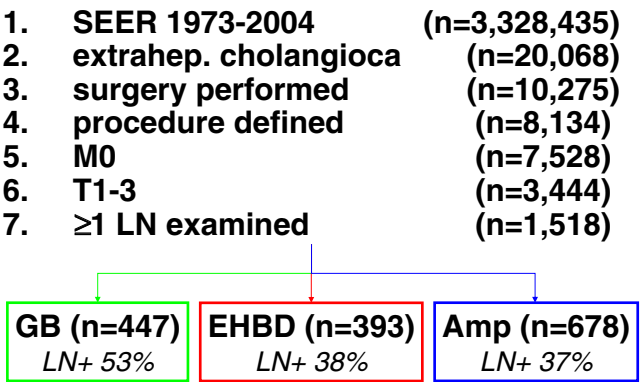

Figure 1 SEER database data extraction process.

Results

Patient Selection and Demographics

Out of a cohort of 20,068 individuals with extrahepatic CC, 10,275 patients were identified to have undergone operative therapy. Specifications for a resective procedure were found in 8,134, of which 7,528 had M0 disease extent. Sufficient

pathologic information for nonendocrine CC with T1 to T3 primary tumor extent was available for 3,444 patients. Of these, 1,518 individuals had pathologic LN information, with at least one LN examined; these represent the cohort for the following analyses. The stepwise patient selection process is depicted in Fig. 1. The median age was 68 (range 22–102). Genders were nearly evenly split, with 766 women and 752 men. Racial categories included white (n=1,224, 81%), black (n=98, 6%), and other patients (n=196, 13%). For the 1,518 individuals selected, primary cancer sites included gallbladder (n=447, 30%), extrahepatic bile ducts (n=393, 25%), and intrapancreatic bile duct/ampulla (n=678, 45%). Transmural tumors were identified in 47% and positive LNs in 42% of patients. The median tumor size was 2 cm (0–10) and did not differ by disease (gallbladder cancers=3, bile duct=2, ampullary=2). Among patients with known tumor grade (n=1,372, 90%), the differentiation degree was well (18%), moderate (49%), poor (31%), and anaplastic (2%). The majority of patients did not undergo radiation therapy (n=1129, 74%). Of those who did, 96% received their radiation postoperatively.

Number of LNs Examined by Disease and Stage Subgroups

The median number for total LNs examined within the surgical specimen was 4 (range 1–39); for positive LNs, 0 (0–34); and for negative LNs, 3 (0–37). The frequency of total LN count categories varied greatly by disease, and also by stage group. Figure 2 depicts total LN counts by diagnosis, where 72% of gallbladder cancer specimens had only one LN identified, but bile duct and ampullary cancer specimens showed a more even distribution of numeric total LN categories, with only 24 or 10% of cases stating one LN examined (p<0.0001). Ten or more LNs were found in 3% of gallbladder, 22% of bile duct, and 29% of ampullary cancers. Accordingly, median total LN counts were 1 for gallbladder (range 1–28), 5 for bile duct (1–37), and 6 for ampullary cancers (1–39). While total LN counts did not differ between nodal negative and nodal positive gallbladder cancers, total LN counts were significantly greater for N+ vs N0 stages for both bile duct and ampullary cancer groups (Fig. 3). Median positive LN counts were 1 for gallbladder (range 0–8), but 0 for bile duct (0–14) and ampullary cancers (0–34). Median negative LN counts were 1 for gallbladder (0–22), 4 for bile duct (0–37), and 5 for ampullary cancers (0–37). Various T categories did not result in different LN counts; median total counts were 4 for intramural, transmural, and even transserosal cancers (p=0.07).

Multivariate Survival Analysis

Survival calculations were performed at a median follow-up of 24 months (range 0 to 166; median follow-up for survivors was 83 months). A backwards elimination multivariate survival analysis yielded these prognostic variables: number of positive LNs, primary site, age (all at p<0.0001), gender (p=0.002), primary tumor size (p=0.005), T category (p=0.009), and total LN count (or number of negative LNs obtained, p=0.01). The risk ratios for these covariates are listed in Table 1. The statistical impact of the number of negative LNs obtained and of the total number of LNs examined was interchangeable, with similar significance levels when substituted for each other. Grade, ethnicity, adjuvant therapy, and the year of diagnosis failed to retain significance levels in this model. Separate multivariate analyses were performed for each diagnostic group; in all three analyses, the total number of LNs examined retained an independent prognostic effect (gallbladder: RR=0.93, p=0.02; bile duct: RR=0.97, p=0.007; ampullary: RR=0.98, p=0.05). For all three groups, the number of positive LNs maintained the strongest survival

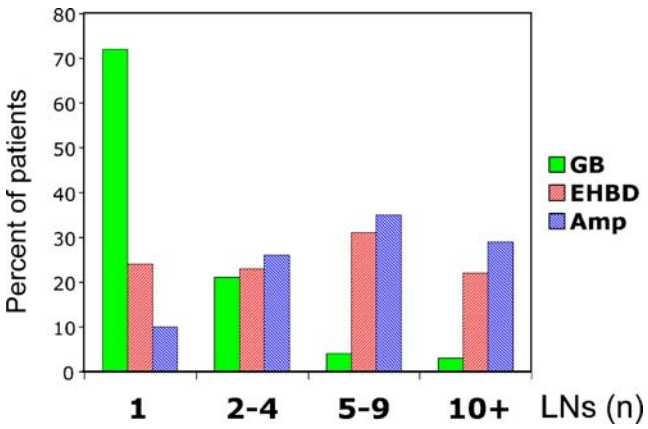

Figure 2 Frequency of categorized number of total LN counts, by disease site. *GB* gallbladder, *EHBD* extrahepatic bile duct, *Amp* ampulla.

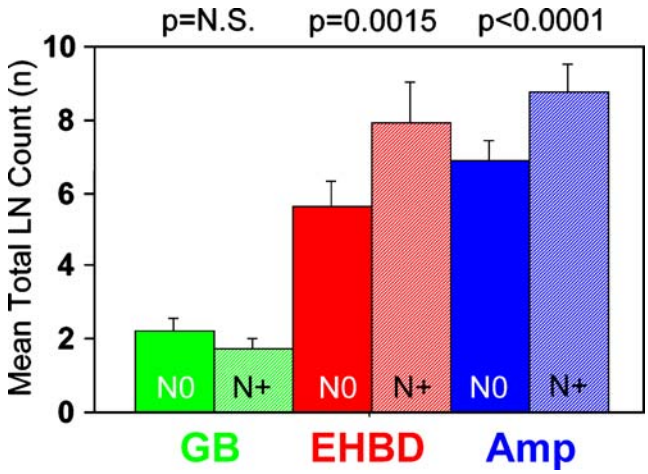

Figure 3 Total LN counts, by cancer site and pathological nodal status. *GB* gallbladder, *EHBD* extrahepatic bile duct, *Amp* ampulla.

Table 1 Multivariate Survival Analysis

Factor	Term (or unit for continuous variables)	Risk Ratio	Lower 95% Confidence Interval	Upper 95% Confidence Interval	*p* Value
Number of LNs examined	*n*	0.98	0.96	1.00	0.01
Number of positive LNs	*n*	1.13	1.10	1.15	<0.0001
Primary site	GB vs Amp	1.11	0.96	1.28	<0.0001
	BD vs Amp	1.22	1.07	1.38	
Age	Years	1.02	1.01	1.03	<0.0001
Gender	Female vs male	0.88	0.81	0.95	0.0019
Tumor size	mm	1.01	1.00	1.01	0.0046
Primary tumor extension	T1/2 vs T3	0.88	0.79	0.97	0.0086

GB=gallbladder, BD=bile duct, Amp=ampulla

impact (gallbladder: $p=0.001$; bile duct and ampullary: $p<0.0001$). Interestingly, adjuvant radiation therapy provided a negative survival impact for ampullary cancers in these calculations (RR=1.12, $p=0.05$), but not for the other diagnostic groups.

Univariate Survival Analysis

Overall actuarial survival was calculated for the entire cohort, for univariate comparisons by disease site and LN counts. Ampullary cancer patients demonstrated better survival than gallbladder or bile duct cancer patients (Fig. 4). Univariate survival impact of the T category yielded significant differences for gallbladder ($p<0.0001$) and ampullary lesions ($p=0.002$), but not for bile duct cancers ($p=0.14$). Pathologically identified nodal involvement resulted in significantly inferior survival than that for N0 disease (Fig. 5). The median actuarial OS was 37 months for LN-negative and 16 months for LN-positive cancers, with a 5-year OS of 40% (N0) vs 14% (N+) ($p<0.0001$). The OS impact of the total number of LNs examined was obvious for both LN-negative (median OS, 1 vs 10 or more LNs examined: 27 vs 51 months, $p=0.002$) and LN-positive disease (10 vs 22 months, $p<0.0001$). Comparison of survival results based on increasing categories of total LN counts showed the best results in patients with 10 or more LNs examined (Table 2), both for median OS and 5-year survival. This trend of better survival results with increasing total LN counts was also visible for all subgroups selected by both nodal status and disease site, always in favor of total LN counts of 10 or more (Table 3). The impact of increasing total LN counts on OS was most pronounced in N+ patients with one or two positive LNs, and was also observed in patients staged as N0 (Fig. 6).

Cutpoint Survival Analysis

In an attempt to identify the optimal LN count cutoff, survival comparisons were created for the entire cohort at increasing total LN counts between 1 and 20. Greatest OS differences were observed between total LN counts of one to three (at $p<0.0001$), but significant differences were still identified at a total LN count cutoff of 10 ($p=0.01$). Generally, survival differences at each cutoff value between 1 and 10 always favored the higher LN count group, with a resulting superior survival in the higher LN count group. When similar cutpoint analyses were performed for N0 patients only, the significance range of survival differences with increasing total LN count cutoff points extended to a count of 8, at $p=0.025$. In an analysis for nodal-positive patients, survival differences favoring higher total LN counts were observed for cutoff values up

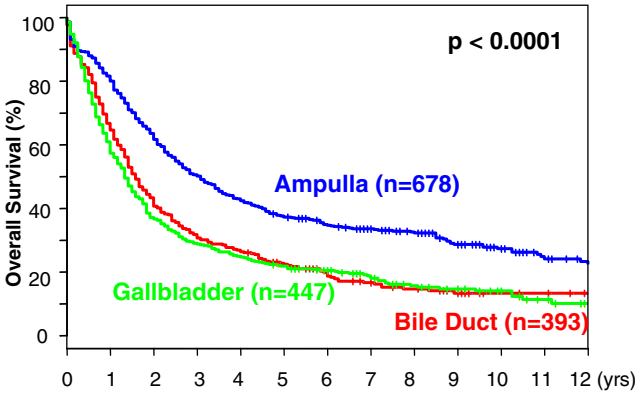

Figure 4 Actuarial OS, by disease site.

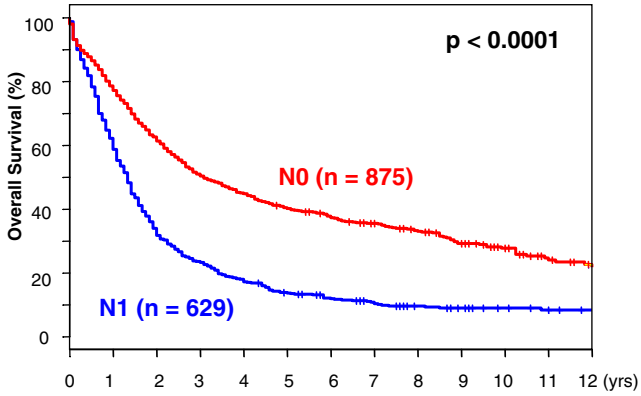

Figure 5 Actuarial OS, by nodal status (all disease sites).

Table 2 Actuarial OS by Total Number of LNs Examined

	Total LNs Examined (*n*)			
	1	2 to 4	5 to 9	10+
5-year OS (%)	19	27	34	39
Median OS (months)	16	18	39	40

All sites combined

to 12 (p=0.003). Cutpoint analyses for the separate diagnostic groups failed to show significant OS differences, with the exception of a total LN count of two or more (vs one only) for gallbladder cancers (p=0.04) and bile duct cancers (p=0.036).

Survival Impact of Negative LN Counts

Numbers of negative LNs showed an obvious impact on survival, as listed in Table 4. The biggest difference in survival was observed between negative LN counts of zero and one, but best survival outcomes were consistently observed with 10 or more negative LNs. This impact of increasing counts of negative LNs on OS in the entire patient cohort is displayed in Fig. 7. When disease sites and nodal status were analyzed separately, median survival varied somewhat between increasing negative LN count categories (Table 5). For N+ disease subgroups, at least one negative LN was linked to a median survival increase of up to 9 months. In both N0 and N+ subgroups, 10 or more negative LNs were associated with the best survival results achieved, with the exception of N+ ampullary cancers.

Projected Numeric LN Impact on OS

To create prognostic information related to LN count impact on survival, a projection model was created based on the statistically assumed linearity as best fit. The resulting curve based on increasing total LN count categories is depicted in Fig. 8. Baseline survival at 5 years, based on only one LN examined, was calculated at 21%. Despite the obvious slope of the graph due to a higher LN count impact, the model failed to reach statistical significance at p=0.074. Similarly, a model created for negative LN count impact on survival did show a gradual increase in survival over baseline, but did not reach statistical significance either. Calculation results and statistical implications of both analyses are listed in Table 6.

Table 3 Median Actuarial OS by Total Number of LNs Examined, by Disease Site and Nodal Status

	Total LNs Examined (*n*)		
	1 to 2	3 to 9	10+
All sites, N0	27	22	51
All sites, N1	11	16	44
Gallbladder, N0	27	22	51
Gallbladder, N1	11	16	40
Bile duct, N0	21	26	34
Bile duct, N1	13	14	16
Ampulla, N0	51	53	86
Ampulla, N1	19	21	24

Median survival time in months

Early Postoperative Deaths and LN Numbers

We investigated whether the removal of increasing LN numbers was associated with worsened postoperative mortality. In the entire cohort, there were 105 deaths (7.0%) within 1 month of diagnosis, and 174 deaths (11.6%) within 3 months of diagnosis. In the combined cohort, total LN counts did not correlate with postoperative mortality (both by logistic regression and chi-square analyses). Similarly, disease site, T category, or overall stage did not correlate with death within 3 months. In contrast, age greater than 70 was associated with increased mortality at 1 month (9.1 vs 5.3%, p=0.005) and at 3 months (16 vs 8.1%, p<0.0001). Additionally, radiation therapy was significantly correlated with death within 1 and 3 months. Lethal events within 3 months occurred in 2.9% of patients who underwent radiation, and 14.8% among those who did not (p<0.0001).

Discussion

Cancers originating in various sites of the extrahepatic biliary system exhibit some inherent differences in survival hazard, as confirmed in this analysis. Nevertheless, only a complete resection, as defined by negative margin (R0) status, can be considered a therapeutic modality with curative potential. How much a regional dissection for the purpose of removing LNs at risk for metastatic disease is able to enhance or contribute to this curative potential remains debated and unproven. Any substantial benefit to more extensive regional LN dissection would depend much on the presence of mechanisms for nonnodal cancer progression, and the likelihood that metastatic activity is indeed limited to a few regional LNs. For all CCs at question, the likelihood for metastatic involvement truly limited to LNs is relatively small, but perhaps larger than that observed in pancreatic cancer or advanced-stage gastric cancer[18,19]. Although positive LNs in gastrointestinal cancers, especially pancreatobiliary cancer, are primarily thought to carry prognostic importance, but not therapeutic relevance[20], it is possible that the removal of involved LNs can induce a curative result in a small patient subset, which, if large enough, can lead to a measurable outcomes benefit.

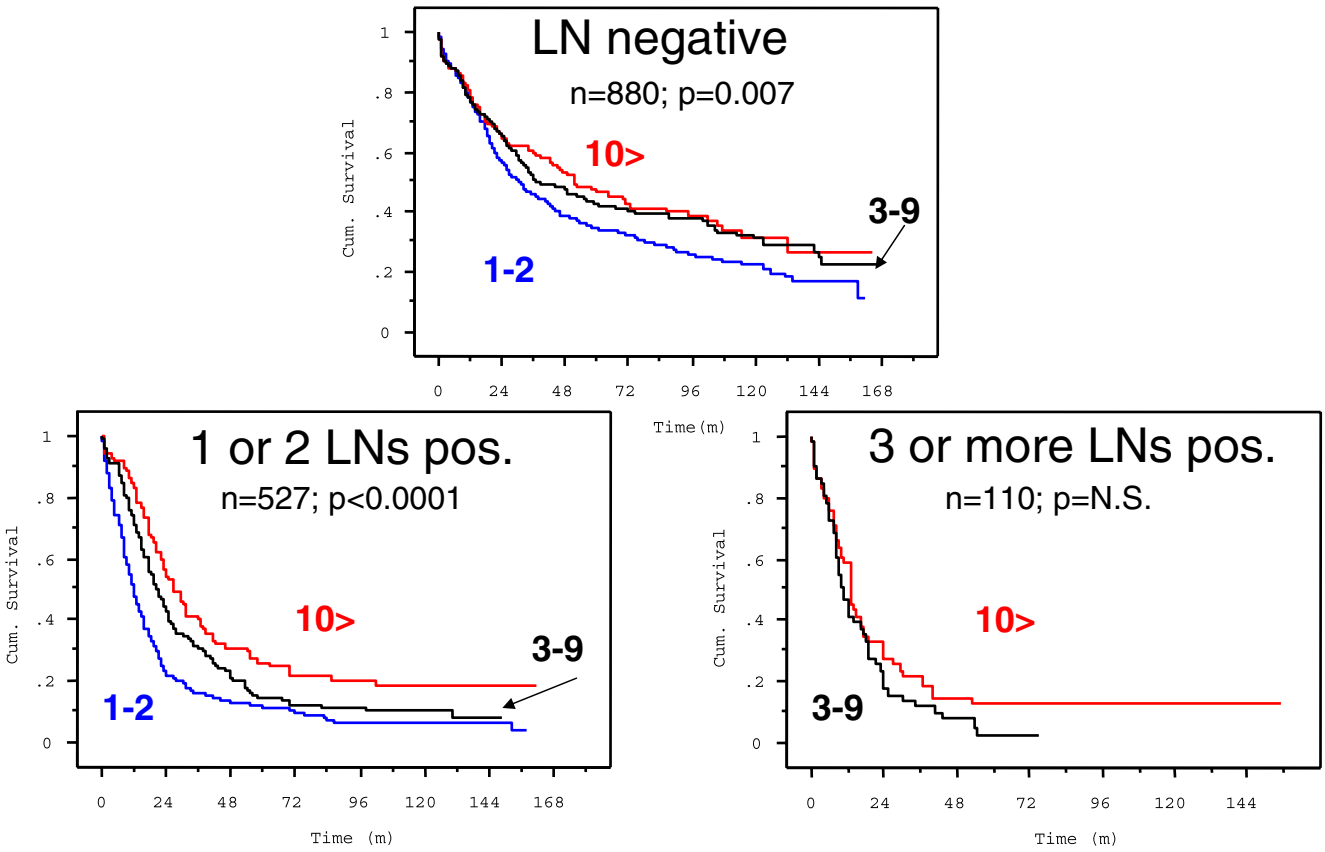

Figure 6 Actuarial OS, by pathological nodal status and total LN counts (all disease sites). Results are displayed separately for N0 patients, N+ patients with one or two LNs involved, and N+ patients with three or more LNs involved.

How to predict which individual belongs to this subset and who may benefit from more extensive LN dissection remains unclear.

No randomized trials exist which would address the question of LN dissection-induced survival benefit in gallbladder or extrapancreatic malignancy. Periampullary CCs are included in a prospective randomized trial that failed to show a survival benefit to extended pancreatoduodenectomy compared to a "standard resection"[10]. The current study based on U.S. population data as captured through the SEER registry is able to link higher total and negative LN counts to longer survival after resection of CCs of various extrahepatic sites. These findings should lend strong credence to an increased awareness of higher LN count benefits, both by the surgeon deciding upon the dissection extent and by the pathologist performing the macroscopic specimen analysis. While the observed effects cannot be extended into a definitive practice recommendation, implications and possible mechanisms of this benefit deserve further discussion.

We have previously reported on a comparable survival impact of LN counts after resections of gastric and pancreatic cancers, although the magnitude appeared to vary somewhat with disease type and stage [11,12]. A similar effect has been described for N0 colorectal cancer, also

Table 4 Actuarial OS by Number of Negative LNs Obtained

	Negative LNs obtained (n)					
	0	1	2	3 to 4	5 to 9	10+
5-year OS (%)	9	30	33	32	33	38
Median OS (months)	11	26	27	28	32	36

All sites combined

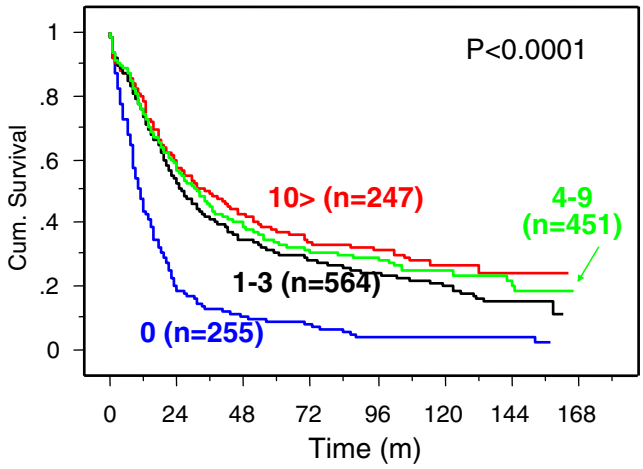

Figure 7 Actuarial OS, by negative LN counts (all disease sites).

Table 5 Median Actuarial OS by Number of Negative LNs Obtained, by Disease Site and Nodal Status

	Negative LNs Obtained (*n*)					
	0	1	2	3 to 4	5 to 9	10+
All sites combined, N0	–	27	38	37	39	51
All sites combined, N1	11	20	17	17	18	23
Gallbladder, N0	–	27	32	11	25	51
Gallbladder, N1	10	19	12	33	39	40
Bile duct, N0	–	20	32	30	24	34
Bile duct, N1	10	13	19	12	16	19
Ampulla, N0	–	50	58	56	51	86
Ampulla, N1	19	28	17	19	21	25

Median survival time in months

Table 6 Projected Numeric Total or Negative LN Impact on Five-year OS

Analysis	Baseline OS at 5 years (based on one LN) (%)	For every additional LN, OS improved by (%)	*p* Value
Total LN counts	21	1.3	0.0742
Negative LN counts	24	1.1	0.2321

All sites combined. Improved survival rate per additional LN examined is based on the linearity model displayed in Fig. 6 (for total LN counts). For example, if a patient had only one LN examined, his expected 5-year OS would be 21%. If a patient had 11 total LNs examined, his expected 5-year OS would be 21%+10×1.3=34%.

based on SEER data[21]. Caution is advised, however, when these findings contrast with the results from prospective randomized trials, which have not resulted in a clear superior survival after extended LN dissections for gastric[22,23] or pancreatic cancer[10,24]. In the gastric cancer trials, there appear to be some confounding hazards, such as increased risks for splenectomy and pancreatectomy-related postoperative sepsis and death. It has to be pointed out that in all reports quoted, the "standard" group to which "extended" dissection was compared had higher LN counts than the highest quartile of patients in the SEER data, comparable to our findings for gastric and pancreatic cancer SEER data[11,12]. It is therefore possible that the LN count–survival relationship observed for CCs, but also for gastric and pancreatic cancers, reflect greater survival hazards due to unacceptably low LN counts removed, and survival benefits observed with higher counts are comparable to those achieved through "standard" or "limited" dissection extents in the randomized trials. If this "standard" extent represents the optimal regional control, any further LN removal would then fail to provide an obvious survival advantage, and even risk increased morbidity as observed in the pancreatectomy trial[25]. Median LN counts in the Hopkins pancreatectomy trial were 16 in the standard vs 26 in the extended group[10], and significantly exceeded those obtained from our data set. As the SEER data reflect U.S. population practice standards, many low-volume providers and institutions are represented. It is quite plausible to assume that some of the observed effect could represent volume characteristics favoring those already represented in the "standard" comparison group.

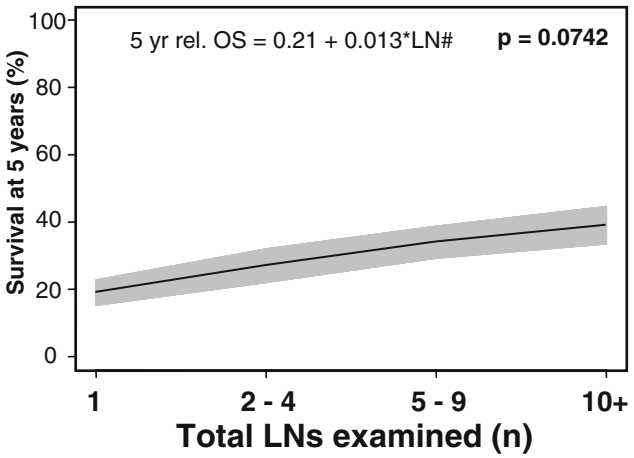

Figure 8 Plots of actuarial OS at 5 years vs the total LN counts. The *shaded area* represents the 95% confidence intervals. The statistically modeled baseline survival at five years with one LN examined is 21%; for every additional LN examined, the projected survival increases by 1.3%.

Other mechanisms may contribute to the observed effect, but cannot be proven. Many of these have been discussed in detail before[11,12]. Is the superior survival with greater LN counts a reflection of higher standards of clinical practice (including the quality of surgical pathology examinations), or a more rigorous selection of healthier patients, aside from the volume considerations mentioned earlier? Could the LN number possibly be a surrogate for other clinical parameters, such as weight loss, comorbid conditions, or a more active immune system with implications for cancer recurrence risks? Some of these factors are known survival predictors, and nearly all are missing in the SEER data, such as margin status, patients' performance status, or comorbidity. It is possible that these are confounding variables with unbalanced distribution among groups of various LN counts.

Last but not least, stage migration is undoubtedly contributing to the observed effect in this study, and may account for a larger portion of the numeric LN survival impact in patients with N0 disease. It is evident that proper stage assignment depends on a critical number of LNs analyzed, and that N+ categories are more likely obtained the more LNs have been analyzed. This process is especially evident for bile duct and ampullary cancers (see Fig. 3), and may perhaps be less obvious in gallbladder cancer, where the majority of individuals had only one LN examined, because the cystic node can be considered the single LN at greatest risk for metastatic involvement for most cancers

arising in the gallbladder. The likely reason for this phenomenon rests with the predominance of laparoscopic resection techniques for gallbladder diseases, including cancers not preoperatively identified[2]. However, it is more difficult to explain the effect seen in N+ patients with mere stage migration. Interestingly, the best survival is consistently observed in groups with 10 or more total LNs analyzed, whereas the biggest increase appears to be observed when at least one negative LN has been identified. Despite a small additional impact of more negative LNs, best survival for nearly all disease sites and stages has been observed for 10 or more negative LNs as well. It appears sensible to consider at least 10 total LNs, with an increased likelihood of at least one negative LN, a minimal goal or minimum acceptable practice standard for the disease sites studied. Obvious detriments to the removal of 10 or more LNs were not encountered, at least as measurable via early postoperative mortality outcomes. We thus conclude that survival prediction of extrahepatic biliary cancer is strongly influenced by total LN counts and numbers of negative LNs obtained. Although the resulting incremental benefit is small and an element of stage migration has to be assumed, benefits are stage-independent. Based on these data, it appears sensible to consider dissection and examination of 10 or more LNs for curative-intent CC resections.

References

1. American Cancer Society. Cancer facts & figures. Atlanta: American Cancer Society 2006.
2. Donohue JH, Stewart AK, Menck HR. The National Cancer Data Base report on carcinoma of the gallbladder, 1989–1995. Cancer 1998;83(12):2618–2628.
3. Yalcin S. Diagnosis and management of cholangiocarcinomas: A comprehensive review. Hepatogastroenterology 2004;51(55):43–50.
4. Jang JY, Kim SW, Park DJ, et al. Actual long-term outcome of extrahepatic bile duct cancer after surgical resection. Ann Surg 2005;241(1):77–84.
5. Kitagawa Y, Nagino M, Kamiya J, et al. Lymph node metastasis from hilar cholangiocarcinoma: Audit of 110 patients who underwent regional and paraaortic node dissection. Ann Surg 2001;233(3):385–392.
6. Kondo S, Nimura Y, Hayakawa N, Kamiya J, Nagino M, Uesaka K. Regional and para-aortic lymphadenectomy in radical surgery for advanced gallbladder carcinoma. Br J Surg 2000;87(4):418–422.
7. Onoyama H, Ajiki T, Takada M, Urakawa T, Saitoh Y. Does radical resection improve the survival in patients with carcinoma of the gallbladder who are 75 years old and older? World J Surg 2002;26(11):1315–1318.
8. Yoshida T, Matsumoto T, Sasaki A, Morii Y, Aramaki M, Kitano S. Prognostic factors after pancreatoduodenectomy with extended lymphadenectomy for distal bile duct cancer. Arch Surg 2002;137(1):69–73.
9. Araida T, Yoshikawa T, Azuma T, Ota T, Takasaki K, Hanyu F. Indications for pancreatoduodenectomy in patients undergoing lymphadenectomy for advanced gallbladder carcinoma. J Hepatobiliary Pancreat Surg 2004;11(1):45–49.
10. Yeo CJ, Cameron JL, Lillemoe KD, et al. Pancreaticoduodenectomy with or without distal gastrectomy and extended retroperitoneal lymphadenectomy for periampullary adenocarcinoma, part 2: Randomized controlled trial evaluating survival, morbidity, and mortality. Ann Surg 2002;236(3):355–366; discussion 366–368.
11. Smith DD, Schwarz RR, Schwarz RE. Impact of total lymph node count on staging and survival after gastrectomy for gastric cancer: Data from a large US-population database. J Clin Oncol 2005;23(28):7114–7124.
12. Schwarz RE, Smith, DD. Extent of lymph node retrieval and pancreatic cancer survival: Information from a large U.S. population database. Ann Surg Oncol 2006;13(9):1189–1200.
13. Surveillance, epidemiology and end results (SEER) program: Public use data (1973–2004). Retrieved April 2005, from http://www.seer.cancer.gov.
14. Greene FL, Page DL, Fleming ID, et al. American joint committee on cancer staging manual (6th ed.). New York: Springer, 2002.
15. Cox DR. Regression models and life tables. J R Stat Assoc 1972;29:187–220.
16. Kaplan EL, Meier P. Nonparametric estimation from incomplete observations. Am Stat Assoc 1958;53:457–481.
17. Peto R, Pike MC. Conservatism in the approximation E(0-E)2/E in the log rank test for survival data or tumor incidence data. Biometrics 1973;29:579–584.
18. Pawlik TM, Abdalla EK, Barnett CC, et al. Feasibility of a randomized trial of extended lymphadenectomy for pancreatic cancer. Arch Surg 2005;140(6):584–589; discussion 9–91.
19. Schwarz RE, Zagala-Nevarez K. Recurrence patterns after radical gastrectomy for gastric cancer: Prognostic factors and implications for postoperative adjuvant therapy. Ann Surg Oncol 2002;9(4):394–400.
20. Gervasoni JE, Jr., Taneja C, Chung MA, Cady B. Biologic and clinical significance of lymphadenectomy. Surg Clin North Am 2000;80(6):1631–1673.
21. Cserni G, Vinh-Hung V, Burzykowski T. Is there a minimum number of lymph nodes that should be histologically assessed for a reliable nodal staging of T3N0M0 colorectal carcinomas? J Surg Oncol 2002;81(2):63–69.
22. Cuschieri A, Weeden S, Fielding J, et al. Patient survival after D1 and D2 resections for gastric cancer: Long-term results of the MRC randomized surgical trial. Surgical Co-operative Group. Br J Cancer 1999;79(9–10):1522–1530.
23. Hartgrink HH, Van De Velde CJ, Putter H, et al. Extended lymph node dissection for gastric cancer: Who may benefit? Final results of the randomized Dutch gastric cancer group trial. J Clin Oncol 2004;22(11):2069–2077.
24. Pedrazzoli S, DiCarlo V, Dionigi R, et al. Standard versus extended lymphadenectomy associated with pancreatoduodenectomy in the surgical treatment of adenocarcinoma of the head of the pancreas: A multicenter, prospective, randomized study. Lymphadenectomy Study Group. Ann Surg 1998;228(4):508–517.
25. Yeo CJ, Cameron JL, Sohn TA, et al. Pancreaticoduodenectomy with or without extended retroperitoneal lymphadenectomy for periampullary adenocarcinoma: Comparison of morbidity and mortality and short-term outcome. Ann Surg 1999;229(5):613–622; discussion 622–624.

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Percutaneous Transhepatic Cholangiodrainage as Rescue Therapy for Symptomatic Biliary Leakage Without Biliary Tract Dilation After Major Surgery

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Abstract Symptomatic biliary leakage following major upper abdominal surgery is a severe complication resulting in increased morbidity and mortality. Treatment options usually include either endoscopic intervention or surgical revision. These options may be burdened by a high perioperative risk for the patient (e.g., patients with severe disease) or simply may not be possible (e.g., nonpreserved gastroduodenal passage). In the past, percutaneous transhepatic cholangiodrainage did only seem to be a viable option for patients with dilated bile ducts. Here, we present our experience in a consecutive series of patients with symptomatic biliary leakage following major upper abdominal surgery and without dilation of the biliary system that underwent percutaneous transhepatic cholangiodrainage. Percutaneous transhepatic cholangiodrainage was feasible in 15 of 18 patients (83.3%). The procedure was technically not possible in three patients (16.7%). In 10 of the 15 patients (66.6%) with feasible percutaneous transhepatic cholangiodrainage, biliary leakage was definitely controlled without the need for surgical revision. Depending on the experience with the interventional procedure, percutaneous transhepatic cholangiodrainage should be considered as an alternative for treatment of symptomatic biliary leakage instead of immediate reoperation.

Keywords Interventional radiography · Biliary leakage · Postoperative complication · Upper abdominal surgery

Introduction

Symptomatic biliary leakage following upper abdominal surgery, e.g., liver or pancreas surgery represents a clinical challenge. Incidence strongly varies between 0.8 and 38%[1,2] and is associated with increased morbidity, mortality, and length of hospital stay.[3–5] The main reasons for leakage are the insufficiency of the cystic duct stump, of the biliodigestive anastomosis, or of the pancreato-jejunostomy, or the existence of accessory bile ducts.[6] Frequently, patients presenting with the clinical feature of symptomatic bile leak are in impaired general condition. The value of preoperative biliary decompression to avoid these problems is discussed controversially.[7–11] Once biliary leakage occurs, treatment concepts consist in draining the bile to support spontaneous closure of the leak by reducing or evading biliary leakage. In cases with preserved gastroduodenal passage, endoscopic biliary drainage is regarded as the first viable option.[12,13]

The impetus of endoluminal biliary stenting for the treatment of biliary fistula is to reduce the pressure in the biliary system (even if this is low), and hereby increase the chance for spontaneous closure of the insufficiency. If endoscopy is not applicable, e.g., after Whipple's operation, interventional procedures including computertomographic- or ultrasonographic-guided percutaneous biliary drainage, aspiration, or percutaneous transhepatic cholangiodrainage (PTCD) may

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Figure 1 Transhepatic puncture of the right major, intrahepatic bile duct in a patient with choledocho-jejunostomy.

be considered.[6,14–17] Special attention has to be addressed to a greater intra-abdominal biliary collection that has to be drained either by percutaneous puncturing or surgically, irrespective of endoluminal stenting. However, as ultima ratio, the option of urgent relaparotomy remains.

Recent reports have shown that, due to advances of radiologic drainage techniques, PTCD has become an attractive and less invasive alternative to reoperation. Although PTCD generally has been reserved for dilated ducts, in experienced hands, transhepatic access to the biliary system is even possible in cases of nondilated bile ducts.[18]

This study summarizes our institutional experience with a consecutive series of patients with symptomatic intra-abdominal bile leakage following upper abdominal surgery and without dilation of the biliary system that underwent PTCD. We evaluated whether PTCD in these patients is a reasonable alternative to reoperation.

Methods

Patients

All patients receiving PTCD between August 2000 and July 2004 for symptomatic biliary leakage following surgery of the upper abdomen in the University Medical Center of Hamburg-Eppendorf were included in this retrospective study. Patients with dilation of the biliary system were excluded. Dilation of the biliary system was defined as when the diameter of biliary ducts of primary order were greater or equal to 3 mm and no evidence of bile ducts within 20 mm from the hepatic capsule was provided sonographically. Biliary leakage was defined as when bilirubin in the abdominal drain doubled the value in serum. Patients were judged to suffer from clinical symptoms originating from a bile leak when at least one of the following symptoms was recorded and an extra-abdominal cause for the onset of these symptoms was excluded: persistent nausea and vomiting, severe abdominal pain, fever, and tachycardia. An intra-abdominal, biliary fluid collection, which would have required either the placement of an additional percutaneous drainage or, in case of an inaccessible fluid collection, relaparotomy, was ruled out prior to PTCD.

Interventional Technique

PTCD for the management of biliary leakage in patients with nondilated biliary tract was carried out as follows. Access to the central biliary tree was obtained via right-sided intercostals percutaneous puncture with local anesthesia. Under fluoroscopic guidance, a first-order paracentral bile duct was punctured with a 21-gauge initial needle and its position was proven with the injection of a small volume of contrast fluid (Fig. 1). A 0.014-in guidewire was inserted through the needle into the bile duct, and a small 18-gauge Teflon cannula (bicoaxial system, Cook, Mönchengladbach, Germany) was passed over the guidewire. Both the initial 21G needle and guidewire were replaced by a 0.035-in hydrophilic guidewire (Terumo, Somerset, NJ, USA) and the Teflon was replaced by a 5F glidecatheter (Terumo). The guidewire was advanced past the spot of leakage (e.g., anastomosis) and then exchanged for a stiff guidewire (Amplatz stiff guide, Cook). Over this wire, a 6.1 to 8.5F polyethylene drainage catheter was placed into the bile duct just proximal of the leakage (Fig. 2). The bile was drained externally, leaving the leakage to subside (e.g., healing of anastomosis) (Fig. 3). A fluoroscopic check was performed at the following day and, if necessary, adjustment of the drainage's position was achieved (Fig. 4).

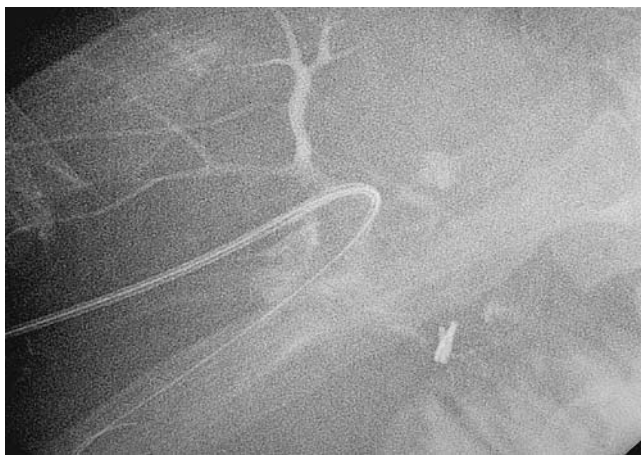

Figure 2 Successful intubation of the insufficient choledocho-jejunal anastomosis in the same patient (see Fig. 1).

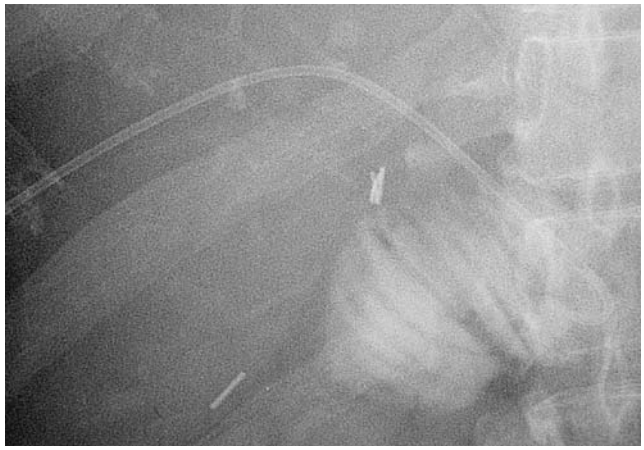

Figure 3 Upper abdominal x-ray showing the PTD with its tip correctly positioned in the Roux-Y-loop of the choledochojejunostomy.

The catheter was pinched off when bilirubin in the abdominal drain decreased significantly and when patients were symptom-free for at least 48 h. When patients did not exhibit recurrence of the bile leak, the catheter was removed and the former drainage tract was embolized to prevent prolonged percutaneous bile leakage using a mixture of n-buthy-cyanoacrylate and lipiodol-ultra fluid (Guerbet, Villepinte, France).

Data Analysis

All computations were done with the Statistical Package for Social Sciences (SPSS), version 11.5 (SPSS, Chicago, IL, USA). Data are reported as mean ± standard deviation in text and tables. Significance of differences between groups was assessed by the Mann–Whitney U test.

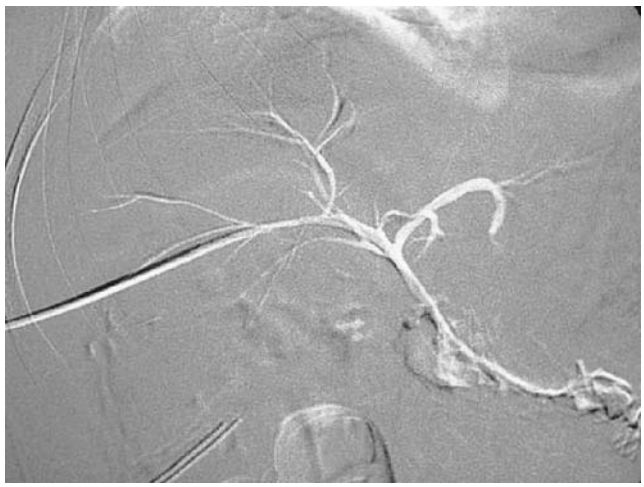

Figure 4 Control fluoroscopy showing adequate drainage of the nondilated biliary tree via PTCD.

Table 1 Operations that Led to Biliary Leakage

Surgical procedure	No. of patients	%
Duodenum-preserving pancreatic head resection	7	38.9
Duodenopancreatectomy with or without pylorus preservation	6	33.3
Pancreas preserving duodenectomy with partial gastric resection	2	11.1
Transhiatal esophagectomy	1	5.5
Total gastrectomy	1	5.5
Subtotal splenopancreatectomy and necrosectomy with partial resection of duodenum and jejunum	1	5.5

Results

A consecutive series of 18 patients (age 35–67) with symptomatic biliary leakage following surgery of the upper abdomen underwent PTCD without dilation of the biliary system. Diagnoses leading to hospital admission and surgery were chronic pancreatitis in seven patients (38.9%); periampullary cancer in five patients (27.8%); ulcer perforation in two patients (11.1%); and esophageal and gastric cancer, choledocholithiasis, and acute hemorrhagic necrotizing pancreatitis in one patient, each (5.5%). None of the patients received placement of a stent prior to surgery. In most of the patients, major pancreatic surgery had been performed (Table 1). The sites of leakage were the cystic duct stump, the pancreaticojejunostomy (each $n=3$ or 16.6%), the site of T-drain insertion, the duodenal stump, the biliodigestive anastomosis (each $n=2$ or 11.1%), the duodenojejunostomy, and the proximal common hepatic duct (each $n=1$ or 5.5%). In four patients (22.2%), the exact site of leakage could not be visualized. Symptoms associated with biliary leakage are summarized in Table 2. PTCD was technically feasible in 15 of 18 patients (83.3%) without complications. The remaining three patients in whom PTCD could not be performed underwent relaparotomy. In 10 of the 15 patients (66.6%) with feasible PTCD, biliary leakage was definitely controlled, whereas in five patients, it failed to definitely control leakage. This resulted in a total of eight patients who eventually had to be reoperated. Three out of these eight patients needing

Table 2 Frequencies of Symptoms of Biliary Leakage

Symptom	*n*	Frequency (%)
Severe abdominal pain	16	88.8
Fever	16	88.8
Persistent nausea and vomiting	11	61.1
Tachycardia	7	38.8

relaparotomy died perioperatively (37.5%). One patient among the 10 that underwent sufficient PTCD died in hospital (10%). Median days with PTC drainage were 27±35.6 (range 3–147). Overall, median postoperative days in hospital were 53±28.4 (range 21–113). Patients with an adequate PTC drainage had a median of 45±24.6 postoperative hospital days, whereas the median postoperative days in hospital in patients needing relaparotomy was 54±33.3 (p=0.194).

Discussion

When endoscopic stenting cannot be performed and biliary dilation is absent, symptomatic biliary leakage following major upper abdominal surgery is usually considered a domain of surgery. Consensus exists that relaparotomy is indicated in case of severe biliary sepsis. Further indications for surgery are intra-abdominal bile collections that can anatomically not be accessed by the placement of an additional percutaneous drain. The present series including patients who had neither biliary sepsis nor had intra-abdominal biliary fluid collections shows that PTCD is a reliable treatment option with reasonably low procedure-related morbidity even in the absence of biliary tract dilation. The key message of our study is that two-thirds of those patients (10/15) in whom PTCD was feasible despite nondilated intrahepatic bile ducts were eventually protected from relaparotomy. Overall, PTCD prevented 10 of 18 patients from further surgery (55%). These findings are consistent with a recently published study of de Castro et al.[18] who found PTCD to be safe without relevant morbidity and reported a failure rate of only 10% (in our study, 16.6%). Two of 10 patients (20%) required relaparotomy for persistent biliary leakage (in our study, 33.3%) despite preceding PTCD. However, de Castro was focusing on biliary leakage following hepaticojejunostomy and unfortunately did not clarify the number of patients without dilation of the biliary tree in the PTCD group, nor did he comment on the issue of perioperative mortality associated with the chosen treatment of leakage.

In our study, out of the eight patients who required reoperation, four had to be surgically treated more than once with peritoneal lavage due to severe peritonitis. In this group, three patients died perioperatively (37.5%), whereas one patient died in the group of sufficient PTCD (10%). Still, the number of patients included in the study is too small to enable making general statements in terms of a risk–benefit analysis. On the other hand, besides its potential of preventing reoperation in a subset of patients with clearly impaired medical condition, PTCD shows tendencies to improve prognosis in these patients.

In the future, prospective trials have to show whether these tendencies prove to be valid. These studies clearly have to compare relaparotomy and PTCD in patients who do not suffer from severe biliary sepsis with attending peritonitis. Future studies may as well answer the question of whether PTCD is a reasonable alternative to endoscopic biliary drainage in patients with preserved gastroduodenal passage.

Conclusion

If endoscopic drainage is not possible for patients with symptomatic biliary leakage after major upper gastrointestinal surgery, PTCD should be considered as rescue therapy instead of immediate reoperation, depending on the patient's clinical status and the experience with the interventional procedure. This also holds true in patients without dilated intrahepatic bile ducts.

References

1. Hopt UT, Makowiec F, Adam U. Leakage after biliary and pancreatic surgery. Chirurg 2004;75:1079–1087.
2. Adam U, Makowiec F, Riediger H, et al. Risk factors for complications after pancreatic head resection. Am J Surg 2004; 187:201–208.
3. Yeo CJ, Cameron JL, Sohn TA, et al. Six hundred fifty consecutive pancreaticoduodenectomies in the 1990s: pathology, complications, and outcomes. Ann Surg 1997;226:248–257; discussion 257–260.
4. Rios G, Conrad A, Cole D, et al. Trends in indications and outcomes in the Whipple procedure over a 40-year period. Am Surg 1999;65:889–893.
5. Ghahremani GG. Postsurgical biliary tract complications. Gastroenterologist 1997;5:46–57.
6. Sohn TA, Yeo CJ, Cameron JL, et al. Pancreaticoduodenectomy: role of interventional radiologists in managing patients and complications. J Gastrointest Surg 2003;7:209–219.
7. Nakeeb A, Pitt HA. The role of preoperative biliary decompression in obstructive jaundice. Hepatogastroenterology 1995;42:332–337.
8. Pitt HA, Gomes AS, Lois JF, et al. Does preoperative percutaneous biliary drainage reduce operative risk or increase hospital cost? Ann Surg 1985;201:545–553.
9. Povoski SP, Karpeh MS Jr., Conlon K C, et al. Association of preoperative biliary drainage with postoperative outcome following pancreaticoduodenectomy. Ann Surg 1999;230:131–142.
10. Pisters PW, Hudec WA, Hess KR, et al. Effect of preoperative biliary decompression on pancreaticoduodenectomy-associated morbidity in 300 consecutive patients. Ann Surg 2001;234:47–55.
11. Sohn TA, Yeo CJ, Cameron JL, et al. Do preoperative biliary stents increase postpancreaticoduodenectomy complications? J Gastrointest Surg 2000;4:258–267; discussion 67–68.
12. Bose SM, Mazumdar A, Singh V. The role of endoscopic procedures in the management of postcholecystectomy and posttraumatic biliary leak. Surg Today 2001;31:45–50.

13. Kromer MU, Maier M, Benz CA, et al. Bile duct stenoses and leakage after cholecystectomy: endoscopic diagnosis, therapy and treatment outcome. Z Gastroenterol 1996;34:167–172.
14. Dawson SL, Mueller PR. Interventional radiology in the management of bile duct injuries. Surg Clin N Am 1994;74:865–874; discussion 75–81.
15. Vaccaro JP, Dorfman GS, Lambiase RE. Treatment of biliary leaks and fistulae by simultaneous percutaneous drainage and diversion. Cardiovasc Intervent Radiol 1991; 14:109–112.
16. Chen XP, Peng SY, Peng CH, et al. A ten-year study on non-surgical treatment of postoperative bile leakage. World J Gastroenterol 2002;8:937–942.
17. Suehiro M, Ishimura J, Fukuchi M. Detection of bile leakage into the thoracic cavity by hepatobiliary scintigraphy. Ann Nucl Med 1991;5:167–169.
18. de Castro SM, Kuhlmann KF, Busch OR, et al. Incidence and management of biliary leakage after hepaticojejunostomy. J Gastrointest Surg 2005;9:1163–1171, discussion, 71–73.

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Suramin Inhibits Not Only Tumor Growth and Metastasis but Also Angiogenesis in Experimental Pancreatic Cancer

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Abstract Suramin inhibits the proliferation of several human tumors in vivo and in vitro. In this study, the effects of Suramin on proliferation and angiogenesis were investigated in human pancreatic cancer cell lines and in an orthotopic nude mouse model of human pancreatic cancer. The effects of Suramin on proliferation, viability, cell cycle, and apoptosis were studied in five human pancreatic cancer cell lines. Suramin inhibited the proliferation of pancreatic cancer cells in a dose-dependent manner and reduced viability at high concentrations. Cell cycle analysis revealed a decreased S-phase fraction in most cell lines, whereas the apoptotic fraction was not notably different. In vivo treatment with Suramin significantly reduced pancreatic tumor size (MiaPaCa-2, −74%; AsPC-1, −41%; and Capan-1, −49%) and metastatic spread (MiaPaCa-2, −79%; AsPC-1, −34%; and Capan, −38%). As a parameter for angiogenic activity, vascular endothelial growth factor (VEGF) secretion was measured, revealing reduced VEGF concentrations under Suramin treatment in both cell culture medium and ascites. Also, microvessel density quantified in primary tumors was reduced in animals treated with Suramin. Therefore, Suramin inhibits the proliferation of human pancreatic cancer in vitro and in vivo. The therapeutic effects seem to involve cell cycle kinetics and may be in part related to the antiangiogenic action of the drug.

Keywords Suramin · Pancreatic cancer · Angiogenesis · Orthotopic mouse model · Microvessel density

Introduction

Adenocarcinoma of the pancreas is the fifth leading cause of cancer-related death in western countries. Despite numerous advances in the treatment of this disease, the prognosis of patients diagnosed with pancreatic cancer remains dismal. More than 80% of the patients have inoperable advanced-stage disease at diagnosis. The poor overall 5-year survival rate of <5%[1] is due to the tumor's propensity toward aggressive tumor growth, early metastasis, and resistance to cytotoxic agents and radiation. Therefore, novel therapeutic strategies are required to improve the prognosis of patients with pancreatic cancer.

The pathological development of vascular supply is a critical step for tumor growth and appears to impact prognosis. Pharmacological control of angiogenesis represents an alternative approach to the management of solid malignancies. The complex process of angiogenesis is mediated in part by the secretion of several growth factors, including vascular endothelial growth factor (VEGF). Antiangiogenic agents decrease tumor growth and metastatic dissemination of numerous solid tumor types. VEGF antagonists are currently being used clinically and several are in development[2,3].

One potential antiangiogenic agent is Suramin. This polysulfonated naphtyl urea derivative was originally developed to treat trypanosomiasis and onchocerciasis and has been used since the 1920s[4]. More recently, Suramin has been found to have antineoplastic effects. The exact

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mechanism of Suramin's antiangiogenic activity is not known, but nonspecifical interactions with numerous growth factors and angiogenic factors, like basic fibroblast growth factor (bFGF), platelet-derived growth factor, transforming growth factor, and VEGF, have been shown[5]. In addition, Suramin interferes with cellular enzymes such as DNA polymerase, topoisomerase II, and protein kinase C. Suramin has been reported to inhibit tumor growth and has been tested in clinical trials, e.g., for the therapy of prostate cancer[6]. The antiangiogenic effect of Suramin has been analyzed in vivo and documented in the chick chorioallantoic membrane assay and in a bFGF-induced model with gel sponges subcutaneously implanted in mice[7,8].

The effect of Suramin in pancreatic cancer has yet to be elucidated. To improve the existing knowledge on the therapeutic activity of Suramin and to characterize in more detail its antiangiogenic potential in human pancreatic cancer, we tested this drug in vitro by evaluating the effects on proliferation, cell viability, and VEGF secretion of human pancreatic cancer cells. Furthermore, we studied the therapeutic and antiangiogenic potential of Suramin in a clinically relevant orthotopic nude mouse model of human pancreatic cancer. To rate the antiangiogenic activity of Suramin, we performed additional in vivo experiments with the angiogenesis inhibitor TNP-470.

Materials and Methods

Drugs

For in vitro assays and intraperitoneal injection, Suramin (Bayer AG, Leverkusen, Germany) was dissolved in 0.9% NaCl (pH 7.5). TNP-470[9] was provided by TAP Pharmaceuticals (Deerfield, IL, USA). For in vivo experiments, TNP-470 was suspended in a vehicle composed of 0.5% ethanol and 5% gum Arabic in physiologic saline solution.

Cell Lines and Culture Conditions

The following human pancreatic adenocarcinoma cell lines were obtained from the American Type Culture Collection (Rockville, MD, USA): Capan-1 (C1) (moderately to well differentiated), Capan-2 (well differentiated), MiaPaCa-2 (MP) (undifferentiated), AsPC-1 (AP) (poorly to moderately differentiated), and PANC-1 (poorly differentiated). MP cells and PANC-1 cells were cultured in Dulbecco's modified Eagle's medium (Gibco, Grand Island, NY, USA), C1 and AP cells in RPMI-1640 medium (Gibco), and Capan-2 in McCoy's 5A medium. All media were supplemented with 10% heat-inactivated fetal bovine serum (Gibco), penicillin G (100 U/ml), streptomycin (100 μg/ml), and 0.1% fungizone. The cells were incubated at 37°C in humidified air with 5% CO_2. The medium was replaced twice weekly, and cells were maintained by serial passaging after trypsinization with 0.1% trypsin.

In Vitro Assessment of Cell Proliferation and Viability

To examine the effect of Suramin on in vitro cell proliferation, 2×10^5 cells from each cell line were seeded in six-well culture plates in 2 ml of the respective cell culture medium. The medium was changed the next day (day 1) and Suramin was added in the following concentrations: 10, 100, 200, and 800 μg/ml. After 72 h (day 4), the cells were trypsinized and counted in a standard hemocytometer. Cell viability was assessed by a colorimetric dye reduction assay with monotetrazolium (MTT) (Boehringer, Mannheim, Germany) according to the manufacturer's instructions. Briefly, cells were seeded in 96-well plates at a density of 5×10^3 cells in 0.2 ml of the respective medium. Medium was changed the next day (day 1) and Suramin was added as described above. After 72 h (day 4), 10 μl of MTT (5 mg/ml) solution and, after and additional 4 h, 100 μl of 10% sodium dodecyl sulfate were added. The plates were allowed to stand overnight (37°C, 5% CO_2). The change in absorbance measured at 570 nm with an ELISA reader (Biotek Instruments, Burlington, VT, USA) has been shown to strongly correlate with the number of viable cells.

Flow Cytometric Analysis of Cell Cycle Distribution and of Induction of Apoptosis

As an early event of apoptosis, phosphatidylserine translocates from the inner cytoplasmic to the outer membrane, which can be determined using Annexin-V-FLUOS (Boehringer). The cells react to Annexin V as soon as chromatin condenses but before the plasma membrane loses its ability to exclude propidium iodide. Hence, it is possible to detect nonapoptotic living cells, early apoptotic cells, and late apoptotic or necrotic cells by staining them with a combination of fluoresceinated Annexin V and propidium iodide. First, 10^6 cells (C1, Capan-2, MP, and AP) were seeded in six-well culture plates in 2 ml of the respective cell culture medium. The medium was changed the next day (day 1) and Suramin was added in different concentrations as described above. After 72 h (day 4), the cells were trypsinized, washed with PBS, and centrifuged at 1,300 rpm for 5 min. The pellets were resuspended in 100 μl staining solution (10 μl Annexin-V-FLUOS, 10 μl prodidium iodide, 500 μl 4-2-hydroxyethyl-1-piperazine-ethanesulfonic acid buffer). After incubation at room temperature for 15 min in the dark, the samples were analyzed by flow cytometry using 488 nm for excitation and 515 nm for detection.

Laboratory Animals and Orthotopic Implantation Technique

Four-week-old male nude mice (Crl:NU/NU-*nu*BR) weighing 20–22 g were obtained from Charles River Laboratories (Wilmington, MA, USA). The animals were housed in microisolator cages with autoclaved bedding, food, and water. The mice were maintained on a daily 12-h light/12-h dark cycle. All experiments were conducted in accordance with the national guidelines for the care and use of laboratory animals, and the experimental protocol was approved by the Chancellor's Animal Research Committee of the University of California, Los Angeles.

The orthotopic pancreatic tumor implantation technique was previously described in detail[10]. First, 5×10^6 cells of each human pancreatic cancer cell line were injected subcutaneously into the flanks of donor nude mice. The animals were killed after 3 to 4 weeks when the subcutaneous tumors had reached a size of 1 cm in the largest diameter. The donor tumors were harvested and minced by a scalpel (no. 11) into fragments of 1 mm^3 in size. The abdomens of anesthetized tumor recipient nude mice were opened by a midline incision under aseptic conditions at a laminar air flow working bench, and the pancreatic tail with the spleen was gently exteriorized. Two small tissue pockets were prepared in the pancreatic parenchyma as an implantation bed with a microscissor (RS-5610 VANNAS; Roboz, Rockville, MD, USA). One donor tumor fragment was placed into each pancreatic tissue pocket so that the tumor tissue was completely surrounded by pancreatic parenchyma. The pancreas was relocated into the abdominal cavity, which was then closed in two layers with 5-0 absorbable sutures (DEXON "S"; Davis+Geck, Manati, Puerto Rico).

In Vivo Treatment with Suramin and TNP-470

Tumor fragments of MP, AP, and C1 cells from the donor animals were orthotopically implanted in the mouse pancreas. The animals were randomized into ten groups of 12 animals each: MP-Con (control), MP-S10 (Suramin 10 mg/kg), MP-S60 (Suramin 60 mg/kg)[11], MP-TNP (TNP 30 mg/kg), AP-Con, AP-S60, AP-TNP, C1-Con, C1-S60, and C1-TNP. Treatment with Suramin and TNP-470 started 3 days after orthotopic tumor implantation. Suramin was administered by intraperitoneal injections twice per week in the first 2 weeks and once a week subsequently, whereas TNP-470 was given every other day subcutaneously at a dosage of 30 mg/kg bodyweight. Both therapeutic substances were administered over a period of 14 weeks. The control groups received the carrier substance (0.9% NaCl). The mice were monitored daily for their clinical condition, weighed weekly, and killed by a lethal dose of sodium pentobarbital (0.5 mg/g body weight) 14 weeks after the orthotopic tumor implantation.

All animals underwent an autopsy at the end of the observation period. The perpendicular diameters of the primary orthotopic tumor were measured with calipers, and the volume was calculated using the following formula: volume = length × width × depth/2. A dissemination score was developed to assess local tumor infiltration, as well as distant metastasis[10]. Local infiltration was determined at the following sites: spleen, stomach, liver (hilus), kidney (hilus), retroperitoneum, diaphragm, mesentery, bowel loops, and abdominal wall. Isolated tumor nodules with no anatomical connection to the primary were judged as distant metastases. The sites of evaluation included liver, kidney, spleen, lung, diaphragm, mesentery, retroperitoneum, mediastinum, and the suture line. Tumor dissemination was quantified as follows: every manifestation of tumor infiltration or metastasis was credited with one point. Additional points were awarded for massive local infiltration (e.g., including more than half of the circumference of the spleen), multiple metastatic nodules (>1 in parenchymal organs; >10 on diaphragm, mesentery, retroperitoneum), and metastatic nodules >50 mm^3. Clinical consequences of the tumor growth were incorporated into this scoring system: formation of ascites (2 points if volume >5 ml), development of jaundice, ileus, and cachexia. The autopsy data were analyzed by one individual (HGH) who was blinded to the treatment groups.

The primary tumor and all sites of potential infiltration or metastasis were harvested, fixed in paraformaldehyde, and embedded in paraffin. Five-micron-thin tissue sections were obtained and stained with hematoxylin and eosin for microscopic examination. The sections were reviewed to confirm the findings of the macroscopic dissemination score.

Determination of VEGF Levels in Ascites and Culture Medium

An ascites sample of each animal (if present) was collected at autopsy. Fifty microliters of EDTA (0.2 M, pH 8.0) was added to 500 μl of ascites, and all samples were centrifuged for 5 min at 2,500 rpm and stored at −80°C. In addition, cell culture supernatants of human pancreatic cancer cells (MP as an undifferentiated cell line, C1 as a well differentiated cell line) were collected. The amount of VEGF in ascites and cell culture medium was quantified by using the Quantikine human VEGF immunoassay kit (R&D Systems, Minneapolis, MN, USA) according to the manufacturer's instructions.

Immunohistochemical Analysis of Microvessel Density

Anti-CD31 was used as an endothelial marker to highlight intratumoral microvessels. The human pancreatic cancer xenograft tumors orthotopically grown in the pancreas of

nude mice were immediately fixed in 10% neutral buffered formalin and embedded in paraffin. Tissue sections (3 μm) were deparaffinized and rehydrated and target retrieval was done by cooking tissues at 97°C for 15 min in 0.01% EDTA (pH 8.0), followed by a 5-min treatment in a 3% hydrogen peroxide solution to block endogenous alkaline phosphatase activity. After blocking slides for 5 min, a purified anti-mouse CD 31 (PECAM-1) antibody (Santa Cruz, San Diego, CA, USA) was applied in a 1:100 dilution and was incubated at 37°C for 30 min. After thorough rinsing in PBS-Tween solution, slides were incubated with a biotinylated secondary antibody for 20 min, followed by a 20-min incubation with streptavidin peroxidase. For color development, slides were incubated for 2 min in DAB (3,3′-diaminobenzidine tetrahydrochloride). Microvessel density was quantified as described by Weidner[12]. Areas of highest neovascularization were found by scanning the sections at low power (×40 and ×100 total magnification). Individual microvessel counts were made on ten ×200 fields (0.74 mm^2 per field).

Statistical Analysis

Data are presented as mean ± SEM. Continuous, normally distributed variables were analyzed by the Student *t* test. Discontinuous variables (dissemination score, microvessel density) were analyzed by the Mann–Whitney rank sum test. Differences were considered significant at a p value<0.05.

Results

Effect of Suramin on Proliferation and Cell Viability In Vitro

The effects of Suramin on the proliferation and viability of all five investigated human pancreatic cancer cell lines, C1, Capan-2, Panc-1, MP, and AP, were studied over a time period of 72 h. The chemotherapeutic agent was applied to the cells using four different concentrations: 10, 100, 200, and 800 μg/ml. Table 1 summarizes proliferation and viability changes during drug treatment. Suramin inhibited the proliferation of all investigated human pancreatic cancer cell lines in a dose-dependent manner. The inhibition was most effective with Panc-1 cells. The highest concentration of Suramin reduced cell proliferation to less than 10% in this cell line. Cell viability was reduced at high concentrations of the drug.

Effect of Suramin on Cell Cycle and Apoptosis

For further analysis of the mechanisms of growth inhibition induced by Suramin, we examined the cell cycle and apoptosis profile of C1, Capan-2, MP, and AP cells either in drug medium or in the presence of 0, 10, 100, 200, and 800 μg/ml of Suramin. Annexin and PI staining of cells treated with Suramin were examined by flow cytometry. Suramin induced a decrease of Capan-2, MP, and AP cells in the phase of DNA synthesis (S phase) and an increase in the G0/1 phase, particularly at a high Suramin concentration of 800 μg/ml. In contrast, the percentage of C1 in the S phase increased. The results are summarized in Table 2. The externalization of phosphatidylserine is a common feature of both early and late apoptotic stages. Annexin staining of cells revealed no notable increase of the apoptotic cell fraction in all four cell lines when treated with Suramin, even at higher concentrations (data not shown).

Table 1 Effect of Suramin on Proliferation and Cell Viability of Pancreatic Cancer Cells

Suramin (μg/ml)	C1 (%)	Capan-2 (%)	Panc-1 (%)	MP (%)	AP (%)
Proliferation (control=100)					
10	99.0± 14.0	89.4± 8.5	53.5± 7.5*	90.4± 6.7	91.0± 8.7
100	85.5± 13.5	67.2± 18.3	40.3± 6.2*	81.7± 5.0	60.5± 4.8*
200	85.3± 14.0	39.6± 7.7*	20.8± 5.4*	65.8± 10.0*	52.7± 4.2*
800	58.9± 10.7*	31.9± 10.6*	8.7± 3.3*	46.3± 6.7*	34.4± 2.6*
Viability (control=100)					
10	105.0± 1.9	93.2± 3.8	100.0± 6.0	98.2± 1.8	105.0± 1.9
100	100.6± 3.8	93.9± 3.2	112.0± 8.0	110.7± 3.6	100.6± 3.7
200	95.4± 1.9	85.1± 2.5	98.0± 6.0	101.8± 3.6	95.4± 1.9
800	75.3± 1.9*	77.2± 1.9*	64.0± 2.0*	58.9± 1.8*	75.3± 1.9*

Pancreatic cancer cells were incubated with different concentrations of Suramin for 72 h. Proliferation was detected by cell counting, whereas cell viability was measured via MTT assay. Data were normalized to 100% (100% = control cells without Suramin) and are expressed as mean ± SEM

*p<0.05

Effect of Suramin on VEGF Secretion in Cell Culture Medium and Ascites

MP and C1 cells were incubated with different doses of Suramin (see above), and VEGF levels were evaluated in the cell culture supernatants. The VEGF secretion in human pancreatic cancer cells was decreased dose-dependently by Suramin (Fig. 1a). In vivo, 75% of control animals and 17% of treated mice with MP tumors developed ascites (p< 0.05). Ascites in animals with C1 tumors occurred in 58% (control group) vs 25% (Suramin group). As expected,

Table 2 Effect of Suramin on Cell Cycle Phase Distribution

Cell line	Suramin μg/ml	S phase %	G0/1 phase %
MP	0	8	68
	800	4	75
Capan-2	0	31	60
	800	19	71
AP	0	12	81
	800	8	83
C1	0	22	60
	800	34	46

Human pancreatic cancer cells were incubated with (800 μg/ml) and without Suramin for 72 h. Cells were analyzed by flow cytometry after cell staining with propidium iodide

VEGF concentration in ascites was significantly reduced in Suramin-treated animals compared to the control groups (Fig. 1b).

Effect of Suramin In Vivo

Low concentration of Suramin (10 mg/kg) had no influence on tumor volume, metastatic behavior, development of ascites, and microvessel density in mice bearing MP tumors. At higher concentrations (60 mg/kg), Suramin showed effects on pancreatic cancers from all used cell lines (MP, AP, and C1).

The volume of tumors derived from the MP cell line was reduced to approximately 25% (3,387±690 vs 889±269 mm^3; $p<0.05$) (Fig. 2a). Suramin-treated animals of the AP and the C1 group developed tumors with about half the volume of the control group (2,052±190 vs 1,222±158 mm^3 and 1,103±159 vs 561±129 mm^3, $p<0.05$). For comparing the groups in terms of tumor spread, we used mean scores, which summarize local infiltration and distant metastasis. Treatment with Suramin resulted in a statistically significant reduction of tumor spread in the group with MP tumors. The incidence of local infiltration and distant metastasis in the control group was more than four times higher than in the treated group [10.9±2.0 vs 2.3±0.9 points; $p<0.01$] (Fig. 2b). The mean dissemination score in the AP and C1 groups diminished to 66 and 62%, respectively (17.1±2.7 vs 11.3±2.3 points and 6.1±0.8 vs 3.8±0.9 points). These differences were statistically not significant. The animal weight was recorded weekly after tumor induction and at autopsy. The average animal weight at autopsy in the control groups was comparable with the weight in the groups treated with 60 mg/kg Suramin (MP, 32.3±1.1 vs 32.8±0.9 g; AsPC, 31.4±1.6 vs 30.4±1.4 g; C1, 32.7±0.7 vs 31.6±1.3 g).

Effect of Suramin on Microvessel Density

The microvessel density as a parameter of angiogenic activity was determined by immunohistochemistry using Anti-CD31 as an endothelial marker. The tumor tissue samples collected from animals treated with 60 μg/kg Suramin revealed a significantly decreased density of microvessels compared to tissue of controls (MP, 49.3±4.3 vs 69.8±6.4/0.74 mm^2; AP, 55.5±6.7 vs 81.3±3.6/0.74 mm^2; C1, 50.5±6.2 vs 75.8±6/0.74 mm^2; $p<0.05$, respectively) (Fig. 2c). In comparison, TNP-470 reduced microvessel densities in tumors more efficiently than treatment with Suramin (MP, 24.8±3.7; AP, 26±3.4; C1, 26.9±2.5; $p<0.05$ vs Suramin).

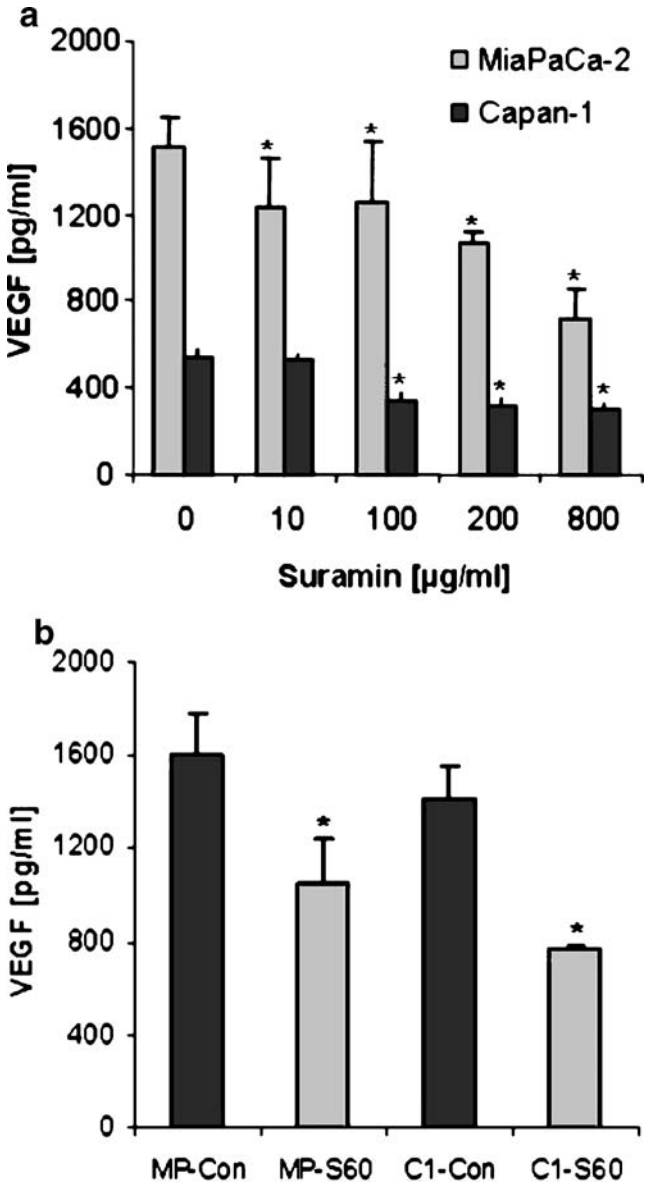

Figure 1 VEGF levels in the cell culture medium (**a**) and ascites (**b**) determined by ELISA. MP and C1 cells were incubated with different concentrations of Suramin. VEGF levels in ascites of animals with MP and C1 tumors treated with 60 mg/kg Suramin (*MP-S60* and *C1-S60*) were compared to control groups (*MP-Con* and *C1-Con*) ($* = p < 0.05$ vs control).

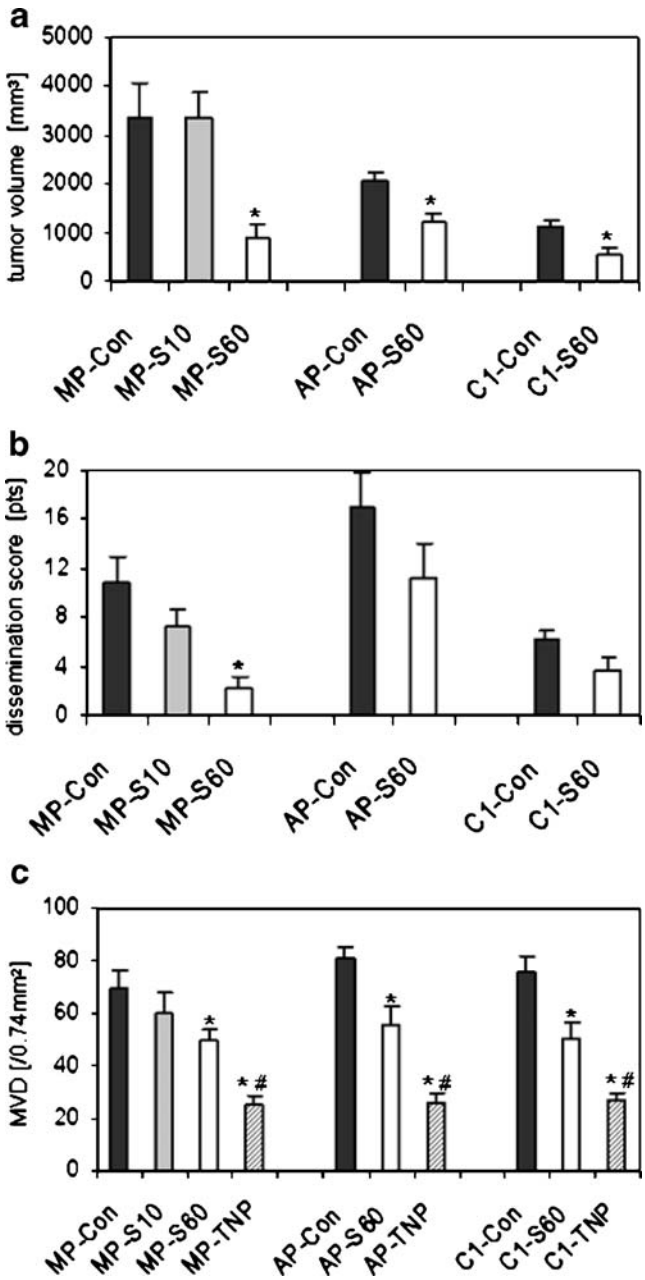

Figure 2 Tumor volume, dissemination scores, and microvessel densities in animals with tumors derived from MP, AP, and C1 cells. Mice were treated with 10 mg/kg (S10) or 60 mg/kg (S60) Suramin or 30 mg/kg TNP-470 (TNP). Each group comprised 12 animals ($* = p < 0.05$ vs control and $\# = p < 0.05$ vs Suramin).

Discussion

Suramin was initially developed to treat sleeping sickness. The drug exhibited antitumor activity first in the treatment of patients with HIV-associated lymphomas and Kaposi's sarcoma. This raised the possibility of using Suramin in the therapy of solid tumors. The substance has been reported to be active in hormone refractory prostate cancer, adrenocortical cancer, and indolent lymphomas[13]. By affecting various intracellular targets, like growth factors, enzymes associated with signal transduction, and DNA replication or polyamines, which are necessary for cell growth and differentiation, Suramin can inhibit tumor growth (Fig. 3). The ability of Suramin to decrease proliferation rates in vitro has been demonstrated in several types of cancer cells, including cells derived from stomach cancer, esophageal cancer, and non-small-cell lung cancer[14–16].

The present study is the first to evaluate Suramin for the treatment of human pancreatic cancer. Our in vitro results confirmed the inhibitory action of Suramin on human pancreatic cancer cells. In all five human pancreatic cancer cell lines, proliferation was decreased dose-dependently by Suramin. Cell viability was influenced at high doses in each cell line. These results indicate that Suramin acts in a cytostatic, rather than in a cytotoxic, manner. This assumption is emphasized by studies on alterations of the cell cycle. DNA staining with propidium iodide revealed a redistribution of cell cycle phases. Suramin caused accumulation of MP, Capan-2, and AP cells mainly in the G0/G1 phase, while the number of cells in the S phase was decreased. Other investigators have demonstrated similar effects of Suramin on the cell cycle in prostate carcinoma cells, glioma cells, and gastric carcinoma cells[14,17,18]. The changes in cell cycle were consistent across the cell lines except in C1 cells, where we observed an increased S phase and decreased G0/1 phase. As a marker for apoptosis, we determined AnnexinV staining of the cells. Apoptosis was not significantly modified after incubation with Suramin. Therefore, we assume an antiproliferative activity for Suramin in human pancreatic cancer cells that is partly based on alterations of cell cycle kinetics.

The effects of Suramin were further evaluated in an orthotopic nude mouse model of human pancreatic cancer, which was previously characterized for moderately to well differentiated C1 cells, poorly differentiated AP cells, and undifferentiated MP cells[10]. The in vivo results showed that low-dose concentrations of Suramin (10 mg/kg) had no influence on primary growth, metastasis, and angiogenesis in animals bearing MP tumors. The high dose of Suramin (60 mg/kg), equivalent to 300 mg/m^2 and achievable in the human setting[11], revealed a strong inhibition of local tumor growth and metastatic progression. The volumes of MP, AP, and C1 tumors in the treated groups were significantly smaller than those of the control animals. Dissemination score of the MP group was significantly reduced in the treated animals. In the AP and C1 groups, the dissemination score was decreased as well, but these effects were not statistically significant.

The role for Suramin as an antiangiogenic drug has been investigated both in vitro and in vivo[7,19]. To evaluate the antiangiogenic potential of Suramin in human pancreatic cancer, we compared its effects on neovascularization with those of TNP-470. TNP-470 (*O*-[chloroacetylcarbamyl]

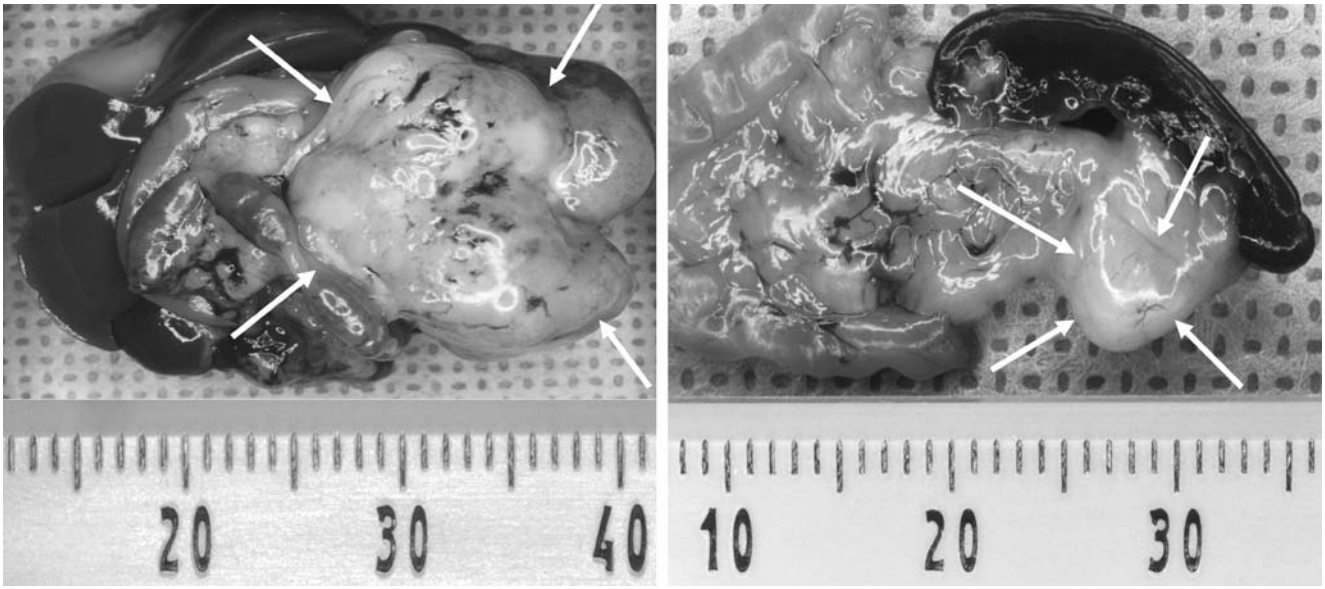

Figure 3 Effect of Suramin on primary tumor growth. Donor tumor fragments derived from MP cells were orthotopically implanted into the pancreatic tail of nude mice, which were killed after 14 weeks. Pancreatic tumor in control animal (*left*) and in an animal treated with 60 mg/kg Suramin (*right*).

fumagillol) is an analog of fumagillin and well known to inhibit neoangiogenesis via nonfactor-specific mechanisms. Numerous investigators have reported that TNP-470 inhibits its growth and metastasis of rodent tumors derived from human cancer cell lines[20,21]. In addition, this therapeutic compound is under clinical investigation in phase I/II studies for the treatment of several tumor types, including Kaposi's sarcoma, renal cell carcinoma, brain cancer, breast cancer, cervical cancer, and prostate cancer[22–24]. In a previous study, we demonstrated that TNP-470 strongly inhibits local tumor growth and metastatic tumor progression in our orthotopic nude mouse model of human pancreatic cancer[25]. Furthermore, immunohistochemical staining of the primary tumors with CD31, a common marker for microvessels, revealed a significant reduction of the neoangiogenic phenotype. A comparison of microvessel densities in the present study revealed that TNP-470 reduces this parameter of neoangiogenesis more efficiently than Suramin does. These findings indicate that Suramin is a less potent antiangiogenic substance in human pancreatic cancer than TNP-470. However, there appears to be some antiangiogenic activity of Suramin. Suramin acts as a functional VEGF-antagonist by binding to VEGF receptor-2 (KDR)[26]. Here, we demonstrated that MP and C1 cells constitutively secrete high levels of VEGF that can be dose-dependently decreased by exposing the cells to Suramin. In contrast to controls, treated animals bearing MP and C1 tumors developed fewer ascites and showed a significant reduction of VEGF levels in ascites.

We did not note any apparent side-effects of Suramin such as a change in food intake or activity in our study. As a surrogate marker of toxicity, animal weights were observed throughout the in vivo study and were not found to be different at autopsy in any group. Other investigators claim that the clinical use of Suramin is limited by its toxicity, which is mainly characterized by the development of a polyneuropathy[27]. As a consequence of Suramin's toxicity, the generation of Suramin analogs is currently being investigated and seems to be a promising approach to circumvent toxic side effects while preserving the advantages of Suramin's antitumor activities[28,29]. A reasonable alternative to Suramin analogs is the application of Suramin in low concentrations. Recently, it has been reported that low-dose administration of Suramin was able to improve the effects of chemotherapy in a mouse model of human breast cancer without enhancing host toxicity[30]. However, this approach has yet to be investigated in pancreatic cancer.

Conclusion

We demonstrated an inhibitory effect of Suramin on proliferation and viability of human pancreatic cancer cells. Therapy with Suramin resulted in a reduction of tumor size and metastatic spread in a clinical relevant orthotopic nude mouse model. In addition, we demonstrated an antiangiogenic effect of Suramin, as verified by the reduction of microvessel density in primary tumors of animals. In summary, our results strongly argue for further investigation of Suramin as a treatment modality in human pancreatic cancer.

References

1. Parker SL, Tong T, Bolden S, Wingo PA. Cancer statistics, 1997. CA Cancer J Clin 1997;47(1):5–27.
2. Kabbinavar F, Hurwitz HI, Fehrenbacher L, Meropol NJ, Novotny WF, Lieberman G, et al. Phase II, randomized trial comparing bevacizumab plus fluorouracil (FU)/leucovorin (LV) with FU/LV alone in patients with metastatic colorectal cancer. J Clin Oncol 2003;21(1):60–65.
3. Fiedler W, Mesters R, Tinnefeld H, Loges S, Staib P, Duhrsen U, et al. A phase 2 clinical study of SU5416 in patients with refractory acute myeloid leukemia. Blood 2003;102(8):2763–2767.
4. Hawking F. Suramin: with special reference to onchocerciasis. Adv Pharmacol Chemother 1978;15:289–322.
5. Coffey RJ Jr, Leof EB, Shipley GD, Moses HL. Suramin inhibition of growth factor receptor binding and mitogenicity in AKR-2B cells. J Cell Physiol 1987;132(1):143–148.
6. Vogelzang NJ, Karrison T, Stadler WM, Garcia J, Cohn H, Kugler J, et al. A phase II trial of Suramin monthly × 3 for hormone-refractory prostate carcinoma. Cancer 2004;100(1):65–71.
7. Gagliardi A, Hadd H, Collins DC. Inhibition of angiogenesis by Suramin. Cancer Res 1992;52(18):5073–5075.
8. Pesenti E, Sola F, Mongelli N, Grandi M, Spreafico F. Suramin prevents neovascularisation and tumour growth through blocking of basic fibroblast growth factor activity. Br J Cancer 1992; 66(2):367–372.
9. Ingber D, Fujita T, Kishimoto S, Sudo K, Kanamaru T, Brem H, et al. Synthetic analogues of fumagillin that inhibit angiogenesis and suppress tumour growth. Nature 1990;348(6301):555–557.
10. Hotz HG, Reber HA, Hotz B, Yu T, Foitzik T, Buhr HJ, et al. An orthotopic nude mouse model for evaluating pathophysiology and therapy of pancreatic cancer. Pancreas 2003;26(4):e89–e98.
11. Walz TM, Abdiu A, Wingren S, Smeds S, Larsson SE, Wasteson A. Suramin inhibits growth of human osteosarcoma xenografts in nude mice. Cancer Res 1991;51(13):3585–3589.
12. Weidner N. Tumoural vascularity as a prognostic factor in cancer patients: the evidence continues to grow. J Pathol 1998; 184(2):119–122.
13. La Rocca RV, Stein CA, Myers CE. Suramin: prototype of a new generation of antitumor compounds. Cancer Cells 1990;2(4):106–115.
14. Choe G, Kim WH, Park JG, Kim YI. Effect of Suramin on differentiation of human stomach cancer cell lines. J Korean Med Sci 1997;12(5):433–442.
15. Shin R, Naomoto Y, Kamikawa Y, Tanaka N, Orita K. Effect of Suramin on human esophageal cancer cells in vitro and in vivo. Scand J Gastroenterol 1997;32(8):824–828.
16. Fujiuchi S, Ohsaki Y, Kikuchi K. Suramin inhibits the growth of non-small-cell lung cancer cells that express the epidermal growth factor receptor. Oncology 1997;54(2):134–140.
17. Takano S, Gately S, Engelhard H, Tsanaclis AM, Brem S. Suramin inhibits glioma cell proliferation in vitro and in the brain. J Neuro-oncol 1994;21(3):189–201.
18. Qiao L, Pizzolo JG, Melamed MR. Effects of Suramin on expression of proliferation associated nuclear antigens in DU-145 prostate carcinoma cells. Biochem Biophys Res Commun 1994;201(2):581–588.
19. Bocci G, Danesi R, Benelli U, Innocenti F, Di Paolo A, Fogli S, et al. Inhibitory effect of Suramin in rat models of angiogenesis in vitro and in vivo. Cancer Chemother Pharmacol 1999;43(3):205–212.
20. Zeng J, Mei W, Huang H, Li X, Kong P. Inhibitory effect of angiogenesis inhibitor TNP-470 on human ACHN renal cell carcinoma. J Huazhong Univ Sci Technolog Med Sci 2002; 22(4):331–333.
21. Arisawa C, Kageyama Y, Kawakami S, Kihara K. TNP-470 combined with nicardipine suppresses in vivo growth of PC-3, a human prostate cancer cell line. Urol Oncol 2002;7(6):229–234.
22. Logothetis CJ, Wu KK, Finn LD, Daliani D, Figg W, Ghaddar H, et al. Phase I trial of the angiogenesis inhibitor TNP-470 for progressive androgen-independent prostate cancer. Clin Cancer Res 2001;7(5):1198–1203.
23. Cattelan AM, Trevenzoli M, Aversa SM. Recent advances in the treatment of AIDS-related Kaposi's sarcoma. Am J Clin Dermatol 2002;3(7):451–462.
24. Kruger EA, Figg WD. TNP-470: an angiogenesis inhibitor in clinical development for cancer. Expert Opin Investig Drugs 2000;9(6):1383–1396.
25. Hotz HG, Reber HA, Hotz B, Sanghavi PC, Yu T, Foitzik T, et al. Angiogenesis inhibitor TNP-470 reduces human pancreatic cancer growth. J Gastrointest Surg 2001;5(2):131–138.
26. Waltenberger J, Mayr U, Frank H, Hombach V. Suramin is a potent inhibitor of vascular endothelial growth factor. A contribution to the molecular basis of its antiangiogenic action. J Mol Cell Cardiol 1996;28(7):1523–1529.
27. Garcia-Schurmann JM, Schulze H, Haupt G, Pastor J, Allolio B, Senge T. Suramin treatment in hormone- and chemotherapy-refractory prostate cancer. Urology 1999;53(3):535–541.
28. Marchetti D, Reiland J, Erwin B, Roy M. Inhibition of heparanase activity and heparanase-induced angiogenesis by Suramin analogues. Int J Cancer 2003;104(2):167–174.
29. Meyers MO, Gagliardi AR, Flattmann GJ, Su JL, Wang YZ, Woltering EA. Suramin analogs inhibit human angiogenesis in vitro. J Surg Res 2000;91(2):130–134.
30. Song S, Yu B, Wei Y, Wientjes MG, Au JL. Low-dose Suramin enhanced paclitaxel activity in chemotherapy-naive and paclitaxel-pretreated human breast xenograft tumors. Clin Cancer Res 2004;10(18 Pt 1):6058–6065.

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Repeated Pancreatectomy after Pancreatoduodenectomy

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Abstract
Background When pancreatic duct dilatation is found in the patient having undergone pancreatoduodenectomy (PD), observation is chosen in most cases. Similarly, recurrent tumor in the remnant pancreas of invasive ductal carcinoma (IDC) of the pancreas is seldom indicated for resection. We have aggressively performed repeated pancreatectomy for these cases and obtained good results.
Methods Repeated pancreatectomy after PD was performed for three types of circumstances: (1) pancreatodigestive anastomotic stricture; (2) neoplasm after intraductal papillary mucinous neoplasm (IPMN); and (3) recurrence of IDC of the pancreas.
Results Resection of anastomosis and reanastomosis was performed for pancreatodigestive stricture in four patients. Symptoms derived from pancreatitis in three patients resolved by the second operation and did not recur during follow-up. None of the four patients required pancreatic enzyme substitution because of clinically overt malabsorption, and the defecation frequency of the four patients was within twice a day. Mild diabetes mellitus has been identified in only one patient who had diabetes mellitus before the second surgery. Completion pancreatectomy and pancreatic tail resection was performed for recurrence in two patients and IDC in one patient, respectively, after PD for IPMN. Intrapancreatic recurrences of IPMN in two patients existed in the main pancreatic ducts. As CT revealed pancreatic duct dilatation but not intraductal tumors, recurrences were not correctly diagnosed before the second operation. Completion pancreatectomy was performed for recurrence of IDC in two patients. One patient who underwent completion pancreatectomy for recurrence of IDC survived 66/44 months after the first/second operation.
Conclusion Repeated pancreatectomy should be performed for patients with pancreatodigestive anastomotic stricture to preserve remnant pancreatic function and for patients with neoplasm or pancreatic duct dilatation after PD for IPMN, and repeated pancreatectomy for recurrence of IDC might be indicated for selected patients.

Keywords Pancreatoduodenectomy · Pancreatic function · Intraductal papillary mucinous neoplasm · Invasive ductal carcinoma of the pancreas

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Introduction

Pancreatoduodenectomy (PD) is performed not only for patients with tumors, but for other indications, such as trauma, chronic pancreatitis, etc. Advances in diagnostic techniques such as computed tomography (CT), magnetic resonance imaging (MRI), and endoscopic ultrasonography (EUS) have revealed increasing numbers of patients with malignant tumors at early stage and benign tumors such as intraductal papillary mucinous neoplasm (IPMN)[1]. Mortality of PD has improved because of improvement in operative techniques and perioperative management[2,3]. For these reasons, patients with long post PD survival have

been increasing in number. Although there are several problems for long survivors, these problems have not received much attention.

We performed repeated pancreatectomy after PD for three types of indications: (1) pancreatodigestive anastomotic stricture; (2) neoplasm after IPMN; and (3) recurrence of invasive ductal carcinoma (IDC) of the pancreas. Some brief clinical case reports describing resection of the remnant pancreas have been published[4–7], but to our knowledge, detailed description of repeated pancreatectomy after PD has not yet been presented. Therefore, in the present study, we retrospectively analyzed our experiences with nine patients undergoing repeated pancreatectomy after PD.

Patients and Methods

Between July 1985 and March 2005, 400 patients underwent pancreatoduodenectomy (PD) at the Department of Surgery, Teikyo University Hospital. In eight of these patients and one who underwent initial surgery at another hospital, repeated pancreatectomy was performed. Thus, nine patients (six men and three women, 38–67 years old, mean 61.9 years at initial surgery) were eligible for this study (Table 1). At initial surgery, the pylorus was preserved in eight of the nine patients. Pancreatic anastomoses of initial surgery were pancreatojejunostomy in seven patients and pancreatogastrostomy in two patients. Indications for PD at initial surgery were intraductal papillary mucinous adenoma (IPMA) in three, IDC of the pancreas in two, ampullary carcinoma in one, intraductal papillary mucinous carcinoma (IPMC) in one, tumor-forming pancreatitis in one, and bleeding duodenal ulcer in one. The interval between initial and second surgery was 22–124 months (mean 60.6) in the 9 patients. Indications of our institution for repeated pancreatectomy after pancreatoduodenectomy were pancreatodigestive anastomotic stricture, neoplasm after PD for IPMN, and IDC recurrence.

Pancreatodigestive Anastomotic Stricture

Pancreatodigestive anastomotic stricture was considered to exist if the diameter of the pancreatic duct had increased to 7 mm or greater in diameter as delineated by CT or MR cholangiopancreatography. Pancreatic duct dilatation accompanying chronic pancreatitis existing before PD was excluded. Reoperation was indicated for patients who complained symptoms and/or were expected long-term survivors.

Resection of anastomosis and reanastomosis was performed by the following steps (Fig. 1): (1) about 1 cm (0.7–1.2 cm, average 1.1 cm) of the proximal portion of the pancreas was dissociated from the jejunal limb or stomach. (2) The anterior wall of the pancreatic duct was opened longitudinally about 1 cm and pancreatic stone was removed if existing. (3) End-to-side pancreatojejunostomy was created in two layers of sutures, with the outer layer comprising the capsular parenchyma of the pancreatic stump and jejunal wall, and the inner layer the pancreatic duct and the whole jejunal muscularis without stent tube. One patient (patient 3) also had stricture of choledocojejunostomy and underwent reanastomosis of choledocojejunostomy simultaneously.

Neoplasm after PD for IPMN

Thirty-two patients underwent pancreatoduodenectomy for IPMN of the pancreas at our institution. Three patients underwent repeated pancreatectomy for neoplasm in the remnant pancreas.

IDC Recurrence

One hundred thirty-five patients underwent pancreatoduodenectomy for IDC of the pancreas between July 1985 and March 2005. Eighty-three patients were followed-up for more than 12 months. In three of the 83 patients, recurrent

Table 1 Background of Nine Patients Who Underwent Repeated Pancreatectomy after Pancreatoduodenectomy

Pt	Age (years)/Gender at Initial Surgery	Diagnosis at Initial Surgery	Operative Procedure	Reconstruction Method
1	57/M	Duodenal ulcer bleeding	Whipple's PD[a]	PJ
2	58/F	Ampullary carcinoma	PPPD	PG
3	54/M	Tumor-forming pancreatitis	PPPD	PJ
4	48/M	IPMA	PPPD	PG
5	62/M	IPMA	PPPD	PJ
6	38/M	IPMC	PPPD	PJ
7	67/M	IPMA	PPPD	PJ
8	72/F	IDC	PPPD	PJ
9	52/F	IDC	PPPD	PJ

PD Pancreatoduodenectomy, *PJ* pancreatoduodenectomy, *PPPD* pylorus preserving pancreatoduodenectomy, *PG* pancreatogastorostomy, *IPMA* intraductal papillary mucinous adenoma, *IPMC* intraductal papillary mucinous carcinoma, *IDC* invasive ductal carcinoma
[a] Whipple's PD means pancreatoduodenectomy with distal gastrectomy.

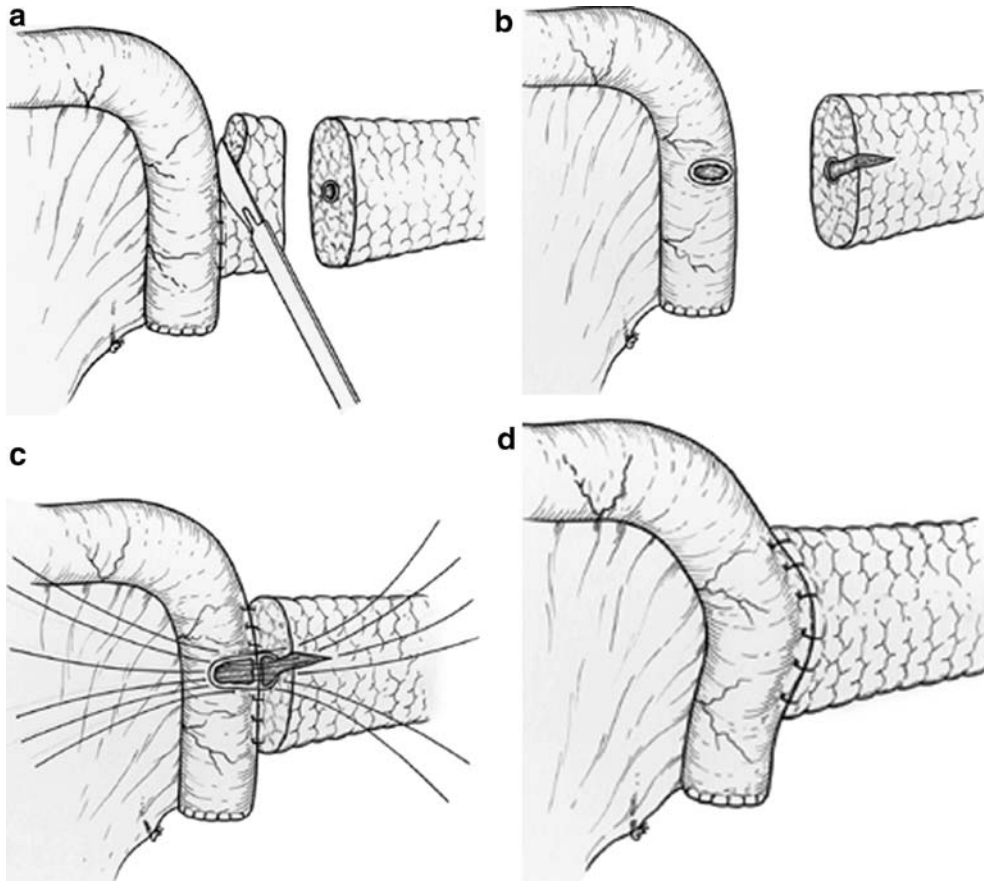

Figure 1 Resection of anastomosis and reanastomosis. **a** About 1 cm of the proximal portion of the pancreas was dissociated from the jejunum. **b** Anterior wall of pancreatic duct was opened longitudinally about 1 cm and the anastomotic hole of the jejunum was extended to fit the pancreatic duct orifice. **c** End-to-side pancreatojejunostomy was created in two layers of sutures. After the posterior outer layer composed of the capsular parenchyma of the pancreatic stump and jejunal wall, the inner layer composed of the pancreatic duct, and the whole jejunal muscularis were sutured without stent tube. **d** Reanastomosis was completed after the anterior outer layer composed of the capsular parenchyma of the pancreatic stump and the jejunal wall were sutured.

tumor developed in the remnant pancreas and two underwent a second operation for the recurrent tumor. The third patient was not indicated for a second operation because of multiple lymph node metastasis.

Analysis

Preoperative diagnosis, operative procedure, postoperative course, and long-term outcome were analyzed retrospectively. Clinical status was determined by examinations in our institution, and from contact with the referring physician. The diagnosis of diabetes mellitus was made based on criteria set by the 1985 World Health Organization study group on diabetes mellitus[8].

Results

Outcomes of the nine patients are shown in Table 2. Operative morbidity and mortality rates were 11.1 and 0%, respectively. One patient (patient 4) developed ileus 7 days after the operation and improved by conservative therapy. Life-threatening complications did not arise in any of the patients. All patients were observed for a minimum 5 months (mean 45.6; range 5–102) after the second operation.

Pancreatodigestive Anastomotic Stricture

In four patients, repeated pancreatectomy was performed for pancreatodigestive anastomotic stricture as described above. All four patients presented with pancreatic duct dilatation on CT. Pancreatolithiasis was identified at the anastomotic site on CT in two patients (patients 1 and 3) with pancreatojejunostomy. Three patients suffered from symptoms derived from pancreatitis: two with abdominal pain and one with diarrhea. Deteriorating diabetes mellitus was identified in one patient.

Histopathologically, abundant fibrosis in the pancreatic parenchyma was found in all four patients, but all were free of any neoplastic changes. Their postoperative courses were uneventful. Pancreatitis-derived symptoms in three patients were resolved by the second operation, and there was no recurrence during follow-up. In all four patients, the caliber of the main pancreatic duct was decreased to less than 4 mm, and hardly increased during follow-up. Body weight ratios, defined as weight 24 months after the second operation per preoperative weight, were 100–106% (mean 102.3%). During follow-up, none required pancreatic enzyme substitution because of clinically overt malabsorption, and defecation frequency of all four patients was within twice a day. Diabetes mellitus was identified in only one patient (patient 2), presenting with diabetes mellitus not

Table 2 Results of Nine Patients Who Underwent Repeated Pancreatectomy after Pancreatoduodenectomy

Pt	Interval (months) from Initial Surgery	CT Findings	Preoperative Diagnosis at Repeated Pancreatectomy	Operative Procedure	Final Diagnosis	Prognosis
1	124	Pancreatic duct dilatation with pancreatolithiasis	Anastomotic stricture	Resection of anastomosis and reanastomosis	Benign anastomotic stricture	Alive 213/89 months after the first/second operation
2	102	Pancreatic duct dilatation	Anastomotic stricture	Resection of anastomosis and reanastomosis	Benign anastomotic stricture	Alive 204/102 months after the first/second operation
3	99	Pancreatic duct dilatation with pancreatolithiasis	Anastomotic stricture	Resection of anastomosis and reanastomosis	Benign anastomotic stricture	Alive 127/28 months after the first/second operation
4	44	Pancreatic duct dilatation	Anastomotic stricture	Resection of anastomosis and reanastomosis	Benign anastomotic stricture	Alive 94/50 months after the first/second operation
5	42	Pancreatic duct dilatation	Anastomotic stricture	Resection of anastomosis and reanastomosis	Recurrence	Third operation was performed after 1 month
			Recurrence	Completion pancreatectomy	Recurrence	Alive 94/52 months after the first/third operation
6	38	Pancreatic duct dilatation	Anastomotic stricture	Completion pancreatectomy	Recurrence	Alive 60/24 months after the first/second operation
7	45	Solitary LDA	Secondary carcinogenesis	Pancreatic tail resection	Secondary carcinogenesis	Alive 61/16 months after the first/second operation
8	29	Solitary LDA LDA in the liver	Recurrence	Pancreatic tail resection lateral segmentectomy	Recurrence	Dead 34/5 months after the first /second operation
9	22	Multiple LDA	Recurrence	Completion pancreatectomy	Recurrence	Dead 66/44 months after the first/second operation

LDA Low density area

requiring insulin treatment before the second surgery, and to date, this status has not deteriorated.

Neoplasm after PD for IPMN

Three patients underwent repeated pancreatectomy after PD for IPMN. Patient 5 underwent PD for branch-type IPMN. Histopathological diagnosis was adenoma and the surgical margin of the pancreatic duct was negative for neoplastic change. CT 42 months after PD revealed dilatation of the pancreatic duct, the diagnosis was stricture of pancreatojejunostomy, and a second operation was performed. Resection of anastomosis was performed as described above. In the resected specimen, an 8-mm papillary tumor was found in the pancreatic duct. Histopathologically, the tumor was diagnosed as papillary adenocarcinoma and the surgical margin was positive for cancer invasion. Recurrence of IPMN was suggested. He underwent completion pancreatectomy (third operation) 3 weeks later. He is alive 52 months after the third operation without recurrence at this writing.

Patient 6 underwent PD for combined-type IPMC. Histopathological diagnosis was papillary adenocarcinoma. The surgical margin of PD was free of atypical cells. CT 37 months after PD revealed dilatation of the pancreatic duct, but intraductal tumor and low-density area in the remnant pancreas were not identified (Fig. 2a). The diagnosis was stricture of pancreatojejunostomy and a second operation was performed. Intraoperative ultrasonography revealed an intraductal tumor near the pancreatojejunostomy, and the patient was diagnosed with recurrent IPMC and underwent completion pancreatectomy. In the resected specimen, a 15-mm papillary tumor existed near the anastomotic site (Fig. 2b). Histopathological diagnosis was papillary adenocarcinoma, similar to the findings of the specimen at the first oeration. He is alive 24 months after the second operation without recurrence at this writing.

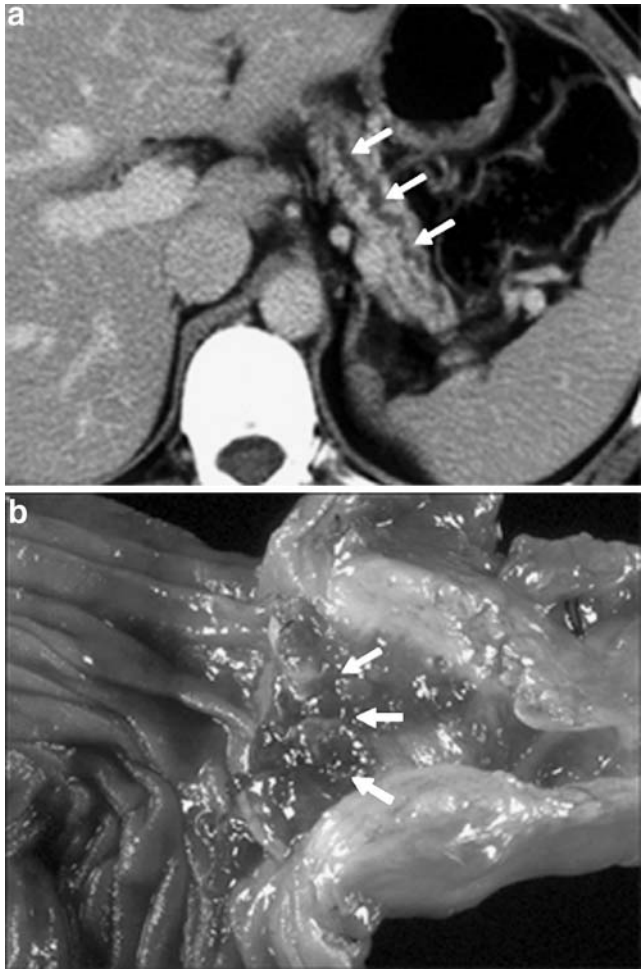

Figure 2 Recurrence of invasive IPMC in patient 6. **a** CT 37 months after PD revealed dilatation of the pancreatic duct (arrows) but intraductal tumor and low-density area in the remnant pancreas were not identified. **b** Completion pancreatectomy was performed. In the resected specimen, a 15-mm papillary tumor (arrows) existed near the anastomotic site.

In patient 7, CT 45 months after PD for IPMA revealed a low-density area 2 cm in size in the tail of the remnant pancreas. Diagnosis was IDC of the pancreas. Pancreatic tail resection and splenectomy were performed, with the pancreatic body measuring 4 cm being preserved. Regional lymph node dissection was also performed. Histopathological diagnosis was IDC. The postoperative course was uneventful and he is alive without recurrence or onset of diabetes mellitus 16 months after surgery at this writing.

IDC Recurrence

Patient 8 had a past history of PD combined with superior mesenteric vein resection for IDC. The original tumor was histopathologically diagnosed as a moderately differentiated tubular adenocarcinoma with infiltration to microvessels and invasion to retropancreatic soft tissue, common bile duct, and superior mesenteric vein (pStage IV, pT3, pN0, M0)[24]. The surgical margins of the resected specimen were free of atypical cells. Twenty-nine months after the initial surgery, CT showed a low-density lesion in the tail of the pancreas and two low-density lesions in the left lateral segment of the liver. A second operation, pancreatic tail resection, with splenectomy and left lateral segmentectomy, was performed. The second tumor and liver metastasis were all moderately differentiated tubular adenocarcinoma. The postoperative course was uneventful except onset of diabetes mellitus. The patient died of liver metastasis 5 months after the second operation.

Patient 9, about whom we previously reported[6], had a past history of PD for IDC. The original tumor was histopathologically diagnosed as papillary adenocarcinoma with infiltration to microvessels and invasion to retropancreatic soft tissue and regional lymph nodes (pStage III, pT3, pN1, M0)[9]. The surgical margins of the resected specimen were free of atypical cells. Twenty months after the initial surgery, CT showed multiple low-density lesions in the body and tail of the pancreas, but no distant metastasis (Fig. 3). A second operation, completion pancreatectomy, with splenectomy and distal gastrectomy, was performed. The second tumor was a papillary adenocarcinoma, showing similar histopathological findings including staining of Ki-67 and p53 protein, and the same pattern of K-ras point mutation as the first tumor. The postoperative course was uneventful except onset of diabetes mellitus, and the patient died of chemotherapy-related complications at another hospital 44 months after the second operation.

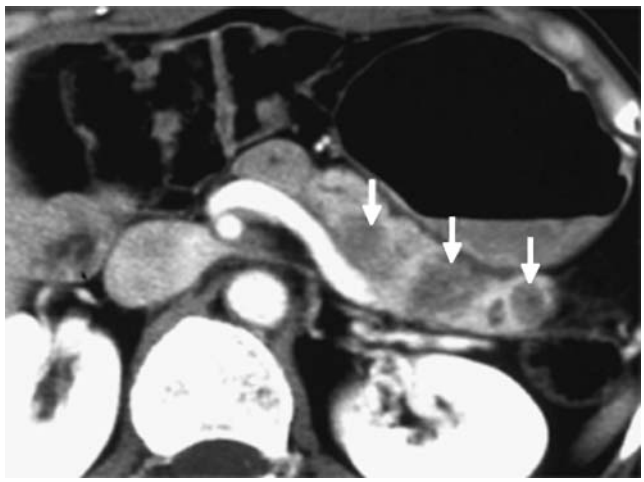

Figure 3 Recurrence of IDC in patient 9. CT 20 months after PD showed multiple low-density lesions (arrows) in the body and tail of the pancreas.

Discussion

We performed repeated pancreatectomy after PD for three circumstances—pancreatodigestive anastomotic stricture, neoplasm after IPMN, and recurrence of IDC.

Pancreatodigestive anastomotic stricture sometimes develops after PD. Patency of the pancreatoenterostomy is one of the most important factors affecting function of the remnant pancreas and quality of life of the patient. Stenosis of the pancreatoenterostomy induces obstructive chronic pancreatitis, a particular form of chronic pancreatitis that occurs as a consequence of primary stenosis or obstruction of the main pancreatic duct, resulting in inflammation of the distal pancreas[10]. The pathogenesis of pain in chronic pancreatitis remains unexplained, but it is believed that in obstructive pancreatitis, the demonstrated elevated pancreatic duct and tissue pressures are presumed to contribute to the generation of pain[11,12].

Some clinical studies have reported that pancreatodigestive anastomotic stricture after PD induces impairment of pancreatic exocrine function[13–16]. Impairment of exocrine function derived from obstructive chronic pancreatitis is thought to be reversible to some degree. It was reported that pancreatic impairment caused by ligation of the major pancreatic duct could be restored in 7 to 46 days by anastomosis of the major pancreatic duct using a microsurgical technique[17]. In patients with dilated pancreatic ducts, drainage of obstructed ducts with pancreatojejunostomy is a reliable means of providing pain relief, with minimal loss of endocrine and exocrine function[18,19]. Pancreatic ductal drainage for chronic pancreatitis will provide complete or significant relief of pain in greater than 80% of patients when the duct is dilated[20,21]. These facts imply that exocrine dysfunction of remnant pancreas due to pancreatodigestive anastomotic stricture could be improved by early treatment. However, there is only one case report from our institution describing the treatment for obstructive chronic pancreatitis after PD[22]. Therefore, it has not been evident whether the remnant pancreatic exocrine function after PD can be preserved or not.

As for endocrine function, although pancreatic islets are preserved long after acinar cells are destroyed and replaced by fibrosis after the obstruction of the pancreatic duct[23], persistent patency of the anastomotic site of pancreatoenterostomy may be a major factor influencing long-term insulin secretion after the operation[24]. In the present series, we performed repeated pancreatectomy for pancreatodigestive anastomotic stricture with the purpose of relieving symptoms and preserving the remnant pancreatic function. Our results showed that resection of anastomosis and reanastomosis in patients with obstructive chronic pancreatitis had low morbidity and succeeded in relieving pain and preserving exocrine and endocrine function of all four patients over long-term follow-up. We believe that resection of anastomosis and reanastomosis is recommendable especially for patients with symptoms derived from pancreatitis because this procedure is safe and is able to relieve symptoms. And also, we consider that resection of anastomosis and reanastomosis is preferable for asymptomatic patients who underwent PD and are expected long-term survivors to preserve the remnant pancreatic function.

IPMN is considered to be a pancreatic neoplasm with relatively favorable prognosis, whereas IPMN, both invasive and noninvasive, has been reported to recur after resection regardless of whether the surgical margin after initial resection is positive or negative[25,26]. The recurrence rates of IPMN after resection are 13–50% in the literature[6,25–27]. One of the major sites of recurrence of IPMN after resection is the remnant unresected pancreas[26,27], where it may have occurred because of residual dysplastic tissue at the surgical margins, multifocal disease with synchronous tumor at this site, or metachronous lesions developing at this site because of a neoplastic tendency of the entire gland[25]. Although distant metastasis is generally considered unresectable and its prognosis is poor, repeated pancreatectomy is recommended when intrapancreatic recurrence without distant metastasis is found[25,26]. However, the long-term outcome of patients undergoing repeated pancreatectomy for the recurrence of IPMN is unknown. In our series, two patients with recurrent tumor of IPMN in the residual pancreas survived 52 and 24 months after reoperation. We believe that good outcome could be obtained by resection of intrapancreatic recurrence of IPMN.

Christensen et al.[28] classified the CT patterns of intrapancreatic recurrence after resection of IPMN into two types: cystic lesion resembling branch-duct tumor and dilatation of the main pancreatic duct. In our study, both of the intrapancreatic recurrences existed in the main pancreatic duct in two patients, and they were invasive carcinomas. It is difficult to differentiate pancreatodigestive anastomotic stricture from intrapancreatic recurrence in the main pancreatic duct[29]. Intraductal tumor is not able to be identified on CT, and endoscopic retrograde pancreatography is often impossible for the patient who has undergone PD. In the present study, we could not identify the recurrent lesions in the main pancreatic duct in two patients (patients 5 and 6) before the second operation. Therefore, we would like to emphasize that repeated pancreatectomy is recommended for patients with pancreatic duct dilatation after PD for IPMN to preserve remnant pancreatic function as well as differentiate between benign anastomotic stricture and intrapancreatic recurrence.

We also performed repeated pancreatectomy for IDC in a patient (patient 7) who had undergone PD for IPMN. IPMN is one of the risk factors of IDC of the pancreas. Yamaguchi

et al.[30] reported that 7 of 76 (9.6%) patients with IPMN had IDC, speculating that the entire pancreatic ductal system including the neoplastic epithelium of IPMN might already be in a premalignant condition. Clinicians should pay attention to not only the possible recurrence of IPMN but also to the development of IDC in patients who had undergone PD for IPMN. Despite the recent advances in diagnostic modalities and treatment methods, the clinical course of patients with IDC of the pancreas remains dismal[31]. The most causative factor for the poor prognosis is the high recurrence rate of the cancer. In contrast to IPMN, the recurrence of IDC in the remnant pancreas is rare. The most frequent recurrence patterns for pancreatic cancer after resection were local recurrence, hepatic metastasis, and peritoneal dissemination. In most cases with recurrence, the tumor is far advanced and there is usually no indication for surgical resection. There are three possibilities for the mode of tumor development in the remnant pancreas after resection for IDC: metachronous intrapancreatic metastasis, local recurrence, or second primary occurrence. It is assumed that only surgical resection is able to provide any cure for primary IDC, although it has so far not been clear whether resection of remnant pancreatic recurrence could provide long-term survival. There are a few reports of resectable remnant pancreatic adenocarcinoma after pancreatectomy for IDC[5–7,32]. The maximal postoperative observation time of these reports was 10 months. In this series, we performed repeated pancreatectomy for IDC recurrence in the remnant pancreas in two patients. We considered that the second tumors of both patients were intrapancreatic metastasis for the following reasons: (1) at the initial operation, the surgically cut end of the pancreas was apparently free of atypical cells, (2) histopatholigically, there were similar findings in the first and second tumors, with both tumors showing immunohistochemical staining of Ki-67 and p53 protein (patient 9), and (3) in a molecular biological study of both the first and second tumors, the same pattern of K-ras point mutation was detected (patient 9). However, we cannot completely rule out the possibility of other patterns and cannot definitively prove the tumor etiology. In any event, our case (patient 9) demonstrated that repeated pancreatectomy could provide long survival for patients with recurrence in the remnant pancreas after PD for IDC.

A drawback of the present study, of course, is the limited number of patients. Further follow-up and additional investigations with larger patient populations will be needed before definitive conclusions can be drawn.

Nonetheless, we believe that repeated pancreatectomy should be performed for patients with pancreatodigestive anastomotic stricture to relieve pain and preserve the remnant pancreatic function and for patients with neoplasm or pancreatic duct dilatation after PD for IPMN to obtain good outcome, and that repeated pancreatectomy for IDC recurrence might be indicated for selected patients.

References

1. Miura F, Takada T, Amano H, Yoshida M, Furui S, Takeshita K. Diagnosis of pancreatic cancer. HPB 2006;8:208–213.
2. Yamaguchi K, Tanaka M, Chijiiwa K, Nagakawa T, Imamura M, Takada T. Early and late complications of pylorus-preserving pancreatoduodenectomy in Japan 1998. Journal of Hepato–biliary–Pancreatic Surgery 1999;6:303–311.
3. Cameron JL, Pitt HA, Yeo CJ, Lillemoe KD, Kaufman HS, Coleman J. One hundred and forty-five consecutive pancreaticoduodenectomies without mortality. Ann Surg 1993;217:430–435 (discussion 435–438).
4. Niiyama H, Yamaguchi K, Shimizu S, Yokohata K, Chijiiwa K, Yonemasu H, et al. Pancreatic carcinoma in remnant pancreas after pancreatectomy for mucinous cystadenoma. European Journal of Gastroenterology and Hepatology 1998;10: 703–707.
5. Eriguchi N, Aoyagi S, Imayama H, Okuda K, Hara M, Fukuda S, et al. Resectable carcinoma of the pancreatic head developing 7 years and 4 months after distal pancreatectomy for carcinoma of the pancreatic tail. Journal of Hepato–biliary–Pancreatic Surgery 2000;7:316–320.
6. Wada K, Takada T, Yasuda H, Amano H, Yoshida M. A repeated pancreatectomy in the remnant pancreas 22 months after pylorus-preserving pancreatoduodenectomy for pancreatic adenocarcinoma. Journal of Hepato–biliary–Pancreatic Surgery 2001;8: 174–178.
7. Takamatsu S, Ban D, Irie T, Noguchi N, Kudoh A, Nakamura N, et al. Resection of a cancer developing in the remnant pancreas after a pancreaticoduodenectomy for pancreas head cancer. J Gastrointest Surg 2005;9:263–269.
8. Alberti K, Hockaday T. Diabetes mellitus. In Weatherall D, Ledingham J, Warrell D. eds. Oxford textbook of medicine, 2nd ed. New York: Oxford Medical Publications, 1987, pp 51–101.
9. Sobin L, Wittekind C. TNM Classification of Malignant Tumours, 6th ed. New York: Wiley-Liss, 2002.
10. Suda K, Mogaki M, Oyama T, Matsumoto Y. Histopathologic and immunohistochemical studies on alcoholic pancreatitis and chronic obstructive pancreatitis: Special emphasis on ductal obstruction and genesis of pancreatitis. Am J Gastroenterol 1990; 85:271–276.
11. Bradley EL, III. Pancreatic duct pressure in chronic pancreatitis. Am J Surg 1982;144:313–316.
12. Jalleh RP, Aslam M, Williamson RC. Pancreatic tissue and ductal pressures in chronic pancreatitis. Br J Surg 1991;78: 1235–1237.
13. Hashimoto N, Fujiwara H, Nomura H, Ohyanagi H. In vivo evaluation of long-term patency of pancreaticoenterostomy after Whipple operation. Hepatogastroenterology 2000;47:866–868.
14. Hall RI, Rhodes M, Isabel-Martinez L, Kelleher J, Venables CW. Pancreatic exocrine function after a sutureless pancreaticojejunostomy following pancreaticoduodenectomy. Br J Surg 1990;77:83–85.
15. Kono T, Okazumi S, Mochizuki R, Ootsuki K, Shinotou K, Matsuzaki H, et al. Clinical application of (11)C-methionine positron emission tomography for evaluation of pancreatic function. Pancreas 2002;25:20–25.
16. Monill J, Pernas J, Clavero J, Farre A, Morales A, Gonzalez M, et al. Pancreatic duct after pancreatoduodenectomy: Morphologic and functional evaluation with secretin-stimulated MR pancrea-

tography. AJR American Journal of Roentgenology 2004;183: 1267–1274.
17. Tiscornia OM, Dreiling DA. Recovery of pancreatic exocrine secretory capacity following prolonged ductal obstruction. Bicarbonate and amylase response to hormonal stimulation. Ann Surg 1966;164:267–270.
18. Jordan GL Jr, Strugm BS, Crowder WE. Current status of pancreatojejunostomy in the management of chronic pancreatitis. Am J Surg 1977;133:46–51.
19. Prinz RA, Kaufman BH, Folk FA, Greenlee HB. Pancreaticojejunostomy for chronic pancreatitis. Two- to 21-year follow-up. Arch Surg 1978;113:520–525.
20. Prinz RA, Greenlee HB. Pancreatic duct drainage in 100 patients with chronic pancreatitis. Ann Surg 1981;194:313–320.
21. Warshaw AL. Conservation of pancreatic tissue by combined gastric, biliary, and pancreatic duct drainage for pain from chronic pancreatitis. Am J Surg 1985;149:563–569.
22. Amano H, Takada T, Ammori BJ, Yasuda H, Yoshida M, Uchida T, et al. Pancreatic duct patency after pancreaticogastrostomy: Long-term follow-up study. Hepatogastroenterology 1998;45:2382–2387.
23. Idezuki Y, Goetz FC, Lillehei RC. Late effect of pancreatic duct ligation on beta cell function. Am J Surg 1969;117:33–39.
24. Kasahara K, Yamashita Y, Ishibashi H, Fukumoto T, Tenmoku S, Kashii A, et al. Factors influencing insulin secretion after pancreatoduodenectomy. Jpn J Surg 1987;17:445–454.
25. Chari ST, Yadav D, Smyrk TC, DiMagno EP, Miller LJ, Raimondo M, et al. Study of recurrence after surgical resection of intraductal papillary mucinous neoplasm of the pancreas. Gastroenterology 2002;123:1500–1507.
26. Sho M, Nakajima Y, Kanehiro H, Hisanaga M, Nishio K, Nagao M, et al. Pattern of recurrence after resection for intraductal papillary mucinous tumors of the pancreas. World J Surg 1998;22: 874–878.
27. Cuillerier E, Cellier C, Palazzo L, Deviere J, Wind P, Rickaert F, et al. Outcome after surgical resection of intraductal papillary and mucinous tumors of the pancreas. Am J Gastroenterol 2000;95: 441–445.
28. Christensen JA, Fletcher JG, Fidler JL, Wold PB, Binstock AJ, Smyrk T, et al. Intraductal papillary mucinous neoplasms of the pancreas: CT patterns of recurrence and multiobserver performance in detecting recurrent neoplasm after surgical resection. AJR American Journal of Roentgenology 2004;183: 1367–1374.
29. Sugiyama M, Abe N, Tokuhara M, Masaki T, Mori T, Takahara T, et al. Magnetic resonance cholangiopancreatography for postoperative follow-up of intraductal papillary-mucinous tumors of the pancreas. Am J Surg 2003;185:251–255.
30. Yamaguchi K, Ohuchida J, Ohtsuka T, Nakano K, Tanaka M. Intraductal papillary-mucinous tumor of the pancreas concomitant with ductal carcinoma of the pancreas. Pancreatology 2002;2: 484–490.
31. Nakagohri T, Kinoshita T, Konishi M, Inoue K, Takahashi S. Survival benefits of portal vein resection for pancreatic cancer. Am J Surg 2003;186:149–153.
32. D'Amato A, Gentili V, Santella S, Boschetto A, Pronio A, Montesani C. Carcinoma of the pancreatic remnant developing after pancreaticoduodenectomy for adenocarcinoma of the head of pancreas. Chir Ital 2002;54:539–544.

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Partial Portal Vein Ligation Plus Thioacetamide: A Method to Obtain a New Model of Cirrhosis and Chronic Portal Hypertension in the Rat

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Abstract To obtain a new model of chronic portal hypertension in the rat, two classical methods to produce portal hypertension, partial portal vein ligation and the oral administration of thioacetamide (TAA), have been combined. Male Wistar rats were divided into four groups: 1 (control; $n=10$), 2 [triple partial portal vein ligation (TPVL); $n=9$], 3 (TAA; $n=11$), and 4 (TPVL plus TAA; $n=9$). After 3 months, portal pressure, types of portosystemic collateral circulation, laboratory hepatic function tests (aspartate aminotransferase, alanine aminotransferase, bilirubin, alkaline phosphatase, and gamma-glutamyl transpeptidase) and liver histology were studied. The animals belonging to group 2 (TPVL) developed extrahepatic portosystemic collateral circulation, associated with mesenteric venous vasculopathy without hepatic destructurization or portal hypertension. Animals from group 3 (TAA) developed cirrhosis and portal hypertension but not extrahepatic portosystemic collateral circulation, or mesenteric venous vasculopathy. Finally, the animals from group 4 (TPVL+TAA) developed cirrhosis, portal hypertension, portosystemic collateral circulation, and mesenteric venous vasculopathy. The association of TPVL and TAA can be used to obtain a model of chronic portal hypertension in the rat that includes all the alterations that patients with hepatic cirrhosis usually have. This could, therefore, prove to be a useful tool to study the pathophysiological mechanisms involved in these alterations.

Keywords Portal hypertension · Partial portal vein ligation · Thioacetamide · Cirrhosis · Portosystemic collateral circulation

Introduction

The most frequently used models to study portal hypertension in the rat are cirrhosis induced by carbon tetrachloride (CCl_4) or by thioacetamide (TAA), both hepatotoxins[1–3], and prehepatic portal hypertension induced by partial ligation of the portal vein (PVL)[4–6].

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Both rats with cirrhosis and those with PVL[5,8] present hyperdynamic systemic and splanchnic circulation. More specifically, with the latter, the animals can be considered to suffer from splanchnic hyperemia[6,8] with the subsequent increase in splanchnic blood flow, which in turn plays an important role in the maintenance of portal hypertension[6,8,9]. This pathological increase in the portal venous inflow is the basis of the "forward" hypothesis of portal hypertension[9].

Although a common factor of experimental models of cirrhosis and PVL is an increase in the portal venous inflow, these are distinguished by a different degrees of shunting of the portal blood flow to the systemic circulation[10]. Hence, in cirrhotic rats, whether by CCl_4 or by TAA, the extent of portal-systemic shunting varies from 15[10] to 30%,[11] respectively, while PVL rats presented a portal-systemic shunting of 95%[10].

Because in human clinical medicine cirrhosis is frequently associated with collateral circulation[12], we considered that an experimental model that combines PVL, which produces a high degree of collateralization with cirrhosis, would be more useful to study the pathophysiological mechanisms taking place in cirrhotic patients. The present experimental study, in which prehepatic portal hypertension was induced by triple partial portal vein ligation (TPVL) and cirrhosis by the oral administration of TAA, was designed for this purpose.

Materials and Methods

Male Wistar rats, with weights ranging between 242 and 259 g, from the vivarium of the Complutense University of Madrid, were used. The experimental procedures employed in this study are in accordance with the principles and practices of the *1986 Guidelines for the Care and Use of Laboratory Animals* published in the RD 1201/2005 in Spain.

Experimental Design

The animals were divided into four groups:

- Control group (group 1, n=10) in which the animals did not undergo any operative intervention or hepatotoxic drug administration
- TPVL group (group 2, n=9), in which prehepatic portal hypertension by TPVL was carried out
- TAA group (group 3; n=11), in which TAA was administered in the drinking water, according to the method described by Li et al.[2] In brief, the rats received an initial dose of TAA (0.04%) and then the dose was modified according to body weight and changes in response to TAA during the time of cirrhosis induction (16 weeks)
- TPVL+TAA group (group 4; n=9), in which TPVL rats at 12 days of po. evolution were administered TAA according to the method described for group 3

All the animals were killed by anesthetics overdose at 3 months of TAA administration and body (BW), liver (LW), spleen, and testes weights were recorded, along with premortem BW and appropriate weight ratios (absolute and relative to BWs) calculated.

Production of Portal Hypertension

The surgical procedure used to produce portal hypertension by TPVL has been described previously[14]. In brief, while rats were under ketamine hydrochloride (100 mg/kg) and xylacine (12 mg/kg) intramuscular anesthesia, the portal vein was isolated and a triple stenosing ligature was performed in its superior, middle, and inferior portions. The stenoses were calibrated by a simultaneous ligature (4–0 silk) around the portal vein with a 20-gauge needle, which was removed when the ties were ended, thus allowing the partial re-expansion of the portal vein. The midline abdominal incision was closed in two layers with poliglycolic acid and 2–0 silk.

Portosystemic Collateral Circulation Study Method

Portosystemic collateral circulation was studied as follows: First, a midline abdominal incision with a large bilateral subcostal extension was performed and then the areas in which the collateral venous circulation developed, i.e., the splenorenal, gastroesophageal, colorectal, and hepatic hilum, were carefully studied for the presence of increased collateral veins[14].

Portal Vein Pressure Measurement

Splenic pulp pressure, an indirect measurement of portal pressure (PP), was measured by inserting a fluid-filled 20-gauge needle into the splenic parenchyma[15]. The needle was joined to a PE-50 tube and then connected to a pressure recorder (PowerLab 200 ML 201) and to a transducer (Sensonor SN-844) with a Chart V 4.0 computer program (ADI Instruments) and was calibrated before each experiment. The pressure reading was considered satisfactory when a stable recording was produced and respiratory variations were observed. Previous studies have demonstrated the excellent correlation between splenic pulp pressure and PP[16]. The external zero reference point was placed at the midpoint of the animal, 1 cm above the operating table.

Gross Mesenteric Vein Study

Three grades of mesenteric venous vasculopathy were considered: grade 0, normal aspect of the branches of the superior mesenteric vein, without dilation and tortuosity secondary to the superior mesenteric compression for 30 sg; grade 1, dilation and tortuosity of these branches secondary to this maneuver; and grade 2, in which the dilation and tortuosity of the branches of the upper mesenteric vein were spontaneous.

Biochemical Tests

Blood samples (1 ml) were drawn by puncture with a 22-gauge needle of the infrahepatic inferior vena cava. After 15 min of centrifugation at 1,500×*g*, the serum was separated and transferred to sterile polypropylene tubes. The serum was then frozen at −40°C until direct bilirubin, alkaline phosphatase (AP), alanine aminotransferase, aspartate aminotransferase, and gamma-glutamyl transpeptidase (GGT) were assayed by biochemical autoanalyzers.

Histopathological Examinations

Representative samples of the middle hepatic lobe were collected and fixed in phosphate-buffered neutral formalin (10%) and embedded in paraffin. Sections 5 μm-thick were stained with hematoxyline eosine and Masson's trichromic dye. The Knodell histological activity index (HAI) was used to evaluate the severity of the necroinflammatory process and fibrosis[17]. HAI comprises four separate scores, including periportal necrosis with or without bridging necrosis (0–10), intralobular degeneration and focal necrosis (0–4), portal inflammation (0–4), and fibrosis (0–4). The sum of the first three categories' scores was used to grade chronic hepatitis activity as minimal chronic hepatitis (1–3), mild chronic hepatitis (4–8), moderate chronic hepatitis (9–12), or severe chronic hepatitis (13–18), in accordance with the Desmet criteria[18]. Fibrosis score was used to stage chronic hepatitis[17]. Other parameters of hepatic lesion were also studied, such as apoptosis, cholangiofibrosis, and proliferation of the bile duct epithelial cell[11].

Statistical Analysis

The results are expressed as the mean ± the standard deviation. Analysis of variance, the Duncan test, and the Student *t* for unpaired data were used for the statistical comparison of the quantitative variables between the different groups. Chi-square test was used for comparison of the qualitative variables. The results are considered as statistically significant if $p<0.05$.

Results

Body and Organ Weights

Animals from the experimental groups (TPVL, TAA, and TPVL plus TAA) present a statistically significant smaller increase in BW than control animals. This increase is minimum in group 3 (TAA) and is significantly lower than that of groups 2 (TPVL) ($p<0.01$) and 4 (TPVL+TAA) ($p<0.01$) (Table 1).

The ratio of LW to BW (LW/BW×100) undergoes a statistically significant increase in groups 3 (TAA) and 4 (TPVL+TAA) compared to groups 1 (control) and 2 (TPVL). In turn, the LW/BW ratio in group 4 (TPVL+TAA) compared to that in group 3 (TAA) is statistically significantly lower ($p<0.01$) (Table 1). In relation to the weights of the superior (SLW) and inferior hepatic lobes (ILW), the only difference is presented by animals from group 4 (TPVL+TAA) that are statistically significantly lower than in group 3 (SLW; $p<0.05$) and group 2 (ILW; $p<0.01$) (Table 1).

The weight of the spleen as a percentage of BW was significantly increased in groups 3 (TAA) and 4 (TPVL+TAA) in relation to groups 1 and 2 (Table 1). In groups 3 (TAA) and 4 (TPVL+TAA) there was no reduction in testicle weight (Table 1).

Porto-collateral Circulation

The animals from groups 2 (TPVL) and 4 (TPVL+TAA) always develop extrahepatic portosystemic collateral circulation. The most frequent type of collateral circulation is splenorenal circulation, which is presented by all the animals from both groups. Moreover, in group 2 (TPVL), it is associated with paraesophageal circulation in 44.4% ($n=4$) of cases, hemorrhoidal-type collateral circulation (pararectal collateral) in 33.3% ($n=3$) of cases, and both types (paraesophageal and pararectal circulations) in 11.1% (n=1) of cases. For group 4 (TPVL+TAA), the type of collateral circulation associated with splenorenal circulation is paraesophageal circulation in 55.5% ($n=5$) of cases (Fig. 1). On the contrary, animals from group 3 (TAA) do not present any type of extrahepatic portosystemic collateral circulation (Fig. 1).

PP Value

The animals from groups 3 (TAA) and 4 (TPVL+TAA) undergo a statistically significant increase in PP (group 3: 14.56±2.92 mm Hg; group 4: 12.53±1.97 mm Hg) compared to groups 1 (control) ($p<0.001$) and 2 (TPVL) ($p<0.001$) (Fig. 2).

Table 1 BW Increase (BWI), LW, LW to BW, SLW (Left Lateral and Middle Liver Lobes) ILW (Right Lateral and Caudate Liver Lobes), Splenic Weight to BW and Testicular Weight to BW in Control Rats, TPVL, TAA, and TPVL Plus TAA

Group	BWI (g)	LW (g)	LW/BW×100	SLW (g)	ILW (g)	SW/BW× 100 (g)	TW/BW× 100
C group 1 (n=10)	216.56±28.64	10.95±0.83	2.70±0.12	7.26±0.55	3.68±0.39	0.21±0.02	0.86±0.07
TPVL group 2 (n=9)	168.63[a]±11.96	11.25±0.55	2.72±0.17	7.24±0.48	4.01±0.33	0.20±0.03	0.82±0.06
TAA group 3 (n=11)	31.11[a,d]±32.98	11.55±1.12	4.96[a,d]±0.67	7.77±0.96	3.68±0.39	0.36[b,e]±0.09	0.83±0.44
TPVL+TAA group 4 (n=9)	66.64[a,d,g]±23.27	9.96 [c,f,g]±1.46	3.82[b,e,h]±0.48	6.72[i]±1.06	3.24[d]±0.44	0.29[a,f,i]±0.06	1.10[c,f,i]±0.12

Mean±SD
BWI = BW increase, TW = testicular weight, SW = splenic weight, C = control rats
[a] $p<0.01$ statistically significant value in relation to group 1
[b] $p<0.001$ statistically significant value in relation to group 1
[c] $p<0.05$ statistically significant value in relation to group 1
[d] $p<0.01$ statistically significant value in relation to group 2
[e] $p<0.001$ statistically significant value in relation to group 2
[f] $p<0.05$ statistically significant value in relation to group 2
[g] $p<0.01$ statistically significant value in relation to group 3
[h] $p<0.001$ statistically significant value in relation to group 3
[i] $p<0.05$ statistically significant value in relation to group 3

Grade of Mesenteric Venous Vasculopathy

The animals from group 2 (TPVL) show either grade 1 (n=3; 33.3%) or grade 2 (n=6; 66.6%) mesenteric venous vasculopathy. In group 3 (TAA), only 54.5% (n=6) present mesenteric venous vasculopathy, which is grade 1. Finally, in group 4 (TPVL+TAA) all the animals develop mesenteric venous vasculopathy of grade 1.

Liver Function

In animals from groups 3 (TAA) and 4 (TPVL+TAA), routine liver function tests show a statistically significant increase in AP compared to control animals (group 1) ($p<0.001$) and with TPVL (group 2) ($p<0.001$). Also, in group 3 (TAA), there is a moderate increase in blood GGT that is statistically significant ($p<0.05$) compared to control values (group 1). (Table 2).

Morphology and Histopathologic Examination

Numerous macro and micronodules (n=9; 81.8%) were seen on the surface of most of the TAA livers (group 3). However, none of the animals belonging to group 4 (TPVL+TAA) presented either macro or micronodules on the hepatic surface. The animals from groups 1 (control) and 2 (TPVL) did not present significant morphological changes in hepatic tissue. In all cases, the Knodell HAI was zero and fibrosis was not observed (Table 3).

The histopathological findings of groups 3 (TAA) and 4 (TAA+TPVL) are summarized in Table 3. Both groups

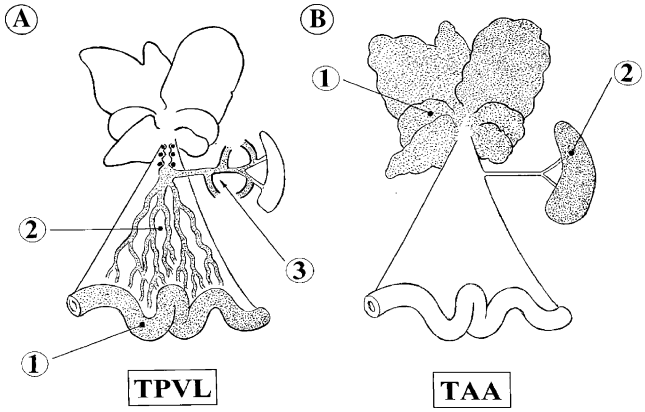

Figure 1 Diagram showing the splanchnic alterations presented by the experimental models secondary to triple stenosing portal ligation (TPVS) and oral administration of TAA. **A** TPVS. In the long term (3 months), TPVS rats manifest hypertensive enteropathy (1), mesenteric venous vasculopathy (2), and portosystemic colateral circulation (3) without portal hypertension. **B** TAA. Animals present hepatosplenomegaly (1 and 2) and portal hypertension.

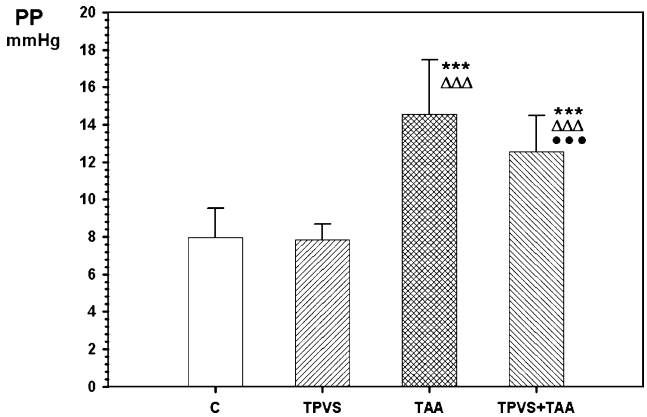

Figure 2 PP (mm Hg) in control (*C*), triple portal vein ligated (*TPVS*), TAA and TPVS plus TAA rats. *Triple asterisks* $p<0.001$: statistically significant value in relation to group 1. *Triple dots* $p<0.001$: statistically significant value in relation to group 3. *Triple triangles* $p<0.001$: statistically significant value in relation to group 2.

Table 2 Direct Bilirrubin, AP, GGT, Aspartatoaminotransferase, and Alanin Aminotransferase in Control Rats, TPVL, TAA, and TPVL Plus TAA

Group	DB mg/dl	AP U/l	GGT U/l	AST U/l	ALT U/l
C (n=10)	0.07±0.02	126.30±33.42	5±2.82	182.40±77.06	23.60±4.90
TPVL (n=9)	0.10±0.49	97.44±31.80[a]	2.67±1.58[a]	64.56±25.11[b]	40.22±28.34[a]
TAA (n=11)	0.09±0.02	370.36±185.42[c,d]	5.36±1.85[f]	134.45±40.44[a,e]	27.18±7.36
TPVL+TAA (n=9)	0.10±0.05	330.22±112.38[c,d]	4.22±2.58[f]	147.67±36.12[d,g]	35.33±6.30

Mean±SD
C=control rats, DB=direct bilirrubin, AST=aspartatoaminotransferase, ALT=aminotransferase, AP=alkaline phosphatase
[a] $p<0.05$ statistically significant value in relation to group 1
[b] $p<0.01$ statistically significant value in relation to group 1
[c] $p<0.001$ statistically significant value in relation to group 1
[d] $p<0.001$ statistically significant value in relation to group 2
[e] $p<0.01$ statistically significant value in relation to group 2
[f] $p<0.05$ statistically significant value in relation to group 2
[g] $p<0.05$ statistically significant value in relation to group 3

presented a statistically significant difference in the grading and staging of chronic hepatitis compared to groups 1 and 2 ($p<0.001$). However, there was no difference between groups 3 and 4.

In group 3, a ductular proliferation was observed in 50% of the animals (statistically significant difference with group 4, $p<0.05$) and cholangitis in 30%. These changes were not observed in any animal from group 4. Finally, a higher degree of apoptosis was observed in group 3 than in group 4 ($p<0.05$) (Table 3, Fig. 3).

Discussion

This study shows that the association of two methods to produce portal hypertension, i.e., partial portal vein ligation by TPVL[14] and oral administration of TAA,[2] permits an experimental model (group 4) to be obtained that associates its pathological characteristics in the long term, such as extrahepatic portosystemic collateral circulation and cirrhosis, respectively.

The extrahepatic portosystemic collateral circulation developed in this new experimental model of portal hypertension is mainly splenorenal and its existence is attributed to portal stenosing ligation (TPVL) because we have not found that the oral administration of TAA (group 3) produces any type of extrahepatic portosystemic collateral circulation. For this reason, it can be considered that the portal systemic shunting described in rats with cirrhosis, whether by CCl_4[10] or by TAA[11], could correspond to the development of intrahepatic portosystemic shunts. More specifically, intrahepatic shunt formation between terminal portal venules and terminal hepatic venules has been described in cirrhotic rats by TAA[19].

Extrahepatic portosystemic collateral circulation that produces TPVL persists in the long term. It has even been described to be maintained at 6 months[14]. However, in this study, although animals with TPVL (group 2) present collateral circulation, this is not associated with portal hypertension (Fig. 1).

In rats with stenosing ligation of the portal vein, the PP increases initially as a consequence of an increased extrahepatic resistance; but, once collaterals have formed, high PP is maintained by an increased splanchnic blood inflow secondary to vasodilatation[8,20]. However, because there is no rise in PP in more advanced stages (3 months), the persistence of extrahepatic portosystemic collateral circulation could be attributed to other mechanisms.

Recently, it has been postulated that the mechanism that determines the formation of portal-system collateral vessels

Table 3 The Knodell Hepatic HAI, Chronic Hepatitis Activity (Minimum, Mild, and Moderate), Chronic Hepatitis Stage, Ductular Proliferation, Apoptosis, and Cholangitis in TAA and TPVL-plus-TAA rats

Group	HAI	CHA			CHS				Ductular proliferation (%)	Apoptosis			Cholangitis (%)
		Min (%)	Mild (%)	Mod (%)	1 (%)	2	3 (%)	4 (%)		0 (%)	+ (%)	++ (%)	
3 (TAA) n=10	8.20±3.91	30	10	60	30	0	40	30	50	0	50	50	30
4 (TPVL+TAA) n=9	6.67±2.78	11.1	44	44	33.3	0	33.3	3.3	26.3 (p=0.013)	11.1	88.9 (p=0.03)	0	0 (p=0.03)

Mean±SD
CHA=chronic hepatitis activity, CHS=chronic hepatitis stage

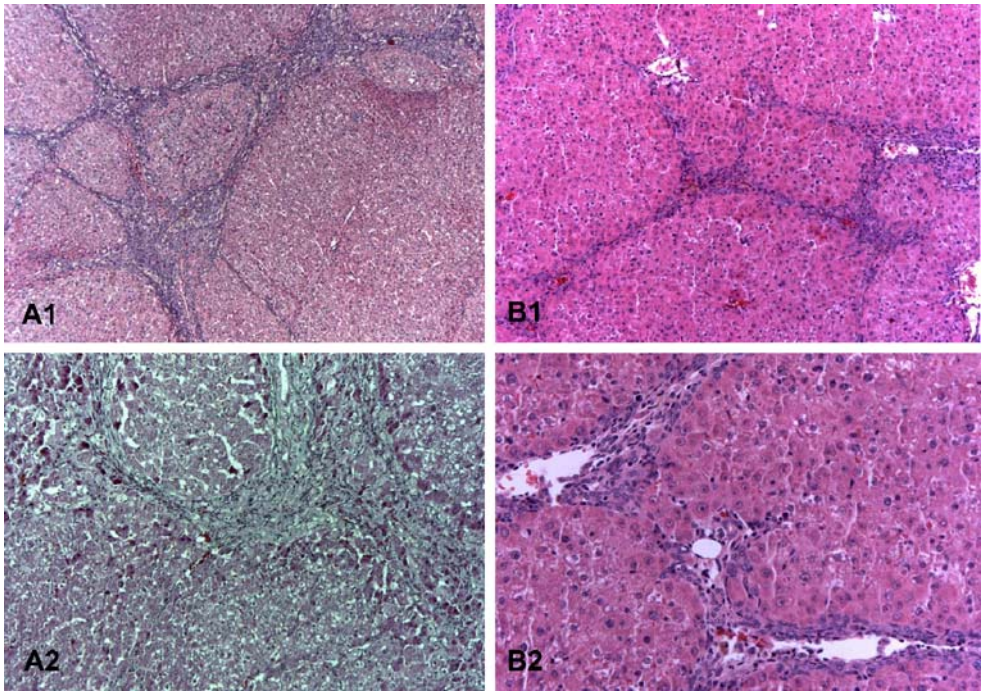

Figure 3 Liver histology. **A**. Liver section showing cirrhosis induced in rats after 16 weeks of TAA treatment. **A1** Hematoxyline eosine (H&E), magnification ×5. **A2** Masson trichromic, magnification ×10. **B** Moderate regression of hepatic cirrhosis in the triple portal vein partial ligation rats treated with TAA. **B1** H&E, magnification ×10. **B2** H&E, magnification ×20.

in partial portal vein-ligated rats is an angiogenic dependent process that can be markedly inhibited by blockading the vascular endothelial growth factor (VEGF) signaling pathway[21,22]. In TPVL rats, angiogenesis also participates in the production of portal hypertensive enteropathy that these animals present, causing an increase in the number, diameter, and area of submucosal small bowel vessels[23]. This process has been attributed to mediators released by the increased mast cell density in the mucosa and submucosa of the small intestine that can induce new blood vessel formation[23,24].

In turn, the TPVL rats also present mesenteric venous vasculopathy; an alteration of the splanchnic venous system characterized by vasodilatation and tortuosity of the branches of the superior mesenteric vein. Although it has been suggested that this macroscopic alteration could be secondary to the existence of splanchnic hyperdynamic circulation[25], it cannot be ruled out that angiogenic mechanisms participate in its production. Similarly, in patients with chronic portal vein hypertension, because the tissue structure of the vascular wall was changed due to long term dilatation, the dilated blood vessels would be hard to recover, even if the effect of vasodilators had been completely eliminated[26].

Therefore, in chronic (3 months) prehepatic portal hypertension (TPVL) in the rat, exacerbated splanchnic angiogenesis could be responsible for both the establishment of intestinal vasculopathy, with a greater increase of the vascular surface in the intestinal submucosa[23]; for the production of mesenteric vasculopathy, with dilatation and tortuosity of the splanchnic venous system[14]; and, finally, for the development of portal-systemic collateral extrahepatic vessels[21]. Therefore, in this case, remodeling of the splanchnic vascular system in chronic prehepatic portal hypertension could be associated with an increase in portal venous inflow, although not with a rise in PP.

In turn, the oral administration of TAA would provide the new experimental model (group 4) with both liver cirrhosis [11,19,27] and an increase in PP[11,28] (Fig. 1). TAA administration to TPVL rats (group 4) produces cirrhosis in spite of the existence of portosystemic collaterals, via which this hepatotoxic agent can bypass the liver. Fibrosis and nodular formation are more prominent in TAA-induced cirrhosis than cirrhosis induced by CCl_4[27]. Angiogenesis plays a known pathogenic role in the production of cirrhosis[29]. Moreover, it has been suggested that the recruitment of mast cells and activated macrophages in the portal area, which are known to be the source of angiogenic substances, could be one of the main factors that governs angiogenesis in alcoholic fibrosis/cirrhosis[30]. In particular, angiogenesis may induce arteriovenous intrahepatic shunting, with the obliteration of small portal veins that also contributes to the induction of a disease-associated portal hypertension[31].

Also, TAA-induced cirrhotic rats presented a splanchnic hyperdynamic circulation with an increase in portal venous inflow and reduced splanchnic arterial resistance[11]. Therefore, portal hypertension is considered to be induced by elevation of the portal venous resistance[11,19] and an increase in portal venous inflow relating to splanchnic hyperdynamic circulation in TAA rats[11].

The changes that characterize prehepatic portal hypertension seem to mainly be a consequence of angiogenesis.

Numerous growth factors (stem cell factor, VEGF, EGF, basic fibroblast growth factor, and platelet-derived growth factor) and cytokines, particularly TNF-α and NO[32,33], have been involved in angiogenesis. In the experimental model of prehepatic portal hypertension by triple stenosing ligation of portal vein, we have shown an increase in hepato-intestinal release of TNF-α, IL-1β, and NO, both in early evolutive (1 and 3 months) and in chronic (15 months) phases[34–36]. Therefore, these mediators could be involved in angiogenesis that occurs in portal hypertensive enteropathy, as well as in the intestinal remodeling process secondary to the chronicity of the splanchnic regional inflammatory response[37].

The angiogenic hyperactivity that occurs in this prehepatic portal hypertension model could be secondary to the portal inflammatory enteropathy that has been proposed to be mediated by mast cells[23,24]. Therefore, it would be angiogenesis and not portal hypertension, nonexistent in this model in the long term, that would be responsible for the development of collateral circulation.

On the contrary, in TAA-cirrhotic rats, hepatic angiogenic hyperactivity would predominate, with the formation of arteriovenous shunts and regeneration nodules[27]. In turn, owing to the nonexistence of mesenteric venous vasculopathy and the collateral vessels in this experimental model, one could suspect that there is no significant intestinal angiogenic or venous splanchnic response. This could be why portosystemic collateral circulation does not develop, in spite of the increased PP (Fig. 2).

Therefore, portal hypertension does not imply the development of collateral vessels provided that intestinal angiogenic and, by extension, mesenteric hyperactivity do not exist. If so, although both alterations are associated with human hepatic cirrhosis, they would result from different etiopathogenic mechanisms. Therefore, the hepatic pathology caused by TPVL could make the liver sensitize the toxic effect of TAA. This would explain why a TPVL-plus-TAA association causes greater hepatic atrophy (Table 1). Hence, in TPVL rats, the administration of TAA (group 4) could cause an increase in PP either by increasing resistance of the liver to portal flow or by an increase in portal venous inflow.

Consequently, in this new experimental model that is achieved by associating partial portal vein ligation (TPVL) and the oral administration of TAA (group 4), different alterations are associated that induce each of the cited production methods of portal hypertension in the rat. Hence, partial portal vein stenosis (TPVL) would produce the mesenteric venous vasculopathy and extrahepatic portosystemic collateral circulation, while the hepatotoxic agent, TAA, would add cirrhosis and portal hypertension. Because these alterations tend to coexist in the cirrhotic patient[6,12], this new experimental model of chronic portal hypertension can be useful to study the pathophysiological mechanisms involved. The coexistence of these alterations in human hepatic cirrhosis, therefore, may mask their different pathophysiological significances.

References

1. Muller A, Machnik F, Zimmermann T, Schubert H. Thioacetamide-induced cirrhosis-like lesions in rats—usefulness and reliability of this animal model. Exp Pathol 1998;34:229–236.
2. Li X, Benjamin IS, Alexander B. Reproducible production of thioacetamide-induced macronodular cirrhosis in the rat with no mortality. J Hepatol 2002;36:488–493.
3. Mas MR, Comer B, Oncu K, Vural SA, Akay C, Tasci I, Ozkomur E, Serdar M, Mas N, Alcigir G, Yener N. The effect of taurine treatment on oxidative stress in experimental liver fibrosis. Hepatol Res 2004;28:207–215.
4. Benoit JN, Barrowman JA, Harper SL, Kvietys PR, Granger DN. Role of humoral factors in the intestinal hyperemia associated with chronic portal hypertension. Am J Physiol (Gastrointest Liver Physiol 10) 1984;247:G486–G493.
5. Lopez-Talavera JC, Merrill WW, Groszmann RJ. Tumor necrosis factor α: A major contributor to the hyperdynamic circulation in prehepatic portal-hypertensive rats. Gastroenterology 1995; 108:761–767.
6. Orloff MJ. Portal hypertension and portocaval shunt. In WW Souba, DW Wilmore Eds., Surgical research vol. 49 London: Academic, 2001, pp. 637–701.
7. Lopez-Talavera JC, Cadelina G, Merrill WW, Groszmann RJ. Role of tumor necrosis factor-alpha in the circulatory abnormalities of cirrhotic rats. Hepatology 1995;22:256A.
8. Sikuler E, Kravetz D, Groszmann RJ. Evolution of portal hypertension and mechanisms involved in its maintenance in a rat model. Am J Physiol (Gastrointest Liver Physiol 11) 1985;248:G618–G625.
9. McMathuna P, Vlavianos P, Westaby D, Williams R. Pathophysiology of portal hypertension. Dig Dis 1992;10(suppl 1):3–15.
10. Chojkier M, Groszmann RJ. Measurement of portal-systemic shunting in the rat by using γ-labeled microspheres. Am J Physiol (Gastrointest Liver Physiol 3) 1981;240:G371–G375.
11. Hori N, Okanoue T, Sawa Y, Mori T, Kashima K. Hemodynamic characterization in experimental liver cirrhosis induced by thioacetamide administration. Dig Dis Sci 1993;38:2195–2202.
12. Sherlock S. The portal venous system and portal hypertension. In S. Sherlock Ed., Diseases of the liver and biliary system 8th ed. London: Blackwell, 1989 pp. 151–207).
13. Monterde G, Rodriguez-Fabian G, Vara E, Lopez L, Arias J, Aller MA, Arias J. Increased plasma levels of corticosterone and prolactin and decreased T_3 and T_4 levels in short-term prehepatic portal hypertension in rats. Dig Dis Sci 2000;45:1865–1871.
14. Dieguez B, Aller MA, Nava MP, Arias JL, Lopez L, Arias J. Chronic portal hypertension in the rat by triple-portal stenosing ligation. J Inves Surg 2002;15:329–336.
15. Castañeda B, Dubernardi-Venon W, Bandi JC, Andreu V, Perez del Pulgar S, Moitinho E, Pizcueta P, Bosch J The role of portal pressure in the severity of bleeding in portal hypertensive rats. Hepatology 2000;31:581–586.
16. Kravetz D, Sikuler E, Groszmann RJ. Splanchnic and systemic hemodynamics in portal hypertensive rats during hemorrhage and blood volume restitution. Gastroenterology 1986;90:1232–1240.
17. Knodell RG, Ishak KG, Black WC, Chen TS, Graig R, Kaplowitz N, et al. Formulation and application of a numerical scoring system for

assessing histological activity in asymptomatic chronic active hepatitis. Hepatology 1981;1:431–435.
18. Desmet VJ, Gerber M, Hoofnagle JH, Manns M, Schever PJ. Classification of chronic hepatitis: diagnosis, grading and staging. Hepatology 1994;19:1513–1519.
19. Nakata M, Nakamura K, Koda Y, Kaminou T, Ugami M, Kaneda K, Yamada R. Hemodynamics in the microvasculature of thioacetamide-induced cirrhotic rat livers. Hepato-Gastroenterol 2002;49: 652–656.
20. Garcia-Tsao G, Wiest R. Gut microflora in the pathogenesis of the complications of cirrhosis. Best Pract Res Clin Gastroenterol 2004 2004;18:353–372.
21. Fernandez M, Vizzutti F, Garcia-Pagan JC, Rodes J, Bosch J. Anti-VEGF receptor-2 monoclonal antibody prevent portal-systemic collateral vessel formation in portal hypertensive mice. Gastroenterology 2004;126:886–894.
22. Moreau R. VEGF-induced angiogenesis drives collateral circulation in portal hypertension. J Hepatol 2005;43:6–8.
23. Diez-Arias JA, Aller MA, Palma MD, Arias JL, Muñiz E, Sanchez M, Arias J. Increased duodenal mucosa infiltration by mast cells in rats with portal hypertension. Dig Surg 2001;18:34–40.
24. Prieto I, Aller MA, Santamaria L, Nava MP, Madero R, Perez-Robledo JP, Arias J. Prehepatic portal hypertension produces increased mast cell density in the small bowel and in mesenteric lymph node in the rat. J Gastroenterol Hepatol 2005;20:1025–1031.
25. Lopez L, Gonzalez-Pardo H, Aller MA, Nava MP, Duran HJ, Arias J, Arias JL. Actividad de citocromo oxidasa esplacnica en ratas con hipertensión portal. Rev Esp Enf Dig Madrid 2001; 93:794–799.
26. Wang JJ, Gao G-W, Gao R-Z, Liu Ch-A, Ding X, Yao Z-X. Effects of tumor necrosis factor, endothelin and nitric oxide on hyperdynamic circulation of rats with acute and chronic portal hypertension. World J Gastroenterol 2004;10:689–693.
27. Dashti H, Jeppsson B, Hägerstrand I, Hultberg B, Srinivas U, Abdulla M, Bengmark S. Thioacetamide-and carbon tetrachloride-induced liver cirrhosis. Eur Surg Res 1989;21:83–91.
28. Chen S, Wang H-T, Yang B, Ou, Q-J. Protective effects of recombinant human growth hormone on cirrhotic rats. World J Gastroenterol 2004;10:2894–2897.
29. Medina J, Arroyo AG, Sanchez-Madrid F, Moreno-Otero R. Angiogenesis in chronic inflammatory liver disease. Hepatology 2004;39:1185–1195.
30. Suneyama TK, Ohba K, Zen Y, Sato Y, Niwa H, Minato H, Nakanuma Y. A comparative histological and morphometric study of vascular changes in idiopathic portal hypertension and alcoholic fibrosis/cirrhosis. Histopathology 2003;43:55–61.
31. Ward NL, Haninec AL, Van Slyke P, Sled JG, Sturk C, Henkelman RM, Wanless IR, Dumont DJ. Angiopoietin-1 causes reversible degradation of the portal microcirculation in mice: Implications for treatment of liver disease. Am J Pathol 165:889–899.
32. Kishore R, Quin G, Luedemann C, Bord E, Hanley A, Silver M, Goukassian D, Losordo DW. The cytoskeletal protein ezrin regulates EC proliferation and angiogenesis via TNF-alpha-induced transcriptional repression of cyclin A. J Clin Invest 2005;115:1785–1796.
33. Ridnour LA, Isenberg JS, Espey MG, Thomas DD, Roberts DD, Wink DA. Nitric oxide regulates angiogenesis through a functional switch involving thrombospondin-1. Proc Natl Acad Sci USA 2005;102:13147–13152.
34. Prieto I, Jimenez F, Aller MA, Nava MP, Vara E, Garcia C, Arias J. Tumor necrosis factor-alpha, interleukin-1 beta and nitric oxide: induction of liver megamitochondria in prehepatic portal hypertensive rats. World J Surg 2005;29:903–908.
35. Aller MA, Vara E, Garcia C, Palma MD, Arias JL, Nava MP, Arias J. Proinflammatory liver and antiinflammatory intestinal mediators involved in portal hypertensive rats. Mediators Inflamm 2005;2:101–111.
36. Palma MD, Aller MA, Vara E, Nava MP, Garcia C, Arias-Diaz J, Balibrea JL, Arias J. Portal hypertension produces an evolutive hepato-intestinal pro- and anti-inflammatory response in the rat. Cytokine 2005;31:213–226.
37. Corcuera MT, Nava MP, Angulo A, Aller MA, Gomez F, Casado I, Alonso MJ, Arias J. Splanchnic remodeling secondary to experimental prehepatic portal hypertension. An Med Inter 2005;22:317–322.

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Rare Localizations of the Hydatid Disease. Experience from a Single Center

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Abstract Hydatid disease is a rare entity that mostly affects the liver and lung, but almost any organ, forming cysts. Although diagnosis is confirmed by serology and imaging studies, a high index of suspicion is required based on the epidemiological background of the patient. However, rare locations of the cysts remain a diagnostic and therapeutic dilemma. Surgical exploration with special attention to avoid parasite spillage is justified in these situations because diagnostic puncture is usually contraindicated. Pericystectomy or fenestration and omentoplasty is strongly recommended, along with the excision of involved organs when feasible.

Keywords Hydatid disease · Echinococcosis

Introduction

Echinococcal disease in humans is a parasitic tapeworm infection caused by a larval stage (the metacestode) of Echinococcus species (*Echinococcus granulosus*, *Echinococcus multilocularis*, *Echinococcus oligarthus*, or *Echinococcus vogelis*). The prevalence of echinococcosis varies considerably but is endemic in the Middle East, Greece, and Africa. The infection can be asymptomatic or severe, causing extensive organ damage and even the death of the patient. Echinococcal cysts usually appear in liver (60%), and lung (30%), but may rarely develop at other sites, including spleen[1], kidney[2], adrenal[3], brain, pericardium, etc. We present our experience with rare sites of hydatidosis, including the diagnostic evaluation and surgical treatment.

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Methods

Between 1990 and 2006, 95 patients with hydatid disease were managed at the 1st Surgical Department of Agia Olga Hospital. Most of our cases localized in the liver; however, echinococcosis was also identified in unusual sites. These atypical locations included the adrenal gland, left kidney, small bowel mesentery, spleen, retroperitoneum with extension in the spinal medulla, gallbladder cyst wall, left thigh, and the biceps branchial muscle (Table 1).

Patients presented with nonspecific symptoms ranging from vague abdominal or back pain to fever spikes and weight loss. In five patients, the disease was completely asymptomatic. A palpable mass was identified in three cases but without tenderness (Table 2). Differential diagnoses involved parasitic and nonparasitic cysts, sarcomas, and rare tumors of mesenchymatic origin.

Diagnosis was confirmed by immunoelectrophoresis and imaging studies. Computed tomography (CT) was applied in all of our patients as the diagnostic tool of choice (Figs. 1, 2, 3, 4, and 5). Daughter cysts with or without cyst wall calcifications in a patient with previous animal exposure were suspicious for hydatid disease. Ultrasound was helpful for further classification according to WHO criteria; however, CT was more precise in identifying the anatomical relations with adjacent structures. Fine needle biopsy (FNA) and magnetic resonance imaging were applied in the diagnostic workup of the patients with biceps

Table 1 Locations of Hydatid Cysts

Location of cysts	Patients
Adrenal gland (right)	3
Biceps brachial muscle	1
Kidney (left) and mesentery	1
Retroperitoneum with daughter cysts in vertebra and spinal medulla	1
Spleen	5
Gallbladder wall	1
Thigh (left)	1

and thigh hydatid cysts to differentiate from other lesions, although FNA is not performed routinely in our center to avoid parasite spread.

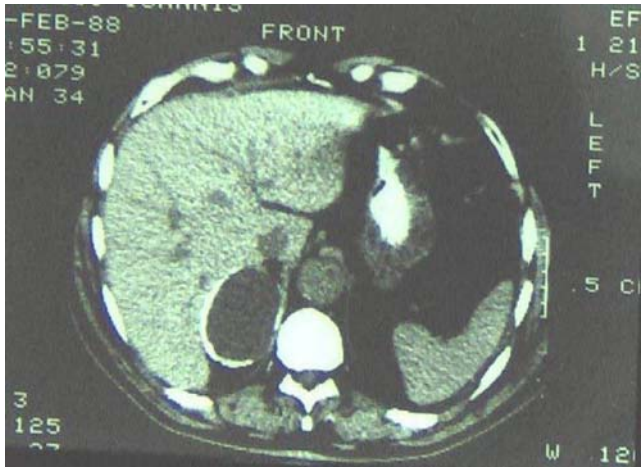

Figure 1 Hydatid cyst of right adrenal gland.

Results

Surgical treatments include resection or fenestration with omentoplasty. Partial nephrectomy, splenectomy, and adrenalectomy were performed whenever cysts invaded these organs. Complete cyst excision (pericystectomy) was our primary goal; however, evacuation of the cyst contents and omentoplasty were applied in the case of hydatid extension to the verebrae. In this particular case, the surgical removal of the affected vertebrae combined with posterior stabilization was also performed, followed by adjuvant chemotherapy. The postoperative hospital stay ranged between 5 and 15 days (median of 7 days). Morbidity was minimal, with a wound infection recorded postoperatively in the case treated with fenestration. Follow up was performed twice a year with CT scan and antibody titers, with one documented recurrence in the patient with hydatidosis of the spine.

Discussion

The most frequent site of hydatid disease is the liver (60%), due to portal vein migration of the parasites. Other organs affected, in order of frequency, are lungs (30%), peritoneum (4%), biliary tract (4%), and pleural cavity (1%). The

Table 2 Clinical Symptomatology in Patients with Hydatid Disease in Nontypical Sites

Signs and symptoms	Patients
Fever spikes	2
Urticaria	1
Abdominal or back pain	3
Palpable mass	2

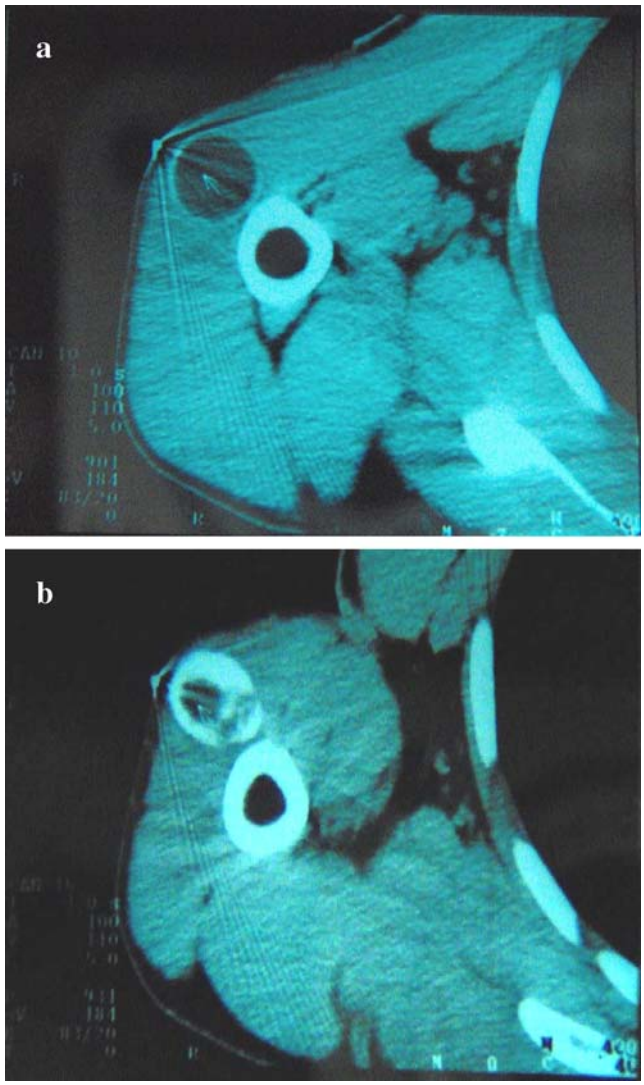

Figure 2 **a** Hydatid disease of biceps brachial muscle, **b** hydatid disease of biceps brachial muscle.

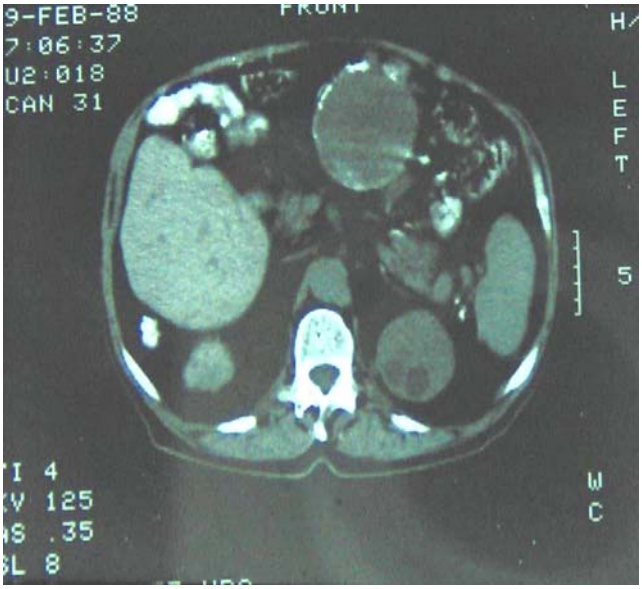

Figure 3 Hydatid cyst of left kidney and mesentery. A calcified rim is demonstrated.

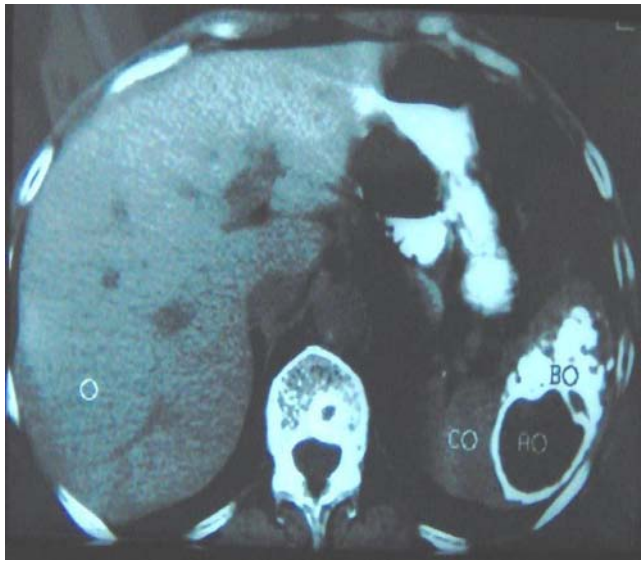

Figure 5 Hydatid disease of the spleen.

prevalence of echinococcosis varies considerably but is endemic in the Middle East and Africa.

In our series, we selected rare locations of hydatid from a single tertiary care center specialized on hepatobiliary diseases. Diagnostic and therapeutic strategies are also summarized from the surgeon perspective. Although portal blood stream remains the main pathway of parasite spread, normally existing porto-caval shunts, lymphatic invasion by the worm, and retrograde migration from vena cava to subclavian vein have been documented and explain some of these rare sites. Diagnosis is based on serology and imaging studies[4]. Plain abdominal radiography can sometimes demonstrate a fluid-type shadow, although the best indicator for hydatid origin of the lesion is the presence of calcifications. Ultrasonography can define the structure of the liver cyst and the number, location, and presence of daughter cysts. CT and magnetic resonance imaging are considered to be very sensitive and accurate because even cysts smaller than 1 cm can be detected and virtually all organs explored. Garbi[5] and WHO modified classification are widely accepted and used by our team routinely. Confirmation of diagnosis should be obtained with immunoelectrophoresis, Western blotting and coelectrosyneresis. ELISA, Western blot, and polymerase chain reaction should be used for other ectopic locations or calcified cysts. It is worthwhile mentioning that specific antibodies increase 1 month after surgery and decrease slowly thereafter. Persistence of high specific antibodies or a secondary increase of titer indicates relapse.

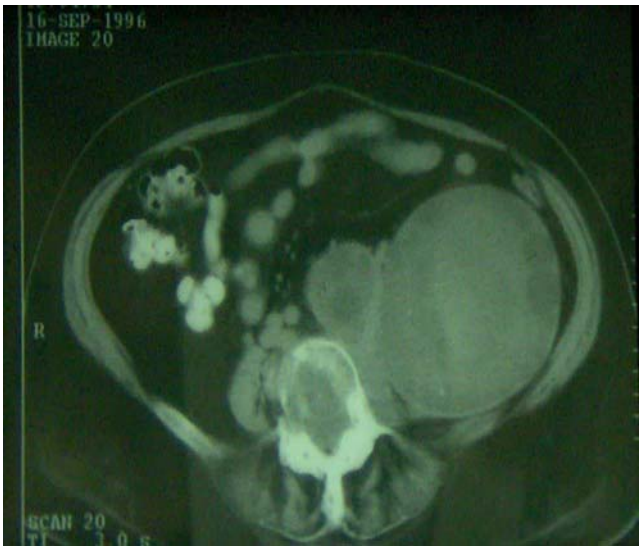

Figure 4 Hydatid disease of retroperitoneum with daughter cysts invading the vertebra and spinal medulla.

The treatment of choice is surgery, with special attention to avoid any spread of hydatid with subsequent secondary echinococcosis. There are many surgical procedures for the management of hydatid cysts. Indications for surgery include large cysts with multiple daughter cysts; superficial location amenable to rupture; cysts exerting pressure on adjacent organs; and cysts in ectopic locations such as lung, brain, bones, spleen, kidneys[4], etc. Although pericystectomy or fenestration and omentoplasty remain popular, new treatment modalities have emerged. Laparoscopic, minimally invasive surgical techniques for certain indications yield good results in expert hands. The proper procedure is selected by taking into consideration the cyst location and Garbi classification. In our series of unusual and rare hydatid cyst locations, we prefer pericystectomy with excision of involved organs unless this is not feasible, as in the case of spinal cord cyst extension. In this particular

case, fenestration with omentoplasty was applied successfully, along with the surgical removal of the affected vertebrae. Partial nephrectomy or splenectomy was performed in this rare group of patients (renal or spleen hydatidosis) to eradicate the disease, avoiding secondary hydatid spread. During abdominal exploration, the abdomen was carefully packed with pads immersed in hypertonic saline solution to avoid peritoneal soilage and contamination from the surgical manipulations. Antiscolecidal agents were infused after evacuation of the cyst in the emptied cavity. Albendazole was also administrated 1 month before surgery according to WHO guidelines to further decrease recurrence. In the follow up period, recurrence was noted only when the parasite invaded the spine, in accordance with the literature.[6,7]

Conclusion

From our experience, echinococcosis can appear at any site of the human body, and so should always be considered in the differential diagnosis of cystic space-occupying lesions or unidentified tumor formations in patients from endemic countries.

References

1. Safioleas M, Misiakos E, Manti C. Surgical treatment for splenic hydatidosis. World J Surg 1997;21(4):374–377.
2. Yildirim M, Erkan N, Vardar E. Hydatid cysts with unusual localizations: diagnostic and treatment dilemmas for surgeons. Ann Trop Med Parasitol 2006;100:137–142.
3. Akcay MN, Akcay G, Balik A, Boyuk A. Hydatid cysts of the adrenal gland: review of nine patients. World J Surg 2004;28:97–99.
4. Dervenis C, Delis S, Avgerinos C, Madariaga J, Milicevic M. Changing concepts in the management of liver hydatid disease. J Gastrointest Surg 2005;9(6):869–877.
5. Gharbi HA, Hassine W, Brauner MW, Dupuch K. Ultrasound examination of the hydatid liver. Radiology 1981;139:459–463.
6. Awasthy N, Chand K. Primary hydatid disease of the spine: an unusual case. Br J Neurosurg 2005;19(5):425–427.
7. Sapkas GS, Machinis TG, Chloros GD, Fountas KN, Themistocleous GS, Vrettakos G. Spinal hydatid disease rare but existent pathological entity: Case report and review of the literature. South Med J 2006;99(2):178–183.

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Larger Amounts of Nitrite and Nitrate-reducing Bacteria in Megaesophagus of Chagas' Disease than in Controls

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Abstract In the megaesophagus of Chagas' disease, chronic esophagitis is caused by stasis of swallowed food and saliva. In this environment, the overgrowth of aerobic and anaerobic bacteria, including nitrate-reducing bacteria, is observed. The reduction of nitrate into nitrite by the action of these bacteria has been associated with the formation of volatile nitrosamines in different situations of gastric bacterial overgrowth. We have hypothesized that this phenomenon could occur in the esophageal lumen of patients with megaesophagus. To evaluate the concentration of nitrite, the presence of volatile nitrosamines and the concentration of nitrate-reducing bacteria in the esophageal lumen of patients with nonadvanced megaesophagus of Chagas' disease and in a group of patients without esophageal disease. Fifteen patients with nonadvanced megaesophagus [megaesophagus group (MG)] and 15 patients without any esophageal disease [control group (CG)] were studied. Saliva samples were taken for nitrate and nitrite quantitative determination and esophageal stasis liquid samples were taken for nitrate and nitrite quantitative determination, volatile nitrosamines qualitative determination and reductive bacteria quantitative determination. MG and CG were equivalent in nitrate and nitrite saliva concentration and in nitrate esophageal concentration. Significant difference was found in nitrite ($p=0.003$) and reductive bacteria concentration ($p<0.0001$), both higher in MG. Volatile nitrosamines were identified in three MG patients and in none of the CG patients, but this was not significant ($p=0.113$). There is a higher concentration of reductive bacteria in MG, responsible for the rise in nitrite concentration at the esophageal lumen and, eventually, for the formation of volatile nitrosamines.

Keywords Chagas' disease · Chronic esophagitis · Nitrate-reducing bacteria

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Introduction

Patients with megaesophagus of Chagas' disease have an elevated risk of developing squamous cell carcinoma of the esophagus.[1–3] The development of such disease may be related to chronic inflammation of the esophageal mucosa due to the stasis of swallowed content in the esophageal lumen.[4–6] A periodic surveillance for early cancer detection in these patients is routinely performed through upper gastrointestinal tract endoscopy and the Lugol's dye test.[7] In the dilated esophageal lumen, overgrowth of aerobic and anaerobic oral bacteria, swallowed with saliva, is observed.[8] Some species of this microbiota have nitrate-reducing activity; therefore, they are able to transform nitrate into nitrite. The latter can react, in certain conditions,

with amines originating in food to form volatile nitrosamines,[9] including some that are proven esophageal carcinogens.[10,11] This phenomenon has already been described in the stomach of patients with different conditions associated with reduced gastric acidity and, as a consequence, gastric bacterial overgrowth.[12,13] To better clarify the mechanism of carcinogenesis in these patients with Chagas' disease, we have hypothesized and planned to investigate if these substances and bacteria were present, and also if these reactions could be occurring in the esophageal lumen of patients with megaesophagus.

Objective

The aim of this study was to evaluate the concentration of nitrite, the presence of volatile nitrosamines, and the concentration of nitrate-reducing bacteria in the esophageal lumen of patients with nonadvanced megaesophagus of Chagas' disease and in a group of patients without esophageal disease.

Material and Methods

Thirty patients were enrolled in the period between August 2003 and September 2004 in the Hospital das Clínicas—University of São Paulo School of Medicine, Department of Gastroenterology, Surgical Division. Their age ranged from 19 to 74 years. Eighteen (60%) were male and 12 (40%) were female. Fifteen patients had nonadvanced megaesophagus of Chagas' disease defined by sorologic test (Elisa—bioMerieux Brasil SA®) and radiologic and manometric criteria,[14] and were classified as the megaesophagus group (MG). Nonadvanced megaesophagus was defined as dilated esophagus with less of 10 cm of diameter in the contrast-enhanced x-ray films and manometric examination demonstrating achalasia of the inferior esophageal sphincter and aperistalsis of the esophageal body but not athonia.[7] Fifteen patients with no esophageal disease defined by clinical and endoscopic criteria were called the control group (CG). The clinical criteria were no complaints of disphagia, odinophagia, regurgitation, or typical symptoms of gastroesophageal reflux disease and negative serological test for Chagas' disease. The endoscopic criteria were macroscopic normal esophagus in the upper endoscopy (biopsies were not performed). Manometry was not performed in the CG. Patients with diabetes, smokers, frequent alcohol drinkers, those with a history of any kind of cancer, and those who had used antibiotics in the last 3 months were excluded from both groups. All the patients gave signed informed consent approved by the Ethics Committee of the Hospital.

Sample Collection

Saliva and esophageal stasis liquid were collected in the morning after 8 to 12 h of fasting. Saliva samples were obtained after stimulation of chewing movements and were taken for quantitative determination of nitrate and nitrite. Esophageal stasis liquid samples were taken using a sterilized no. 14 Levine tube, which was passed through a sterilized no. 7.5 orotracheal tube. These tubes were introduced through the mouth into the inferior third of the esophagus with the Levine catheter placed inside the endotracheal tube to avoid contamination with microorganisms in the oropharynx.[15] The catheter was then pushed until it reached the esophageal lumen, and the stasis liquid was then aspirated with a 20-ml syringe connected to its extremity. Total static liquid at the esophageal lumen of Chagas' patients was not measured. In the CG, the collection was performed by washing the esophageal lumen with 10 ml of sterile saline solution, after which the contents were aspirated. The dilution was considered for concentration measure. The collected material was prepared for quantitative determination of nitrate and nitrite, qualitative determination of volatile nitrosamines, and quantitative determination of nitrate-reducing bacteria.

Nitrate, Nitrite, and Nitrosamine Determination

Nitrate and nitrite were determined by chemiluminescent assay (Nitric oxide analyzer, Mod 280, Sievers®). Nitrosamine concentrations were measured by means of gas chromatography and mass spectrometry (QP 5050 A, Shimatzu®), using the following five nitrosamines as standards: *N*-nitrosodimethylamine, *N*-nitrosoetilmethylamine, *N*-nitrosodiethylamine, *N*-nitrosomorpholine, and *N*-nitrosopiperidine (Sigma-Aldrich®—Fig. 1).

Determination of Nitrate-reducing Bacteria

The separated bacterial colonies for the microbiologic study were diluted, seeded in specific culture media, and incubated under aerobic and anaerobic conditions. Representative colonies were chopped and added to indol-nitrate juice to test for nitrate formation. For comparison, only nitrate-reducing colonies present in the MG or CG were considered.

Statistical Analysis

In the analysis of the differences between the MG and CG salivary and esophageal nitrate and nitrite levels, the Mann–Whitney test was employed. For the analysis of differences between salivary and esophageal nitrate and nitrite levels within the same group, the Wilcoxon test was

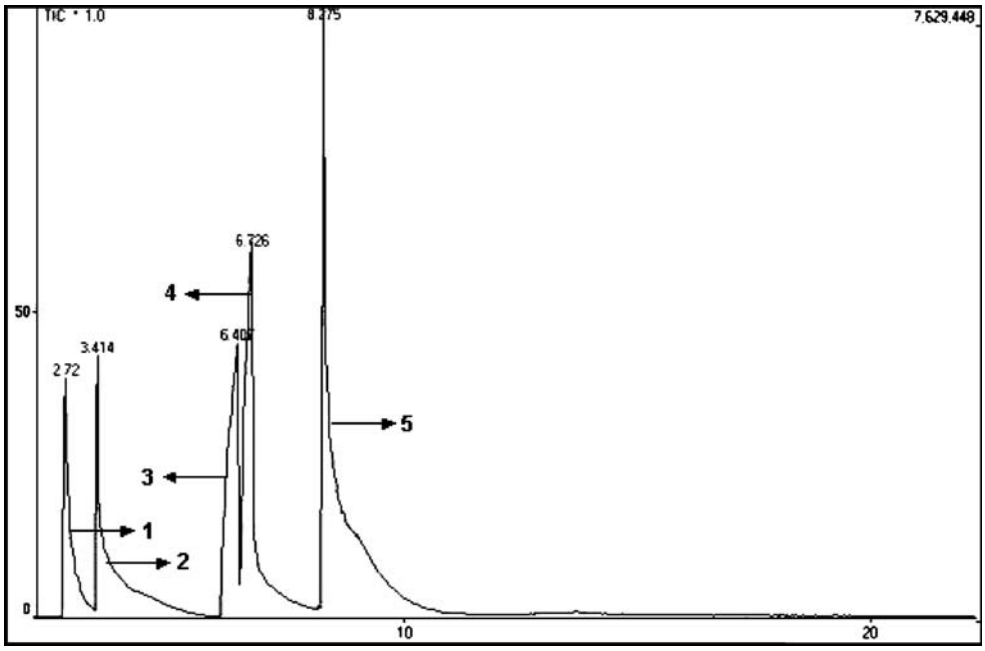

Figure 1 Standard curve of nitrosamines. *1 N*-nitrosodimethylamine; *2 N*-nitrosoetilmethylamine; *3 N*-nitrosodiethylamine; *4 N*-nitrosopiperidine; *5 N*-nitrosomorpholine.

selected. For the association analysis among the groups and the presence or absence of nitrosamines or nitrate-reducing bacteria, the chi-square test was chosen.

Results

Nitrate-reducing bacteria found in MG and CG are presented in Table 1. The MG and CG had equivalent concentrations of nitrate and nitrite in their saliva (Table 2). Their esophageal nitrate concentrations were also equivalent; however, significant differences were found in the concentrations of nitrite ($p=0.003$) and nitrate-reducing bacteria ($p<0.0001$), both being higher in the MG (Table 3). Volatile nitrosamines were identified in three MG patients and in none of the CG patients. Nevertheless, this difference was not significant (Table 3).

Discussion

One of the purposes of this investigation was to test the hypothesis that individuals with megaesophagus have significantly greater amounts of esophageal nitrate-reducing bacteria than are found in normal controls. Specific tests able to identify the bacterial-reducing activity were applied, and a larger quantity of reducing species was found in the MG than in the CG. The activity of this nitrate-reducing flora was suspected by the finding of spontaneous modification of nitrogenous compounds in the esophageal lumen and the formation of a higher concentration of nitrite than that expected in a normal situation, with nitrosamine formation as the eventual consequence.

Nitrate and nitrite concentrations were measured in the saliva and esophagus of the megaesophagus patients and patients without esophageal disease, and the presence of nitrosamines in the esophageal lumen was investigated in both groups. This study model has been applied previously, through the same evaluation method, in patients with several gastric diseases.[12,13]

The sources of nitrate to the humen body are food (mainly vegetables) and water. The quantity of nitrate ingested by water varies with the concentration of nitrate in the water of different places. In São Paulo, were all the patients that participated in the study live, the concentration of nitrate in water is 10 mg/dl. The ingested nitrate is almost completely absorbed in the upper gastrintestinal tract and 25 to 30% of the circulating nitrate is secreted in saliva. Higher concentrations of nitrate in saliva are observed between 1 and 2 h after drinking a vegetable juice.[16] In different studies, large differences in nitrate saliva concentrations between patients are observed, probably due to different amounts of ingested nitrate.[17] Nitrite is formed by the reduction of ingested nitrate by nitrate-reducing bacteria. This physiologic phenomenon responds to 80% of nitrite body exposure and occurs mainly in the oral cavity and less in the stomach in normal situations.[18]

Samples were collected from patients after an approximately 12-h fast, a required condition for a safe collection of material from the esophagus. In this situation, the nitrate concentration in the saliva is probably at its lowest level of daily variation. Nevertheless, the purpose was to let the MG and CG nitrate saliva concentrations reach some

Table 1 Prevalence (%) and Concentration (CFU/ml) of Nitrate-reducing Bacteria in MG (n=15) and CG (n=15)

	Prevalence (%) (megaesophagus)	Concentration (megaesophagus)	Prevalence (%) (control)	Concentration (control)
Lactobacillus sp.	66	10^1–10^7	13	10^1–10^2
Peptococcus sp.	40	10^1–10^5	6.6	10^1
Veillonella sp.	80	10^1–10^7	0	0
Enterococcus sp.	40	10^2–10^7	0	0
Fusobacterium sp.	6.6	10^3	0	0

CFM/ml = colony-forming units per milliliter

equivalence, so that this parameter would not interfere in the next analysis.

Our results of the average salivary nitrate concentration in the CG and MG were both compatible with data from other studies.[17,18] There was no significant difference between the two groups. However, in both groups, the relative nitrite concentration compared to nitrate is a little higher than that reported in other studies in which similar methodology was employed.[17,18] The average high nitrite concentration in both groups was affected by a few subjects who presented with very high concentrations (up to 245 μmol/l in the CG and 343 μmol/l in the MG). A possible explanation for this phenomenon could be the poor conservation of teeth and deficient oral condition of some of the subjects in both groups, resulting in an overgrowth of nitrate-reducing bacteria and, consequently, greater reducing activity in the saliva. This condition was characteristic of some of the people studied that belonged to a low socioeconomic level. Regardless, it can be said that the salivary nitrate and nitrite levels were equivalent in the two groups. The equivalent levels of nitrite in saliva indicate that there is a similar concentration of nitrate-reducing bacteria in the saliva of both groups and that this concentration does not account for the differences found in the esophageal lumen.

Although the difference between the MG and CG in esophageal nitrate concentrations was not significant, there seems to be proportionally more nitrate in the MG. We can suppose that, once there is stasis liquid in the megaesophagus (and not in the normal esophagus), the total nitrate quantity present in the megaesophagus and available for reduction to nitrite is much larger than in the normal esophagus.

The difference between the CG and MG nitrite concentrations was significant, suggesting that patients with megaesophagus have a tendency to have higher nitrate reduction activity than those without esophageal disease. The relative concentration of nitrate/nitrite in the saliva in the CG group was 1.6/1, and in the MG group, it was 1.9/1. In the esophagus, the relative concentration in CG was 4.6/1, and in MG it was 2.1/1, showing that, in MG, there are similar conditions for nitrate reduction as it is observed in saliva but not in the esophagus of CG.

The present study found nitrosamines in the esophageal stasis liquid of three MG patients (*N*-nitrosomorpholine in two cases and *N*-nitrosodimetilamine in one case), and in none of the CG. Although not statistically significant, the event sequence (a nitrate-reducing microbiota, leading to an increase in nitrite concentration, leading in turn to nitrosamine formation) suggests that all the local conditions required for the nitrosation reaction are present in the megaesophagus. No correlation between manometric findings and concentration of bacteria or nitrite at the

Table 2 Concentration (μM) of Nitrate (NO_3) and Nitrite (NO_2) in Saliva

	CG	MG	p
NO_3 (μM)	32.2–267.16 (mean=118.87)	34.9–487.0 (mean=166.34)	0.443
NO_2 (μM)	9.90–245.78 (mean=74.93)	11.48–343.22 (mean=87.24)	0.290

Table 3 Concentration (μM) of Nitrate (NO_3) and Nitrite (NO_2) in Esophagus and the Presence of Nitrosamines and Nitrate-reducing Bacteria in the Esophageal Lumen (%)

	CG	MG	p
NO_3 (μM)	2.83–102.0 (mean=30.85)	2.25–314.0 (mean=90.96)	0.101
NO_2 (μM)	0.05–27.0 (mean=6.72)	0.81–115.06 (mean=42.77)	0.003
Nitrosamines (%)[a]	0	20	0.113[b]
Nitrate-reducing bacteria (%)	13.3	100	<0.0001[b]

[a] Nitrosamines found: *N*-nitrosomorpholine in two cases and *N*-nitrosodimetilamine in one case

[b] χ^2 and Fisher tests

esophageal lumen was observed. However, higher concentrations could have been seen if grade IV or advanced[7,14] megaesophagus cases had been included.

Conclusions

We conclude that, in the megaesophagus of Chagas' disease, there is a higher concentration of nitrate-reducing bacteria, and that this can be the causative factor for an increase in nitrite concentration in the esophageal lumen and, eventually, the formation of volatile nitrosamines. Therefore, it is possible to hypothesize that stasis might establish the condition to lumen production of carcinogens.

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References

1. Carter R, Brewer LA. Achalasia and esophageal carcinoma: Studies in the early diagnosis for improved surgical management. Am J Surg 1975;130:114–120.
2. Chuong JJ, Dubovik S, McCallum RW. Achalasia as a risk factor for esophageal carcinoma. Dig Dis Sci 1984;29:1105–1108.
3. Meijssen MAC, Tilanus HW, Van Blankenstein M, Hop WCJ, Ong GL. Achalasia complicated by esophageal squamous cell carcinoma: A prospective study in 195 patients. Gut 1992;33:155–158.
4. Peracchia A, Segalin A, Bardini R, Ruol A, Bonavina L. Esophageal carcinoma and achalasia: Prevalence, incidence and results of treatment. Hepato-gastroenterol 1991;38:514–516.
5. Loviscek LF, Cenoz MC, Badaloni AE, Agarinakazato O. Early cancer in achalasia. Dis Esophagus 1998;11:239–247.
6. Sandler RS, Nyren O, Ekbom A, Eisen GM, Yuen J, Josefsson S. The risk of esophageal cancer in patients with achalasia. A population-based study. JAMA 1995;274:1359–1362.
7. Yamamuro EM, Cecconello I, Iriya K, Tomishigue T, Oliveira MA, Pinotti HW. Lugol dye endoscopy for analysis of esophageal mucosa in achalasia. Hepato-gastroenterol 1999;46(27):1687–1691.
8. Pajecki D, Zilberstein B, Santos MAA, Ubriaco JÁ, Quintanilha AG, Cecconello I, Gama-Rodrigues JJ. Megaesophagus microbiota: A qualitative and quantitative analysis. J Gastrointest Surg 2002;6:723–729.
9. Dallinga JW, Pachen DMFA, Lousberg AHP, Van Geel JAM, Houben GMP, Stockbrugger RW, Van Maanen JMS, Kleijans JCS. Volatile N-nitrosamines in gastric juice of patients with various conditions of the gastrointestinal tract determined by gas chromatography-mass spectrometry and nitrite levels. Cancer Lett 1998;124;119–125.
10. Ribeiro U Jr, Posner MC, Safatle-Ribeiro AV. Risk factors for squamous cell carcinoma of the esophagus. Br J Surg 1996;3:1174–1185.
11. Soner GD, Gupt A. Etilogy and chemoprevention of esophageal squamous cell carcinoma. Carcinogenesis 2001;22:1737–1746.
12. Verdu E, Viani F, Amstrong D, Fraser R, Siergrist HH, Pignatelli B, Fried M. Effect of omeprazole on intragastric bacterial counts, nitrates, nitrites, and N-nitroso compounds. Gut 1994;35:455–460.
13. Vermeer ITM, Danielle MFA, Dallinga JW, Kleijans JCS, Van Maanen JMS. Intragastric volatile N-nitrosamines, nitrite, pH and *Helicobacter pylori* during long-term treatment with Omeprazole. Gastroenterology 2001;121:517–525.
14. Rezende JM, Lauar KM, Oliveira AR. Aspectos clínicos e radiológicos da aperistalse do esôfago. Rev Bras Gastroenterol 1960;12:247–262.
15. Gagliardi D, Makihara S, Corsi PR, de Toledo Viana A, Wiczer MVFS, Nakakubo S, Mímica LMJ. Microbial flora of the normal esophagus. Dis Esophagus 1998;11:248–250.
16. Van Maanen JMS, Welle IJ, Hageman G, Dallinga JW, Mertens PLJM, Kleijans JCS. Nitrate contamination of drinking water: Relationship with HPRT varian frequency in linphocyte DNA and urinary excretion of N-nitrosamines. Environ Health Perspect 1996a;104:521–528.
17. Van Maanen JMS, Van Geel AA, Kleinjans JC. Modulation of nitrate–nitrite conversion in the oral cavity. Cancer Detec Prev 1996;20:590–596.
18. Van Maanen JM, Pachen DM, Dallinga JW, Kleijans JC. Formation of nitrosamines during consumption of nitrate- and amine-rich foods, and the influence of the use of mouthwashes. Cancer Detec Prev 1998;22:204–212.

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Percutaneous Drainge and Ileocolectomy for Spontaneous Intraabdominal Abscess in Crohn's Disease

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Abstract
Background Historical studies have shown that percutaneous drainage alone for intraabdominal abscess secondary to Crohn's disease is successful in avoiding surgery in only approximately 50% of patients. Failure, however, can require urgent surgery and is then associated with increased morbidity, extended hospital stays, and increased risk for stoma creation. Because of this, our current protocol is initial percutaneous drainage of the abscess, 5–7 days of broad spectrum IV antibiotics with simultaneous high-dose steroids and hyperalimentation, followed by planned one stage resection with primary anastomosis. The aim of the present study was to evaluate the success of this protocol with regard to length of stay, complications associated with the protocol, and its ability to avoid stoma creation.
Methods A retrospective chart review was performed for all Crohn's disease patients with intraabdominal abscess who underwent the above protocol from 1992 to the present.
Results Nineteen patients (11 male) were identified. Sixteen underwent ileocolectomy with primary anastomosis while only three patients required an upstream diverting ileostomy in addition to resection due to incompletely drained abscesses. The mean length of hospital stay was 13.9±0.6 days including 6.4±0.4 postoperative days. Four patients had post-op complications that did not require surgery (two self-limited anastomotic bleeds, one wound infection, and one pelvic abscess treated with a percutaneous drain). One patient needed reoperation for a small bowel obstruction.
Conclusions Crohn's disease patients with intraabdominal abscess can safely undergo planned resection with primary anastomosis if initially treated with successful percutaneous drainage and aggressive antibiotic and steroid management. Such a protocol provides a standard of care against which nonsurgical management can be compared and judged.

Keywords Crohn's disease · Ileocolectomy · Intraabdominal abscess · Percutaneous drainage

Introduction

Crohn's disease (CD) is an immune-mediated illness that primarily affects the gastrointestinal tract. The inflammatory process in the intestine is a full-thickness injury that can lead to significant complications such as fistulae, abscesses, and phlegmons from perforations of the intestine. The organs most commonly involved in fistula formation are the perianal skin, bladder, colon, small bowel, vagina, and abdominal wall. Intraabdominal fistulae are commonly treated with resection of the diseased intestine and simple closure of the fistula in the involved but typically undiseased organ.

Intraabdominal abscesses occur in 7–28% of patients with CD.[1,2] The optimal treatment regimen for intra-abdominal abscess in CD is unclear. Medical management without abscess drainage is usually ineffective. Historical studies have shown that percutaneous drainage alone for intraabdominal abscess is successful in only about 50% of patients.[3–11] Failure of percutaneous drainage requiring unplanned surgery is associated with increased morbidity

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and extended hospital stays averaging greater than 30 days.[10,12] Clearly, the management of the typically immunosuppressed Crohn's disease patient with an intra-abdominal abscess is a difficult life-threatening problem that can understandably result in a complicated and prolonged hospital course. Operating on such patients can frequently require stoma creation and can be associated with significant complication. Due to the perceived lack of consensus in the surgical and medical literature on how to treat such patients, we instituted a protocol for treatment that included initial computed tomography (CT) scan or ultrasound (US)-guided percutaneous drainage, 5–7 days of broad spectrum intravenous (IV) antibiotics, high-dose IV steroids and hyperalimentation, followed by planned one-stage resection with primary anastomosis during the same hospital admission. Our goal behind this protocol was to minimize sepsis, maintain nutritional status, treat the underlying Crohn's disease with aggressive immunosuppressive therapy, while preparing the patient for definitive surgery to resect the diseased segment of bowel in one stage, so avoiding a stoma. This study reviews our experience treating patients with this protocol.

Methods

Patients A retrospective chart review was performed for all patients with ileocolonic CD patients and intraabdominal abscesses from 1992 to the present. An abscess was defined as a ring enhancing fluid collection with or without air identified on CT scan. Patients who presented with intra-abdominal abscess as a postoperative complication were excluded. All patients who underwent the described protocol were selected for review. Data abstracted from the chart included patient characteristics, treatment course, length of hospital stay, complete blood count (CBC), length of bowel resected, and complications. The study was approved by the Milton S. Hershey Medical Center Institutional Review Board.

Treatment All patients were admitted to the hospital and started on high-dose IV steroids (the equivalent of 300 mg hydrocortisone per day) and broad spectrum antibiotics: ampicillin, gentamicin, and metronidazole (13 patients), vancomycin, gentamicin, and metronidazole (three patients), or Unasyn, gentamicin, and metronidazole (two patients). The patients underwent an abdominal CT scan with IV and oral contrast. Rectal contrast was used as necessary. After identification of an abscess, the patients underwent either a CT-guided or US-guided drainage of the abscess cavity. CT vs US as a modality for drainage was chosen at the radiologist's discretion. Cavities were either aspirated dry (seven patients), or a drain was left in place (12 patients). Whether or not to leave a drain was a determination made on a case by case basis by the radiologist based on the size of the cavity.

Patients were initially kept nothing per orem (NPO) and on total parental nutrition (TPN) to minimize nutritional deficit, but as they clinically improved, they were allowed an oral elemental diet as tolerated in addition to the TPN. The patients underwent ileocolic resection with the intent to perform a primary anastomosis and avoid an ileostomy after at least 5 days of treatment with antibiotics and steroids. Any patient who clinically deteriorated while on this protocol was taken to surgery urgently.

Results

Patient Characteristics

Nineteen consecutive patients (11 male) with spontaneous intraabdominal abscesses from CD who underwent the described protocol were identified. Only patients who underwent percutaneous drainage and subsequent surgery on the same admission were included. Since the initiation of this protocol, all patients have been treated in this manner. The patients presented here are sequential. Patient characteristics, admission medications, and abscess location are shown in Table 1.

Table 1 Patient Characteristics

Characteristics	Numbers/Values
Total patients	19
Sex (M/F)	11/8
Mean age	31.2±1.8 years[a]
Duration of disease	7.3±1.8 years[a]
Prior ileocolectomy	3 (16%)
Meds on admission	
None	4 (21%)
Steroids	13 (58%)
5-ASA	9 (47%)
6-MP/Immuran	1 (5%)
Infliximab	1 (5%)
Antibiotics	9 (47%)
Abscess location	
Right lower quadrant	9 (47%)
Right psoas muscle	1 (5%)
Abdominal wall	1 (5%)
Pelvis	4 (21%)
Intraabdominal	4 (21%)

m Male, *f* female, *5-ASA* 5-aminosalicylic acid, *6-MP* 6-mercaptopurine
[a] Mean ± standard error

Hospital Course

Pain was the most common presenting symptom seen in 18 of 19 patients. Patients also complained of elevated temperature (four patients), increased bowel movements (four patients), pyuria, nausea, and vomiting, or palpable mass, one patient each. Of 19 patients, 16 (84%) had an elevated white blood cell count on admission. The mean white blood cell (WBC) for the entire group was 13.8±0.8, range, 8.5–19.5. Of 19 patients, seven (37%) had an elevated temperature on or within 12 hours of admission. Three patients had temperatures of 39°C or greater. Mean temperature for the group was 37.6±0.2, range, 35.8–39.5.

All patients were treated with broad spectrum antibiotics, high-dose IV steroids, and placed on TPN. The abscess was drained by either CT guidance (16 patients) or US guidance (three patients). Indwelling catheters were left in 11 patients ranging in size from 6–14 French. One patient had a second catheter placed because of incomplete drainage, and one patient had the catheter manipulated when drainage decreased. No catheters were downsized. Only one patient with an indwelling catheter had it removed before surgery. All the patients who had only aspiration of the cavity had 50 or less milliliters of purulent material removed. Those with indwelling catheters had minimal to greater than 2 L of drainage from the catheter. Catheters were not injected with contrast to determine if there was an enteric fistula as all patients were to undergo ileocolectomy.

Patients were treated for a mean of 7.4±0.4 (range, 5–11) days before surgery. They then all underwent ileocolonic resection at which time a mean of 17.0±1.8 cm of terminal ileum or 26.9±2.3 cm when the normal cecum was included, was resected. One of the 19 patients was brought to the operating room urgently, due to deteriorating symptoms in spite of percutaneous drainage. Primary anastomosis was performed in 18 of 19 (95%) patients. Three patients required an ileostomy (two diverting loops and one end ileostomy). One stoma was given to a patient who had an unsuccessful initial drainage procedure with increasing abdominal pain and fever who was brought urgently to the OR. Two other stomas were in patients who were found to have residual loculated purulence at the time of surgery in spite of prior percutaneous drainage. Six of the seven patients who had only percutaneous aspiration avoided stoma placement at the time of resection.

In addition to the primary ileocolectomy, 15 fistulas were closed in 10 patients, 4 patients required a sigmoid resection and anastomosis, 2 patients had single stricturoplasties, and 1 patient had a small bowel resection. The fistulas were five entero–entero, six entero–colic, two entero–rectal, one entero–vesicle, and one enterocutaneous. In five patients, a minimal amount of residual purulence was found and was not felt to necessitate ileostomy. The diagnosis of CD was confirmed in every patient pathologically by review of the resected specimen.

The average total length of hospital stay was 13.9±0.6 (range, 11–19) days with an average of 6.5±0.4 (range, 4–11) days postoperatively. Five patients had post-op complications (two self-limited anastomotic bleeds, one small bowel obstruction, one wound infection, and one pelvic abscess requiring repeat postoperative percutaneous drainage). Two of these complications required readmission, the other three occurred during the initial hospitalization. Only one required reoperation (small bowel obstruction requiring lysis of adhesions).

Follow-up and Recurrence

Mean follow-up for these patients is 32±8 months with a range of 1–112 months. One patient had a recurrent abscess at 4 weeks postoperatively. It resolved with percutaneous drainage and oral antibiotics, and no further surgery was required. Two patients have undergone repeat ileocolectomy at 18 and 87 months, both for recurrent Crohn's disease at the anastomosis. Neither patient had a second intraabdominal abscess.

Discussion

Intraabdominal abscesses occur in 7–28% of patients with CD.[1,2] The primary goal in treating these patients is to drain the abscess and treat the sepsis. Treating the underlying diseased intestine is often a secondary consideration. Although not all intraabdominal abscesses in CD patients are associated with a persistent fistula, the majority of them are, and in such patients, the abscess is prone to recur if the underlying fistula is not treated.

An early treatment described by Nagler et al. was a two-stage surgical approach with the first stage being extraperitoneal surgical abscess drainage, followed by definitive surgical treatment of the diseased intestine 6 weeks later.[13] Nearly all patients treated in this manner developed an entero-cutaneous fistula before undergoing definitive surgical resection. Of the patients, 100% who underwent incision and drainage (I+D) only in the series by Ayuk et al., developed an enterocutaneous fistula.[3] Ribeiro et al. looked at 129 patients with intraabdominal abscesses and CD. In their series, patients underwent either primary en bloc resection of the abscess and diseased intestine (48%), I+D without removal of the diseased intestine (33%), bypass (12%), or ileostomy without removal of the diseased intestine (7%).[14] Fifty-seven percent of those who underwent I+D as their only procedure developed a fistula.[14] Twenty-four percent of patients who underwent en bloc

surgical treatment of the diseased intestine without prior abscess drainage developed a complication including two enterocutaneous fistulas and seven recurrent abscesses. Clearly, from these early reports, surgical protocols for management left much to be desired.

In the past two decades, image-guided percutaneous drainage of CD abscesses has been added to the armamentarium of the clinician. However, most reports describing patients treated with percutaneous drainage do not routinely have the diseased intestine subsequently removed. In many of these protocols, surgery is often reserved for patients with persistent symptoms or recurrent abscesses. A review of the literature of series with at least five patients with spontaneous intraabdominal abscesses from CD who underwent percutaneous drainage reveals a range of 7–72% success in avoidance of surgery with a mean of only 34% (Table 2).[3–11,15] In addition, the reported hospital stays for patients with intraabdominal abscesses in CD treated only by percutaneous drainage have been long. In the study by Doemeny et al., the patients were hospitalized 20 days to 3 months with a mean stay of 56 days.[12] Many patients had drains in place averaging 2–4 weeks with some catheters in place as long as 64 days.[4,7,15]Unplanned surgical resection after failed percutaneous drainage can also lead to long hospitalizations. In the report by Sahai et al. the patients who were successfully drained and did not go onto surgery were hospitalized 16.3±6.9 days, while the patients who failed and required surgery on the same admission were hospitalized 29.1±16.1 days.[10] Thus, protocols using percutaneous drainage, only in an attempt to avoid surgery, had remarkably long periods of morbidity and hospitalization.

Table 2 Review of Patients Undergoing Percutaneous Drainage for Spontaneous Intraabdominal Abscess in CD

Author	Year	Number of Patients	Number of Successful Drainages[a]
Ayuk et al.[3]	1996	8	3 (38%)
Bernini et al.[5]	1997	5	2 (40%)
Casola et al.[15]	1987	11	8 (72%)
Garcia et al.[6]	2001	7	33%
Gervais et al.[7]	2002	19	7 (37%) at 60 days 3 (16%) long term
Jawhari et al.[4]	1998	8	4 (50%)
Lambiase et al.[8]	1988	8	3 (38%)
Safrit et al.[9]	1986	7	2 (28%)
Sahai et al.[10]	1997	20	9 (45%)
Yamaguchi et al.[11]	2004	15	1 (7%)
Total		108	37 (34%)

Where possible, when a series included postoperative and spontaneous intraabdominal abscesses, only those spontaneous abscesses were included in this chart.

[a] Successful drainage indicates resolution of the abscess without surgery.

It was due to this clear need for an improved paradigm for management of this disease process that led us to implement a strict protocol of percutaneous drainage of the abscess, IV antibiotics, high-dose steroids, and definitive surgical resection on the same hospitalization, in an attempt to decrease complications and hospital stay while still avoiding the creation of a stoma.

With the presently described protocol, the patients had a mean hospital stay of only 13.2 days. The incidence of recurrent abscesses (1/19) and postsurgical complications were very acceptable and generally less than that seen in the literature describing patients treated with only one modality. By draining the abscess before surgery and using broad spectrum antibiotics, the amount of purulence found at surgery was minimized to a degree that permitted the performance of a primary anastomosis without a stoma in most of the patients. In the three patients that had ileostomies created, there was a large amount of residual purulence found at the time of surgery making unprotected primary anastomosis unsafe. One of these patients had failed management requiring urgent operation. Thus, only 2 of 18 successfully drained patients, required stomas. The use of IV steroids, we believe, helps to decrease the inflammation of CD and may possibly reduce the amount of intestine that needs to be resected. The length of bowel resected averaged only 26.9±2.3 cm (terminal ileum and cecum), which is a very reasonable resection in the context of a patient with CD and intraabdominal abscess.[16,17]

In this age of Infliximab and other biologic therapies for CD, the clinician might be tempted to use such drugs to treat these patients. However, one must be cautious in using Infliximab in the presence of undrained sepsis. Even if the sepsis was drained before the institution of Infliximab, results from such a care protocol must still be compared to one that includes definitive surgery as is presented here. We feel that this protocol, with its very favorable length of stay, the avoidance of a stoma in 84% of patients, and low overall recurrence and complication rate, provides a standard of care against which other protocols should be compared.

Conclusions

The use of percutaneous drainage has been a definite advance in the treatment of abscesses in CD, but it should not be the only form of treatment. Unless the diseased intestine is removed, the recurrence rate by historical reports is unacceptably high. CD patients presenting with intraabdominal abscess can safely undergo planned resection with primary anastomosis if initially treated with successful percutaneous drainage and aggressive medical management. Such a

protocol is associated with a very acceptable morbidity and LOS and avoids stoma creation in the majority of patients.

References

1. Keighley MRB, Eastwood D, Ambrose NBS, Allan RN, Burdon DW. Incidence and microbiology of abdominal and pelvic abscesses in Crohn's disease. Gastroenterology 1983;83:1271–1275.
2. Greenstein AJ, Sachar DB, Greenstein RJ, Janowitz HD, Aufses AH Jr. Intraabdominal abscess in Crohn's (ileo) colitis. Am J Surg 1982;143:727–730.
3. Ayuk P, Williams N, Scott NA, Nicholson DA, Irving MH. Management of intra-abdominal abscesses in Crohn's disease. Ann R Coll Surg Engl 1996;78:5–10.
4. Jawhari A, Kamm MA, Forbes A, Bartram CI, Hawley PR. Intra-abdominal and pelvic abscess in Crohn's disease: results of non-invasive and surgical management. Br J Surg 1998;85:367–371.
5. Bernini A, Spencer MP, Wong D, Rothenberger DA, Madoff RD. Computed tomography-guided percutaneous abscess drainage in intestinal disease. Factors associated with outcome. Dis Colon Rectum 1997;40:1009–1013.
6. Garcia JC, Persky SE, Bonis PAL, Topazian M. Abscesses in Crohn's disease outcome of medical versus surgical treatment. J Clin Gastroenterol 2001;32:409–412.
7. Gervais DA, Hahn PF, O'Neill MJ, Mueller PR. Percutaneous abscess drainage in Crohn's disease: technical success and short-term and long-term outcomes during 14 years. Radiology 2002;222:645–651.
8. Lambiase RE, Cronan JJ, Dorfman GS, Paolella LP, Haas RA. Percutaneous drainage of abscesses in patients with Crohn's disease. Am J Roentgenol 1988;150:1043–1045.
9. Safrit HD, Mauro MA, Jacques PF. Percutaneous abscess drainage in Crohn's disease. Am J Roentgenol 1987;148:859–862.
10. Sahai A, Belair M, Gianfelice D, Cote S, Gratton J, Lahaie R. Percutaneous drainage of intra-abdominal abscesses in Crohn's disease: short and long term outcomes. Am J Gastroenterol 1997;92:275–278.
11. Yamaguchi A, Matsui T, Sakurai T, Ueki T, Nakabayashi S, Yao T, Futami K, Arima S, Ono H. The clinical characteristics and outcome of intraabdominal abscess in Crohn's disease. J Gastroenterol 2004;39:441–448.
12. Doemeny JM, Burke DR, Meranze SG. Percutaneous drainage of abscesses in patients with Crohn's disease. Gastrointest Radiol 1988;13:237–241.
13. Nagler SM, Poticha SM. Intraabdominal abscess in regional enteritis. Am J Surg 1979;137:350–354.
14. Ribeiro MA, Greenstein AJ, Yamazaki Y, Aufses AH. Intra-abdominal abscess in regional enteritis. Ann Surg 1991;213:32–36.
15. Casola G, vanSonnenberg E, Neff CC, Saba RM, Withers C, Emarine CW. Abscesses in Crohn disease: percutaneous drainage. Radiology 1987;163:19–22.
16. Canin-Endres J, Salky B, Gattorno F, Edye M. Laparoscopically assisted intestinal resection in 88 patients with Crohn's disease. Surg Endosc 1999;13:595–599.
17. Bemelman WA, Slors JFM, Dunker MA, van Hogeznd RA, van Deventer SJ, Ringers J, Griffioen G, Gouma DJ. Laparoscopic-assisted vs open ileocolic resection for Crohn's disease, a comparative study. Surg Endosc 2000;14:721–725.

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Total Pancreatectomy: Indications, Operative Technique, and Postoperative Sequelae

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Abstract Total pancreatectomy has been used to treat both benign and malignant disease of the pancreas, but its use has been limited by concerns about management of the apancreatic state with its attendant total endocrine and exocrine insufficiency. Here, we review the indications for total pancreatectomy, operative technique, and improvements in the postoperative management of patients. Total pancreatectomy remains a viable option in the treatment of intractable pain associated with chronic pancreatitis, multicentric or extensive neuroendocrine tumors, patients with familial pancreatic cancer with premalignant lesions, and in patients with intraductal papillary mucinous neoplasia with diffuse ductal involvement or invasive disease. Improvements in postoperative management include auto-islet cell transplantation, advances in insulin formulations, and the use of glucagon rescue therapy which allow much tighter control of blood glucose than previously possible. This markedly lessens the risk of life-threatening hypoglycemia and decreases the risk of long-term complications, resulting in improved quality of life for these patients.

Keywords Pancreatic neoplasms · Chronic pancreatitis · Pancreatectomy · Diabetes

Abbreviations
IPMN intraductal papillary mucinous neoplasia

Billroth performed the first reported total pancreatectomy for pancreatic cancer in 1884[1]. This patient was said to have done well postoperatively, which would be questionable in the pre-insulin era. The first modern report of total pancreatectomy for pancreatic adenocarcinoma was by Rockey in 1943, with early patient death in the perioperative period from a bile duct leak[2]. Priestley performed the first successful total pancreatic resection in a hypoglycemic patient with a non-palpable 8×5 mm islet cell tumor in 1944[2]. As reports of long-term metabolic complications of the apancreatic state have accumulated, total pancreatectomy for benign, premalignant, and malignant disease has generally been avoided because of the perceived difficulty of managing the associated brittle diabetes. However, new formulations of long-acting insulin and improvements in the use of autologous islet cell transplantation have made total pancreatectomy an increasingly viable option in the treatment of both benign and selected malignant pancreatic diseases. Here, we will review the indications for total pancreatectomy, technical considerations, and postoperative management strategy for the apancreatic state.

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Indications

Chronic Pancreatitis

Warren first performed total pancreatectomy in patients with recurrent pancreatitis, proposing at that time that the

Table 1 Possible Indications for Total Pancreatectomy

Intractable pain of chronic pancreatitis
Neoplasm
Sporadic adenocarcinoma
Familial pancreatic cancer
Neuroendocrine tumors

procedure should be limited to patients with intractable pain and intraductal obstruction not amenable to a drainage procedure (Table 1)[3]. He theorized that if the pain associated with end-stage chronic pancreatitis arose from the inflamed pancreatic parenchyma, then total pancreatectomy should result in complete relief of symptoms. Initial experiences did not bear out this theory (Table 2). Several large retrospective studies showed that only 30 to 60% of patients undergoing total pancreatectomy experienced significant pain relief, and a large percentage of patients were readmitted for diabetic complications[4–7]. The Lahey Clinic's experience with total pancreatectomy, reported in 1978, showed that of the 25 patients studied, 12 had late deaths secondary to diabetic complications[6]. Many of these patients who died suffered from substance abuse, highlighting that this subset of patients was poorly suited for the intensive patient involvement required for the maintenance of normoglycemia following total pancreatectomy and were more likely to complain of continued pain postoperatively[8].

Advancements in the use of autologous islet cell transplantation have led to renewed interest in the use of total pancreatectomy for relief of the pain associated with chronic pancreatitis. The University of Minnesota has the largest experience with the use of total pancreatectomy and autologous islet transplantation in the current era, as described by Gruessner and colleagues[9]. The records of 112 patients were reviewed, with follow-up ranging from 4 months to 26 years. Islets were isolated following pancreatectomy using the semiautomated technique originally described by Ricordi, followed by intraoperative infusion into the portal venous system[10]. Of 112 patients, 70% experienced significant pain relief based on comparison of pre- and postoperative narcotic requirements. Importantly, 72% of patients who had not undergone previous pancreatic resection did not require insulin postoperatively. Those with previous pancreatic resection (and thus, with fewer islets available for autotransplantation) fared worse, with only about 20% of patients achieving insulin independence. Patients actively abusing alcohol were not enrolled in the study. Two additional reports from the University of Cincinnati and University of Leicester (with patient follow-up between 3 months–3 years and 7 months- –6 years, respectively) show similarly excellent pain control in patients who were not actively abusing alcohol. The University of Cincinnati group reported that 82% of the 22 patients in their series were able to be weaned entirely from narcotics. At both centers, approximately 40–50% of patients became insulin independent following the procedure[11,12]. The need for insulin postoperatively following this procedure appears to be due to a number of factors, including number of islets transplanted, similar to that observed with non-autologous transplantation used to treat type 1 diabetes[13]. There is evidence that maintenance of normoglycemia in the immediate postoperative period is necessary to insure recovery of transplanted islet cell function, which can occur as late as 1 year following transplantation[12]. Because higher islet recovery rates translate into better outcomes, proponents of this approach argue for early pancreatectomy in patients with chronic pancreatitis before they develop glucose intolerance due to loss of β-cell mass.

Pancreatic allotransplantation has also been used as an alternative approach in patients with previous total pancreatectomy for chronic pancreatitis; long-term results in the small number of patients[14] who have undergone this procedure at the University of Minnesota show a 3-year graft survival of 77% in the tacrolimus era[9]. There were no transplant-related deaths in this group. With total organ transplantation, these patients benefit from restoration of exocrine and endocrine function if enteric rather than bladder drainage is provided. The potential benefits of the procedure must be balanced with the need for lifelong immunosuppression and the resultant morbidity and mortality associated with rejection, infection, and malignancy.

Table 2 Review of Series of Total Pancreatectomy for Chronic Pancreatitis

Author	Year	Number of patients	Median follow-up	Percentage of pain improvement	Autotransplant	Percentage of nondiabetic
Gruessner, et al.[9]	2004	132	0.3–26 yr	72	Yes	33
Clayton, et al.[12]	2003	31	2–6 yr	50	Yes	0
Rodríguez-Rilo, et al.[11]	2003	19	19 mos (3 to 41)	94	Yes	40
Easter, et al.[66]	1991	8	29 mos (8 to 51)	75	No	0
Stone, et al.[4]	1988	15	9.1 yr (2.1 to 13.1)	67	No	0
Braasch, et al.[6]	1978	26	Unknown	78	No	0

Sporadic Pancreatic Adenocarcinoma

Historically, the rationale for total pancreatectomy for the treatment of pancreatic adenocarcinoma stems from: (1) the desire to avoid the complications of pancreatic fistula; (2) the belief that the disease is frequently multicentric; and (3) the view that total pancreatectomy represents a more definitive oncologic resection than a partial pancreatic resection, with greater lymph node clearance and an increase in the percentage of R0 resections. Approaching these arguments point by point: (1) Recent retrospective reviews of the complications of pancreatic resection show pancreatic fistula rates between 3 and 11% at high volume centers[14,15]. More than 90% of those patients who develop pancreatic fistulas are now managed successfully with percutaneous drainage, without the relatively high mortality rate (up to 40%) previously reported with this complication[16]. (2) Brooks et. al. in the 1960s reported that up to 34% of patients with pancreatic adenocarcinoma undergoing resection had multicentric disease[17]. This view was supported by data in two other reports[18,19]. More recent studies using immunohistochemistry and PCR have found multicentricity to be much less prevalent, ranging between 0 and 6%[20,21]. The prevalence of multicentric disease in earlier studies may have been due to sampling error, or operator bias, in the identification of truly discontinuous lesions. It could also be due to the inclusion of a disproportionate number of cases of familial pancreatic cancer[3]. Regarding the efficacy of total pancreatectomy as an oncologic operation, large retrospective series have shown no long-term survival benefit. In fact, several studies show that perioperative mortality is higher with total pancreatectomy than with subtotal pancreatectomy. Ihse, et al. reported an in-hospital mortality of 27% of their 89 patients undergoing total pancreatectomy for cancer[22]. In a more recent report from Memorial Sloan Kettering Hospital, 28 patients who underwent total pancreatectomy for adenocarcinoma had a 5-year survival rate of 9%, no better than that seen in patients undergoing partial pancreatectomy for adenocarcinoma[24]. Given the fact that pancreatic fistulas are now better managed, most tumors are not multicentric and that total pancreatectomy results in higher perioperative morbidity and mortality with no increased long-term survival, there is no role for routine consideration of total pancreatectomy in the management of sporadic pancreatic adenocarcinoma.

Familial Pancreatic Adenocarcinoma

In families affected by this condition, first degree relatives with three or more affected family members have up to a 57-fold increase in the risk of developing pancreatic cancer[25]. The susceptibility to pancreatic cancer is inherited in an autosomal dominant fashion[26]. Germline mutations in BRCA2 have been identified in up to 20% of affected families, and recently, a susceptibility locus has been mapped to chromosome 4q32–34[27–29]. In patients with familial pancreatic cancer, there is often an early age of onset, and some question of anticipation (i.e., the disease presents earlier with a more aggressive clinical course in succeeding generations)[30–32]. Other familial cancer syndromes also predispose to pancreatic cancer, most notably Peutz–Jegher syndrome, familial adenomatous polyposis (FAP), hereditary non-polyposis colorectal carcinoma (HNPCC), familial breast–ovarian cancer, and familial atypical multiple mole melanoma (FAMMM). Hereditary pancreatitis, a result of either a mutation in the cationic trypsinogen gene (PRSS1) or in the serine protease inhibitor (SPINK1), also results in increased susceptibility to pancreatic cancer. Screening with endoscopic ultrasound has been recommended in asymptomatic patients with two or more first-degree relatives with pancreatic cancer, one first-degree relative with cancer diagnosed before the age of 50, or with two or more second-degree relatives, one of whom was diagnosed before the age of 50[33]. The exact timing for initiating surveillance is up to some debate, with experts agreeing that surveillance should begin somewhere between 5 to 10 years before the onset of pancreatic cancer in the youngest affected relative or by the age of 40 or 50, or at the onset of symptoms (including the development of diabetes or weight loss)[25,33]. In a study by Rulyak and colleagues of 35 family members undergoing surveillance, 12 had positive findings of a mass lesion on endoscopic ultrasound (EUS) and/or endoscopic retrograde cholangiopancreatography (ERCP)[35]. These patients underwent either total or partial pancreatectomy and all 12 had pancreatic dysplasia on histological examination without invasive adenocarcinoma. These patients were found to have extensive, multicentric lesions. A larger group of 50 kindreds has been followed at the University of Washington, of which 10 patients have undergone total pancreatectomy. All were found to have Pan IN-3 lesions (carcinoma in situ). One patient with postoperative hypoglycemic unawareness underwent solid organ pancreas transplantation and had normal glucose homeostasis 1-year post-transplant[35]. Clearly, surveillance and total pancreatectomy have the potential to avert the development of invasive pancreatic adenocarcinoma in the setting of familial pancreatic cancer and should be considered as a prophylactic procedure in some patients.

Neuroendocrine Tumors

In 1993, a large retrospective study from the Mayo Clinic showed completion pancreatectomy for recurrent insulinoma to be associated with 10 year decrease in mean survival when

compared to patients undergoing repeat partial pancreatectomy[36]. It should be noted that all of these patients underwent resection before 1977. More recent analysis suggests that endocrine tumors do not have as benign a course as previously thought, and that aggressive resection might be warranted, including total pancreatectomy. Doherty et al. examined a group of 34 distinct kindreds of multiple endocrine neoplasia type I (MEN-I) syndrome with 1,838 members and found that 46% of MEN-I patients died as a result of their endocrine tumors at a median age of 47 years[37]. These patients succumbed to metastatic islet cell or carcinoid tumors, ulcer disease, or complications of hypercalcemia. Aggressive screening of MEN-I patients using endoscopic ultrasound was therefore suggested[38]. In 2003, Norton et al. reported three patients who had undergone total pancreatectomy for locally advanced neuroendocrine tumors without postoperative complications[39]. As our understanding of the natural history of pancreatic neuroendocrine tumors has evolved, it is clear that a place remains for completion pancreatectomy in the endocrine surgeon's armamentarium.

Intraductal Papillary Mucinous Neoplasm (IPMN)

Ohhashi et al. first described IPMN of the pancreas in 1982; initially, it was thought to be an indolent disease with a favorable prognosis[40,41]. It is now widely recognized to be a premalignant lesion with between 30 and 72% of patients having invasive or noninvasive carcinoma at the time of presentation[42,43]. Those patients found to have invasive carcinoma after resection have a poor prognosis, with a 5-year survival ranging from 24–60%[42,44–46]. The distribution of IPMN has been proposed as a predictor of progression: lesions involving the main pancreatic duct have a higher rate of malignancy discovered at the time of resection than lesions arising from a branch duct[47]. Intraoperative frozen sections following planned partial resection for localized lesions are necessary to assure negative margins. If there is evidence of severe dysplasia or invasive cancer at the resection margin, the resection should be extended, up to and including total pancreatectomy. In patients with diffuse noninvasive disease, total pancreatectomy should be considered in select patients to minimize the chance of recurrent, invasive disease.

Operative Technique

The operative technique utilized for total pancreatectomy depends upon whether the patient has undergone previous pancreatic resection. Distal pancreatectomy can be performed in patients who had a previous pancreatoduodenectomy. Patients with a previous distal pancreatectomy are candidates for either duodenum-preserving pancreatectomy or completion pancreaticoduodenectomy. Preservation of the spleen should be considered whenever possible if it is felt not to compromise the oncologic nature of the operation. In cases when accompanying splenectomy is planned, the patient should be vaccinated 2 weeks preoperatively against pneumococcus, Hemophilus influenza group B, and meningiococcus group C to minimize the likelihood of developing potentially lethal post-splenectomy sepsis.

The operative procedure begins with a thorough exploration to evaluate the presence of extra-pancreatic disease. The right colon and hepatic flexure of the colon are mobilized to provide access to the second part of the duodenum. A wide Kocher maneuver is performed, and the duodenum and pancreas are elevated off the inferior vena cava until the left border of the abdominal aorta can be palpated. The Kocher maneuver is extended by continuing mobilization of the third portion of the duodenum until the superior mesenteric vein is encountered. The gastrocolic ligament is widely divided to allow access to the body of the pancreas. The anterior surface of the superior mesenteric vein is identified and dissected under direct vision. Using a Cushing vein retractor, the neck of the pancreas is lifted, and entering this avascular plane, the superior mesenteric vein is traced proximally to its confluence with the portal vein. Following cholecystectomy, the peritoneal reflection over the hepatoduodenal ligament is carefully opened, and the common bile duct and common hepatic artery are carefully dissected, and vessel loops are placed around them. The gastroduodenal artery is identified and ligated in continuity to facilitate access to the portal vein at the superior aspect of the pancreas. The splenorenal ligament is divided, and the spleen is drawn medially together with the tail of the pancreas, thus, opening the retropancreatic plane. The splenic vein and artery are ligated. Next, the distal part of the stomach is mobilized and transected. The duodenojejunal flexure is located and dissected free from the retroperitoneum by dividing the ligament of Treitz. Approximately 10 to 15 cm distal to the duodenojejunal flexure, the vessels within the mesentery and subsequently the small bowel are divided. The pancreas, distal stomach, duodenum, and spleen are removed en bloc (Fig. 1). To restore gastrointestinal continuity, an end-to-side choledochojejunal anastomosis is performed. The stomach is then anastomosed to the jejunum in two layers.

Metabolic Consequences of Total Pancreatectomy

Endocrine Insufficiency

The diabetic state induced by total pancreatectomy is characterized by complete insulin deficiency (as confirmed

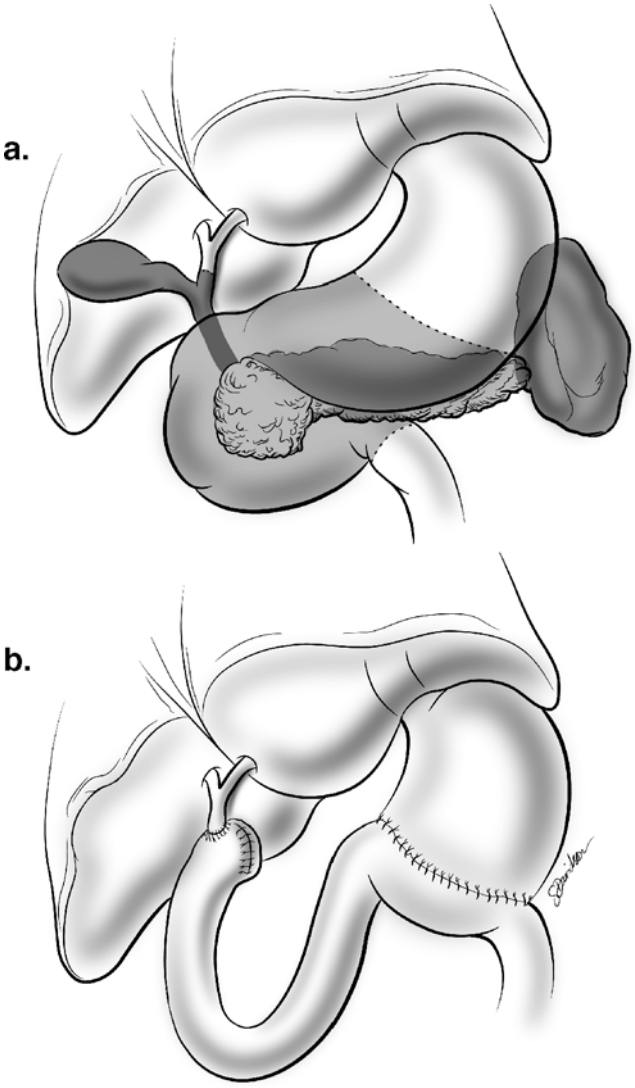

Figure 1 Total pancreatectomy with partial gastrectomy, duodenectomy, cholecystectomy, and splenectomy with choledochojejeunostomy and gastrojejeunostomy.

by the absence of C-peptide in the serum), pancreatic polypeptide deficiency, and an absence of functional glucagon[48,49]. Because the apancreatic state is characterized by a defect in gluconeogenesis secondary to hypoglucagonemia, daily insulin requirements in these patients are typically lower than in type I or type II diabetics[50,51]. However, the therapeutic window is narrowed, resulting in frequent episodes of mild to severe postprandial hypoglycemia following insulin administration (Table 3)[52]. Patients who are chronically hypoglycemic have been shown to upregulate cerebral endothelial glucose transporters which are responsible for the initiation of the autonomic response to hypoglycemia[53]. This results in an attenuation of epinephrine secretion and may account for episodes of diabetic unawareness in pancreatectomized individuals[54,55]. Reductions in the adrenal secretion of epinephrine also decrease hepatic glucose production[56]. The combination of insulin sensitivity and hypoglycemic unawareness was termed "brittle" diabetes by R.T. Woodyatt in the 1930s[57].

Insulin therapy for the apancreatic patient has been simplified by the availability of the long-acting insulin, glargine, a recombinant human insulin analogue which can be dosed once or twice daily. Because glargine is less soluble than native human insulin at physiological pH, there is a delayed absorption, resulting in a relatively 'peakless' insulin profile. The combined use of glargine with supplemental short-acting insulins such as insulin lispro or insulin aspart at mealtimes helps to prevent postprandial hypoglycemia after intestinal carbohydrate absorption has been completed. Most patients are able to achieve adequate glycemic control using these insulin preparations. However, continuous subcutaneous insulin infusion pumps have also been used to simplify dosing for patients[58]. The use of insulin lispro or insulin aspart via continuous infusion has been shown in several open-label, randomized, crossover trials in type I and type II diabetic patients to provide better control of postprandial hyperglycemia and a significantly lower glycosylated hemoglobin level, with lower daily insulin requirements and less hypoglycemic episodes than with the use of regular insulin in the pump[58].

Current clinical work in patients with type I diabetes suggests that glucagon rescue injections can help prevent late postprandial hypoglycemia[59]. Glucagon replacement therapy has been attempted in small numbers of apancreatic patients[60,61]. Tankjoh et al., reported that when a physiological dose of glucagon is given proportional to the amount of insulin administered, the utilization of glycogenic amino acids and lipids increased along with a marked improvement in the utilization of carbohydrates.

Exocrine Insufficiency

Exocrine insufficiency also complicates postoperative management following total pancreatectomy. Even with aggressive pancreatic enzyme replacement (up to 120,000 IU of lipase per meal taken in conjunction with a proton pump

Table 3 Clinical Differences Between Type I Diabetes and the Apancreatic State

Conditions	Type I	Apancreatic diabetes
Glycemic instability	++	+++
Insulin requirement	+++	+
Hypoglycemia	++	+++
Ketoacidosis	+++	+
Vascular complications	+++	+

inhibitor to prevent early inactivation of the enzymes by gastric acid), patients continue to have moderate steatorrhea which causes glucose malabsorption and further complicates diabetic management[62,63]. Because of fat and glucose malabsorption, these patients can require an intake of up to 5,000 k/cal per day to maintain their body weight[64]. High-calorie/complex carbohydrate diets with aggressive vitamin and calcium supplementation can help prevent weight loss, control postprandial glycemic shift, and prevent the osteoporosis associated with the apancreatic state. Some simple sugars should be taken at the beginning of a meal following the injection of short acting insulin because of its rapid onset of action. In some patients, it will be necessary to delay insulin injection until after the meal to reduce the risk of hypoglycemia.

Maintenance of continuity of the upper gastrointestinal tract has been proposed as a mechanism to improve absorption following total pancreatectomy. Buchler et al. have reported improved glucose tolerance following duodenum-preserving pancreatic head resection when compared to pylorous-preserving pancreaticoduodenectomy, leading some to speculate that patients undergoing total pancreatectomy might also benefit from more conservative resection[65,66]. Easter in 1991 reported that of eight patients undergoing duodenum-sparing total pancreatectomy for the pain of chronic pancreatitis, none experienced problems with the control of diabetes or any hypoglycemic attacks requiring medical treatment[66]. Proposed mechanisms include improved intestinal transit, improved oral intake, and maintenance of sufficient insulin- and pancreatic polypeptide-secreting tissue to ameliorate the effects of pancreatic resection. However, more recent series of patients undergoing duodenum-sparing total pancreatectomy for chronic pancreatitis reported no statistically significant differences in diabetic complications[67].

Steatohepatitis and Liver Failure

Another metabolic consequence of the apancreatic state is the development of steatohepatitis with progressive liver failure. Dressler, et al. noted that three of the patients in their series of 49 followed at Memorial Sloan-Kettering died of complications of hepatic failure, only one of whom had significant preoperative alcohol abuse[64]. Centrilobular steatosis was documented in two of these patients. All 49 patients demonstrated a durable elevation in levels of serum aspartate aminotransferase and alkaline phosphatase, but the degree of elevation did not correlate with an increased risk for the development of steatohepatitis. It has been hypothesized that decreased hepatic stimulation by glucagon results in progressive fatty deposition in the liver[69]. Periodic evaluation of hepatic aminotransferases, serum bilirubin, and prothrombin time are recommended to evaluate hepatic function in pancreatectomized patients. These patients are also at increased risk for the development of marginal ulcers or peptic ulcer disease secondary to lack of bicarbonate secretion, mandating proton pump inhibitor therapy[63].

Quality of Life Following Total Pancreatectomy

A relative paucity of data exist on quality of life following total pancreatectomy in the current era. In 2004, 20 patients who had undergone total pancreatectomy at the University of Verona, a mean of 34 months earlier (range, 1.5–112 months), were surveyed with the European Organization for Research and Treatment of Cancer Quality of Life Questionnaire (EORTC QLQ). They were found to have a median insulin requirement of 30.5 IU/day with one patient requiring subcutaneous insulin infusion. Among the patients, 88% had a normal HbA1C level, while 72% patients claimed to have hypoglycemic episodes at least weekly. The median QOL score was 5.5 (range, 3–7) and the median health status score was 5 (range, 3–7), similar to age-matched patients with type II diabetes[69]. More recently, 34 patients who had undergone total pancreatectomy at the Mayo Clinic were surveyed with multiple quality-of-life instruments (SF-36, Audit of Diabetes Dependent Quality of Life, EORTC PAN26) and were found to have quality of life scores equivalent to age- and sex-matched diabetics[70]. Three alcoholic patients died of late hypoglycemic episodes, emphasizing the importance of screening for substance abuse before performing total pancreatectomy. The authors concluded that in appropriately selected patients, total pancreatectomy can provide an acceptable quality of life.

Conclusions

In conclusion, total pancreatectomy remains a viable option in the treatment of (1) intractable pain associated with chronic pancreatitis, (2) multicentric or extensive neuroendocrine tumors, (3) patients with familial pancreatic cancer with premalignant lesions, and (4) in patients with IPMT with diffuse ductal involvement or invasive disease. The use of islet autotransplantation in selected patients with chronic pancreatitis and pancreatic allografts in young patients with premalignant disease can also be included in the armamentarium of treatment options for pancreatectomized patients. In patients who are not candidates for transplantation, advances in insulin formulations and the use of glucagon rescue therapy allow much tighter control of blood glucose than previously possible, markedly lessening the risk of life-threatening hypoglycemia and

decreasing the risk of long-term complications, resulting in improved quality of life for these patients.

References

1. Sauve L. Des pancreatectemies et specialement de la pancreatectomie cephalique. Rev Chir 1908;37:113–152.
2. Rockey EW. Total pancreatectomy for carcinoma: Case Report. Ann Surg 1943;118:603–611.
3. Warren KW, Poulantzas JK, June GA. Life after total pancreatectomy for pancreatitis. Ann Surg 1966;164:830–834.
4. Stone WM. Chronic pancreatitis: results of Whipple's resection and total pancreatectomy. Arch Surg 1988;123(7):815–819.
5. Gall FP, Muhe E, Gebhardt C. Results of partial and total pancreaticoduodenectomy in 117 patients with chronic pancreatitis. World J Surg 1981;5(2):269–275.
6. Braasch JW, Vito L, Nugent FW. Total pancreatectomy of end-stage chronic pancreatitis. Ann Surg 1978;188(3):317–322.
7. Sarr MG, Sakorafas GH. Incapacitating pain of chronic pancreatitis: a surgical perspective of what is known and what is not known. Gastrointest Endosc 1999;49:S85–S89.
8. Warshaw AL, Banks PA, Fenandez-del Castillo C. Treatment of pain in chronic pancreatitis. Gastroenterology 1998;115 (3):765–776.
9. Gruessner RWG, Sutherland DER, Dunn DL, Najarian JS, Jie T, Hering BJ, Gruessner AC. Transplant options for patients undergoing total pancreatectomy for chronic pancreatitis. J Am Coll Surg 2004;198:559–567.
10. Ricordi C, Lacy PE, Scharp DW. Automated islet isolation from human pancreas. Diabetes 1989;38:140–142.
11. Rodriguez-Rilo HL, Ahmad SA, D'Alessio D, Iwanaga Y, Kim J, Choe KA, Moulton JS, Martin J, Pennington LJ, Soldano DA, Biliter J, Martin SP, Ulrich CD, Somogyi L, Welge J, Matthews JB, Lowy AM. Total pancreatectomy and autologous islet cell transplantation as a means to treat severe chronic pancreatitis. J Gastrointest Surg 2003;7(8):978–989.
12. Clayton HA, Davies JE, Pollard CA, White SA, Musto PP, Dennison AR. Pancreatectomy with islet autotransplantation for the treatment of severe chronic pancreatitis: the first 40 patients at the Leichester General Hospital. Transplantation 2003;76:92–98.
13. Ricordi C, Inverardi L, Kenyon NS, Goss J, Bertuzzi F, Alejandro R. Requirements for success in clinical islet transplantation. Transplantation 2005;79(10):1298–1300 (May 27).
14. Buchler MW, Wagner M, Schmied BM, Uhl W, Friess H, Z'graggen K. Changes in morbidity after pancreatic resection: toward the end of completion pancreatectomy. Arch Surg 2003;138:1310–1314.
15. Lin JW, Cameron JL, Yeo CJ, Riall TS, Lillemoe KD. Risk factors and outcomes in postpancreaticoduodenectomy pancreaticocutaneous fistula. J Gastrointest Surg 2004;8(8):951–959 (Dec).
16. Munoz-Bongrand N, Sauvanet A, Denys A, Sibert A, Vilgrain V, Belghiti J. Conservative management of pancreatic fistula after pancreaticoduodenectomy with pancreaticogastrostomy. J Am Coll Surg 2004;199(2):198–203 Aug.
17. Collins JJ, Craighead JE, Brooks JR. Rationale for total pancreatectomy for carcinoma of the pancreatic head. N Engl J Med 1966;274:599–602.
18. Van Heerden JA, ReMine WH, Weiland LH, McIlrath DC, ILstrup DM. Total pancreatectomy for ductal adenocarcinoma of the pancreas: Mayo Clinic experience. Am J Surg 1981;142:308–311.
19. Ihse I, Lilja P, Anresjo B, Bengmark S. Total pancreatectomy for cancer: an appraisal of 65 cases. Ann Surg 1977;186:675–680.
20. Kloppel G, Lohse T, Bosslet K, Ruckert K. Ductal adenocarcinoma of the head of the pancreas: incidence of tumor involvement beyond the whipple resection line. Histological and immunocytochemical analysis of 37 total pancreatectomy specimens. Pancreas 1987;2:170–175.
21. Motojima K, Urano T, Nagata Y, Shiku H, Tsurfune T, Kanematsu T. Detection of point mutations in the Kristen-ras oncogene provides evidence for the multicentricity of pancreatic adenocarcinoma. Ann Surg 1993;217(2):138–143.
22. Ihse I, Anderson H, Andren-Sandberg A. Total pancreatectomy for cancer of the pancreas: is it appropriate? World J Surg 1996;20:288–294.
23. Sarr MG, Behrns KE, vanHeerden JA. Total pancreatectomy: an objective analysis of its use in pancreatic cancer. Hepatogastroenterol 1993;40:418–421.
24. Karpoff H, Klimstra DS, Brennan MF, Conlon KC. Results of total pancreatectomy for adenocarcinoma of the pancreas. Arch Surg 2001;136:44–47.
25. Bartsch DK. Familial pancreatic cancer. Br J Surg 2003;90:386–387.
26. Lynch HT, Lanspa SJ, Fitzgibbons RJ, Smyrk T, Fitzsimmons ML, McClellan J. Familial pancreatic cancer: genetic pathology review. Nebr Med J 1989;74:109–112.
27. Klein AP, Brune KA, Petersen GM, Goggins M, Tersmette AC, Offerhaus GJ, Griffin C, Cameron JL, Yeo CJ, Kern S, Hruban RH. Prospective risk of pancreatic cancer in familial pancreatic cancer kindreds. Cancer Res 2004;64(7):2634–2638.
28. Hahn SA, Greenhalf B, Ellis I, Sina-Frey M, Rieder H, Korte B, Gerdes B, Kress R, Ziegler A, Raeburn JA, Campra D, Grutzmann R, Rehder H, Rothmund M, Schmiegel W, Neoptolemos JP, Bartsch DK. BRCAZ germline mutations in familial pancreatic cancer. J Natl Cancer Inst 2003;5:180–181.
29. Eberle MA, Pfutzer R, Pogue-Geile KL, Bronner MP, Crispin D, Kimmey MB, et al. A new susceptibility locus for autosomal dominant pancreatic cancer maps to chromosome 4q32–34. Am J Hum Genet 2002;70:1044–1048.
30. Greenhalf W, McFaul C, Earl J, Howes N, Neoptolemos J, Kress R, Sina-Frey M, Rieder H, Hahn SA, Bartsch D. Anticipation in familial pancreatic cancer. Gut 2006;2155(2):252–258.
31. Reider H, Sina-Frey M, Ziegler A, Hahn SA, Przypadlo E, Kress R, et al. German national case collection of familial pancreatic cancer-clinical-genetic analysis of the first 21 families. Onkologie 2002;25:262–266.
32. Tersmette AC, Petersen GM, Offerhaus GJ, Falatko FC, Brune KA, Goggins M, et al. Increased risk of incident pancreatic cancer among first-degree relatives with familial pancreatic cancer. Clin Cancer Res 2001;7:738–744.
33. Brentnall TA. Cancer surveillance of patients from familial pancreatic cancer kindreds. Med Clin North Am 2000;84 (3):707–718.
34. Rulyak SJ, Brentnall TA. Inherited pancreatic cancer: surveillance and treatment strategies for affected families. Pancreatology 2001;1(5):477–485.
35. Charpentier KP, Brentnall TA, Bronner MP, Byrd D, Christopher M. A new indication for pancreas transplantation: high grade pancreatic dysplasia. Clin Transplant 2004;18:105–107.
36. Thompson GB, Service FJ, van Heerden JA, Carney JA, Charboneau JW, O'Brien PC, Grant CS. Reoperative insulinomas, 1927 to 1992: an institutional experience. Surgery 1993;114 (6):1196–1206.
37. Doherty GM, Olson JA, Frisella MM, Lairmore TC, Wells SA, Norton JA. Lethality of multiple endocrine neoplasia type I. World J Surg 1998;22:581–587.

38. Thompson NW. Invited commentary. World J Surg 1998;22 (6):586–587.
39. Norton JA, Kivlen M, Li M, Schneider D, Chuter T, Jensen RT. Morbidity and mortality of aggressive resection in patients with advanced neuroendocrine tumors. Arch Surg 2003;138:859–866.
40. Ohhashi K, Murakami Y, Takekoshi T, Ohta H, Ohhashi I. Four cases of "mucin-producing" cancer of the pancreas on specific findings of the papilla of Vater. Progress of Digestive Endoscopy 1982;20:348–351.
41. Rickaert F, Cremer M, Devière J, Tavares L, Lambilliotte JP, Schroder S, et al. Intraductal mucin-hypersecreting neoplasms of the pancreas: a clinicopathologic study of eight patients. Gastroenterology 1991;101:512–519.
42. Jang JY, Kim SW, Ahn YJ, Yoon YS, Choi MG, Lee KU, et al. Multicenter analysis of clinicopathologic features of intraductal papillary mucinous tumor of the pancreas: is it possible to predict the malignancy before surgery? Ann Surg Oncol 2005;12(2):124–132 (Feb).
43. Sarr MG, Kendrick ML, Nagorney DM, Thompson GB, Farley DR, Farnell MB. Cystic neoplasms of the pancreas: benign to malignant epithelial neoplasms. Surg Clin North Am 2001;81 (3):497–509.
44. Cuillerier E, Cellier C, Palazzo L, Deviere J, Wind P, Rickaert F, Cugnenc PH, Cremer M, Barbier JP. Outcome after surgical resection of intraductal papillary and mucinous tumors of the pancreas. Am J Gastroenterol 2000;95(2):441–445.
45. Azar C, Van de Stadt J, Rickaert F, Deviere M, Baize M, Kloppel G, Gelin M, Cremer M. Intraductal papillary mucinous tumours of the pancreas. Clinical and therapeutic issues in 32 patients. Gut 1996;39(3):457–464.
46. Raimondo M, Tachibana I, Urrutia R, Burgart LJ, DiMagno EP. Invasive cancer and survival of intraductal papillary mucinous tumors of the pancreas. Am J Gastroenterol 2002;97(10):2553–2558 (Oct.).
47. Obara T, Maguchi H, Saitoh Y, Itoh A, Arisato S, Ashida T, Nishino N, Ura H, Namiki M. Mucin-producing tumor of the pancreas: natural history and serial pancreatogram changes. Am J Gastroenterol 1993;88:564–569.
48. Doi K, Prentki M, Yip C, Muller WA, Jeanreneaud B, Vrainic M. Identical biological effects of pancreatic glucagons and a purified moiety of canine gastric immunoreactive glucagons. J Clin Invest 1979;63:525–531.
49. Muller WA, Brennan MF, Tan MH, Aoki TT. Studies of glucagon secretion in pancreatectomised patients. Diabetes 1974;23:512–516.
50. Vigili de Kreutzenberg S, Maifreni L, Lisato G, Riccio A, Trevisan R, Tiengo A, DelPrato S. Glucose turnover and recycling in diabetes secondary to total pancreatectomy: effect of glucagons infusion. J Clin Endocrinol Metab 1990;70:1023–1029.
51. Muller WA, Brennan MF, Tan MH, Aoki TT. Studies of glucagon secretion in pancreatectomized patients. Diabetes 1974;23(6):512–516 (Jun).
52. Slezak LA, Andersen DK. Pancreatic resection: effects on glucose metabolism. World J Surg 2001;25:452–460.
53. Boyle PJ. Alteration in brain glucose metabolism induced by hypoglycaemia in man. Diabetologia 1997;40(Suppl 2):S69–S74.
54. Horie H, Matsuyama Namba M, et al. Responses of catecholamines and other counterregulatory hormones to insulin-induced hypoglycemia in totally pancreatectomized patients. J Clin Endocrinol Metab 1984;59:1193–1196.
55. Amiel SA, Sherwin RS, Simonson DC, Tamborlane WV. Effective of intensive insulin therapy on glycemic thresholds for counterregulatory hormone release. Diabetes 1988;37:901–907.
56. Butler PC, Rizza RA. Regulation of carbohydrate metabolism and response to hypoglycemia. Endocrinol Metab Clin N Am 1989;18 (1):1–25 (Mar).
57. Tattersall RB. Brittle diabetes revisited: the Third Arnold Bloom Memorial Lecture. Diabet Med 1997;14(2):99–110 (Feb.).
58. Radermecker RP, Scheen AJ. Continuous subcutaneous insulin infusion with short-acting insulin analogues or human regular insulin: efficacy, safety, quality of life, and cost-effectiveness. Diabetes/metab Res Rev 2004;20(3):178–188 (May–Jun).
59. Heptulla RA, Rodriguez LM, Bomgaars L, Haymond MW. The role of amylin and glucagon in the dampening of glycemic excursions in children with type 1 diabetes. Diabetes 2005;54 (4):1100–1107.
60. Bajorunas DR, Fortner JG, Jaspan J, Sherwin RS. Total pancreatectomy increases the metabolic response to glucagon in humans. J Clin Endocrinol Metab 1986;63:439–446.
61. Tankjoh K, Tomita R, Mera K, Hayashi N. Metabolic modulation by concomitant administration of insulin and glucagon in pancreatectomy patients. Hepatogastroentereology 2002;49 (44):538–543.
62. Petrides A, DeFronzo RA. Glucose metabolism in cirrhosis: a review with some perspective for the future. Diabetes/metab Rev 1989;5:691–709.
63. Kahl S, Malfertheiner P. Exocrine and endocrine pancreatic insufficiency after pancreatic surgery. Best Pracitce and Research in Clinical Gastroenterology 2004;18:947–955.
64. Dressler CM, Fortner JG, McDermott K, Bajorunas DR. Metabolic consequences of (regional) total pancreatectomy. Ann Surg 1991;214:131–140.
65. Buchler M, Freiss H, Muller M, Wheatley A, Berger H. Randomized trial of duodenum-preserving pancreatic head resection versus pylorus-preserving Whipple in chronic pancreatitis. Am J Surg 1994;65:458–467.
66. Easter DW, Cuschieri A. Total pancreatectomy with preservation of the duodenum and pylorus for chronic pancreatitis. Ann Surg 1991;214:575–580.
67. Alexakis N, Ghaneh P, Connor S, Raraty M, Sutton R, Neoptolemos JP. Duodenum- and spleen preserving total pancreatectomy for end-stage chronic pancreatitis. Br J Surg 2003;90: 1401–1408.
68. DeFronzo RA. Pathogenesis of type 2 diabetes mellitus. Med Clin North Am 2004;88(4):787–835 (Jul).
69. Landoni L, Salvia R, Festa L, Muselli P, Giardino A, Butturini G, Falconi M, Bassi C, Pederzoli. Quality of life after total pancreatectomy. Ten-year experience. J Pancreas (Online) 2004;5(5 Suppl):441.
70. Billings BJ, Christein JD, Harmsen WS, Harrington JR, Chari ST, Que FG, Farnell MB, Nagorney DM, Sarr MG. Quality-of-life after total pancreatectomy: is it really that bad on long-term follow-up? J Gastrointest Surg 2005;9(8):1059–1067 (Nov–Dec).

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Open and Laparoscopic Roux-en-Y Gastric Bypass: Our Techniques

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Obesity has become a national health crisis of epidemic proportions. The explosion (and realization) of interest from both medical circles and from the lay public in bariatric surgery has stimulated the development of many bariatric surgical programs across the United States. This article will be a purely technical one addressing how we perform the Roux-en-Y gastric bypass (RYGB) using open and laparoscopic techniques. Our intent is to describe our specific techniques and innuendoes, but we acknowledge other approaches that may offer advantages either to others or in specific situations. We believe strongly that bariatric surgeons should have both approaches in their armamentarium.

Roux-en-Y gastric bypass, and most other bariatric procedures, can be accomplished via minimal access techniques by experienced laparoscopic surgeons. These laparoscopic approaches, however, are very challenging technically, and the patients (and the topic of bariatric surgery) are "high risk." We tend to choose a primary open approach in very large patients (body mass index [BMI] >55), most reoperative bariatric procedures, and those patients known to have multiple adhesions. We also convert readily from a laparoscopic to an open approach if visibility is not satisfactory, if there is a question about too much tension at the gastrojejunostomy, or if there is concern about the safety of any anastomosis related to the minimal access approach.

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Open RYGB

After careful positioning to avoid hyperextension or traction-type injury if one or both arms need to be abducted, an upper midline incision is made beginning 2 cm caudal to xiphoid and ending 2 cm rostral to umbilicus. Avoid extending the incision to the xiphoid—it does not increase exposure, complicates fascial closure, and may predispose to ectopic bone formation at the rostral end of fascial closure. Blunt lateral retraction ("tearing apart" of the wound) minimizes bleeding and separates the fat exactly in the midline. Careful incision of the linea alba without exposure of rectus abdominis muscle facilitates later closure. The peritoneum is entered to the left of the midline in the avascular plane where the preperitoneal fat joins the left posterior rectus abdominis fascia. Exploration excludes gallstones (but the gallbladder is removed routinely anyway—there is a 30% chance of synchronous gallstones, 30% of metachronous development of gallstones with early rapid weight loss), umbilical hernia defect (20–30% prevalence), adnexal pathology, and colon cancer

(somewhat higher incidence in morbid obesity). Some form of mechanical retractor is a must; we prefer the Pilling Bariatric Retractor (Pilling Surgical, Horsham, PA, USA).

Creation of Proximal Pouch of Cardia

The blade of the notched liver retractor should be positioned just anterior to the phrenoesophageal ligament to retract the left lobe of liver rostrally; this maneuver exposes the fat pad of the esophagogastric junction and the underlying cardia. There is almost never a need to take down the triangular ligament to the left hepatic lobe, and for a very fatty left lateral sector, it will not aid exposure because you cannot fold a fatty liver over to the right anyway.

Next, the left esophagogastric junction (angle of His) at the left crura is exposed by sharply dissecting the fat pad off the underlying cardia (Fig. 1a); cautery dissection fully under vision prevents making the proximal pouch too big by including gastric fundus. A reliably "avascular" area of about 4 cm along the greater curvature extends from the esophagogastric junction down to the first short gastric vessel. A 32-Fr orogastric tube aids markedly in this part of the dissection.

Next, the lesser omentum (gastrohepatic ligament) is opened in the bare area while carefully avoiding the more rostrally located hepatic branches of the vagus nerve and/or a replaced left hepatic artery. The surgeon's left hand then enters the lesser sac distal to the left gastric artery (Fig. 1b). Any congenital adhesions to the anterior surface of the retrogastric peritoneum are transected bluntly (if feasible or safe) or sharply either via the lesser omental window or by exposure via opening the gastrocolic ligament—note, the retrocolic, antegastric Roux-en-Y limb to be created will require a window through the gastrocolic ligament anyway (see below). Next, the index finger of the surgeon's left hand is placed behind the stomach through the defect in the lesser omentum and feels for the his/her own right index finger placed anterior to the stomach at the angle of His; there is usually a thin, avascular veil located posteriorly (not laterally), requiring blunt avulsion to permit the fingers to touch one another. Two separate, 18-Fr Silicone® bladder catheters (with balloon-inflating side arms cut off) are passed from the left esophagogastric junction behind the stomach and out to the lesser omental window with the catheter end at the lesser curvature (Fig. 2a). We specifically avoid a clamp for this maneuver but rather use a manual technique.[1] Caudal retraction of the first catheter aids placement of the second one. Next, a 1-cm window into the lesser sac right at the edge of the lesser curvature of the stomach ~2 cm (no further) distal to the esophagogastric junction is made (usually bluntly; on occasion, one small vessel requires ligation). The catheter ends are repositioned from the previous defect in the lesser omentum to be brought out at this lesser curvature window, thereby excluding the neurovascular bundle along the lesser curvature from the gastric cardia around which the catheters encircle (Fig. 2b). The linear anvils of two 90-mm linear staplers are docked with the Silicone® catheters, which then guide the anvils of the staplers posteriorly and around the cardia. The proximal stapler is then oriented as vertically as possible (~45% with the longitudinal axis of the esophagus) and tilted inferiorly to allow more anterior wall of cardia and less posterior wall (the cardiojejunostomy anastomosis will be made to use this larger anterior wall) (Fig. 3). The pouch size should be kept as small as possible (usually <10 ml), just large enough to allow a 21 or 25-mm circular stapled anastomosis. The distal stapler is approximated to the proximal one, both are fired, and the cardia between is transected, thereby creating a "disconnected" anatomy. Two separate staplers markedly facilitate

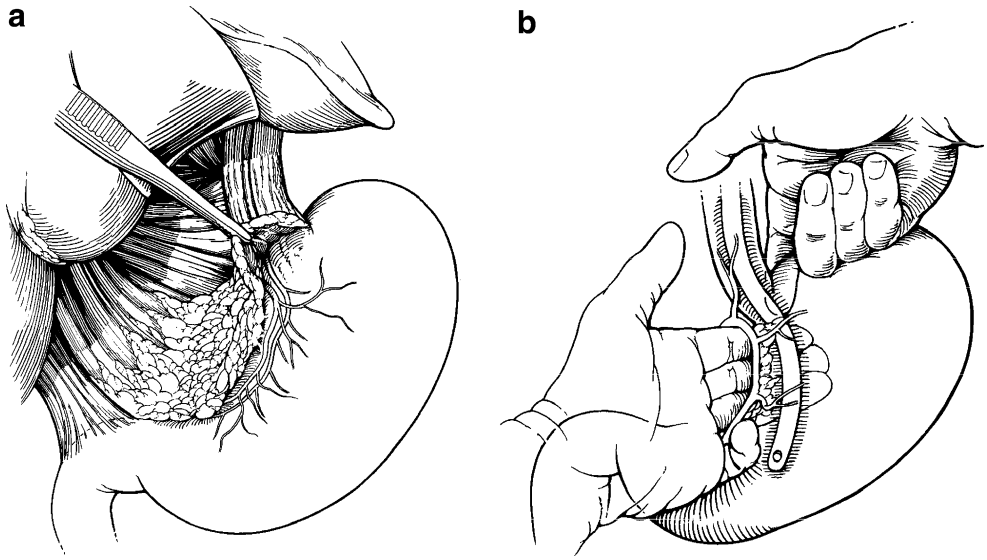


Figure 1 Abdomen open, left lobe of liver retracted rostrally. **a** Fat pad elevated off the cardia exposing the esophagogastric junction, the angle of His, and the left crux of diaphragm. **b** Fingers of left hand in lesser sac posterior to stomach meet index finger of right hand passed behind cardia at angle of His.

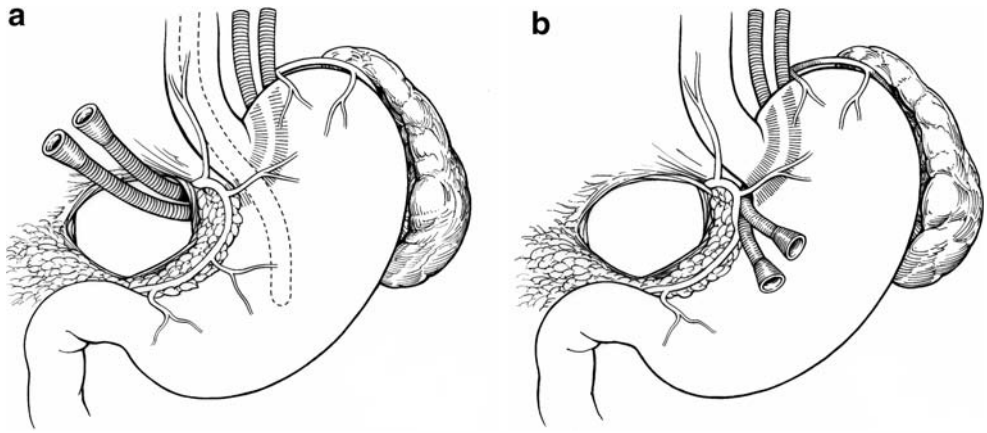


Figure 2 Placement of catheters to guide stapler. **a** Two catheters passing through defect in lesser omentum, behind stomach, and exiting at angle of His. **b** Catheters replaced between neurovascular bundle and lesser curvature of stomach.

this procedure; it is quite difficult to fire one stapler, remove it, and fire another distal to the first staple line angling the distal stapler in a similar fashion. We also oversew the proximal staple line distal to the staple line with a running absorbable suture.

Creation of Roux-en-Y Limb

Next, a Roux-en-Y limb is created from the proximal jejunum, usually fully 50–70 cm distal to the ligament of Treitz. Positioning the overhead light at the head of the bed and shining it through the jejunal mesentery (transillumination) facilitates visualization of the mesenteric vessels. By making the Roux limb further from the ligament of Treitz, a long mesenteric transection is possible in this region, allowing a long, dominant, mesenteric artery and vein to supply the most distal aspect of the Roux limb (Fig. 4). Usually, one arcade needs to be ligated; we always suture-ligate this small arcade to prevent a simple tie from being pulled off while transpositioning the Roux limb through the mesocolon. The jejunum 4–5 cm proximal to the highest point on the Roux limb is transected with a gastrointestinal stapler, and a marking suture is placed on the end of the Roux limb to assure that the correct end of the transected jejunum is always brought up for the cardiojejunostomy. Making the Roux limb closer to the ligament of Treitz minimizes Roux mobility (the mesentery here is short) and often requires ligation of multiple mesenteric vessels, thereby decreasing the effective blood supply to the end of the Roux limb.

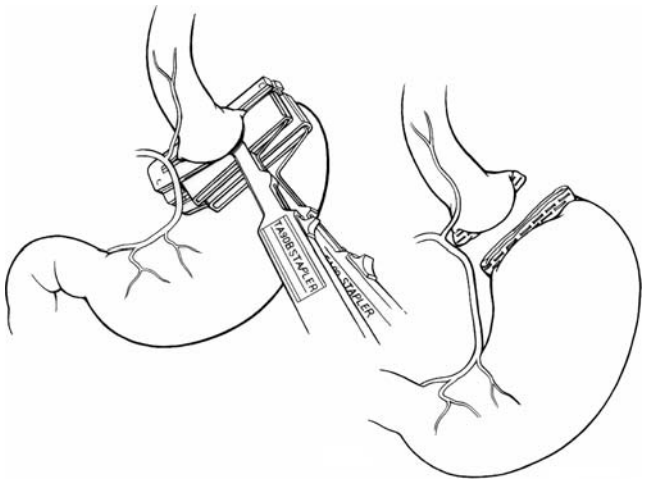


Figure 3 Gastric partitioning. Left panel shows positioning of linear staplers angled to exaggerate anterior wall of cardia and excluding all of the fundus. Right panel shows divided stomach; note mild overhand of anterior wall of proximal pouch of cardia.

Next, a window is made in the gastrocolic ligament in about the midline, and with the transverse colon retracted anteriorly, the surgeon's left hand is passed into the lesser sac. A defect in the avascular part of the mesocolon just to the left of the middle colic vessels (Fig. 4b) is made bluntly and widened, being careful not to disrupt the mesenteric arcade to the transverse colon; no vessel requires ligation. The fingers of the surgeon's left hand grasp the Roux limb, bringing it retrocolic and then antegastric up toward the proximal pouch of cardia. Bringing an extra 12" of Roux limb through the mesocolic defect provides an additional few inches of "length", because a bit more of the Roux mesentery is brought through the mesocolic defect. On rare occasions (a very fat or foreshortened jejunal mesentery), there may be tension; in this situation, usually multiple transverse relaxing incisions through just the peritoneum on both sides of the mesentery of the Roux limb will gain ~2 cm. If necessary (just once in over 1,000 RYGB by the senior author), the entire small bowel mesentery can be mobilized rostrally from the retroperitoneum beginning at the distal ileum/cecum, allowing the entire mesentery to fold anterorostrally.

The Cardiojejunostomy

The Roux limb is draped over the proximal pouch, the highest point (least tension) is identified, and a small cautery mark is made on the antimesenteric serosal surface to identify the site for the anastomosis (usually 5–10 cm

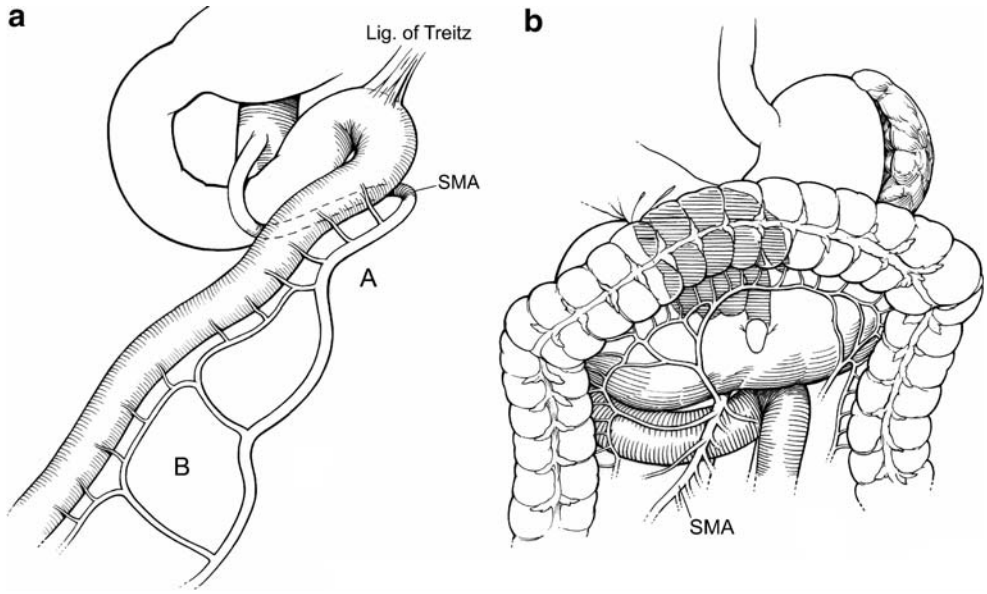


Figure 4 Creation/passage of Roux limb. **a** Anatomy of blood supply to proximal jejunum. Note the close short branches off superior mesenteric artery to jejunum just distal to ligament of Treitz at subpanel **a**, but the longer branches at subpanel **b** 40–60 cm distally. **b** Creation of retrocolic tunnel. Left hand is passed through gastrocolic ligament into lesser sac; index finger identifies appropriate avascular site in mesocolon.

from the stapled end of the Roux limb). Before creating the anastomosis, the small mesenteric vessels to the redundant end of the Roux limb are ligated; it is much easier to do so before the cardiojejunostomy anastomosis fixes the distal end of the Roux limb at the anastomosis.

Now, a 1-cm cardiotomy is made in the anterior wall of the proximal pouch at least 2 cm proximal to the distal staple line, and the circular anvil of a 21-mm, intraluminal circular stapler is positioned bluntly into the proximal gastric pouch. A 2-0 polypropylene pursestring suture obliterates the cardiotomy tight onto the stem of the anvil (Fig. 5a). Careful inverting bites of the pursestring suture, incorporating only 2 mm of the wall of the cardia at the edge of the cardiotomy will ensure a small enough "donut" of proximal cardia such that it will fit inside the 21-mm, circular knife blade of the cartridge end. Next, an enterotomy in the end of the Roux limb allows the cartridge end of the stapler (we prefer the curved circular stapler made by Autosuture Corporation, Norwalk, CT, USA) to be inserted into the Roux limb, advanced to the serosal cautery mark at the highest point of

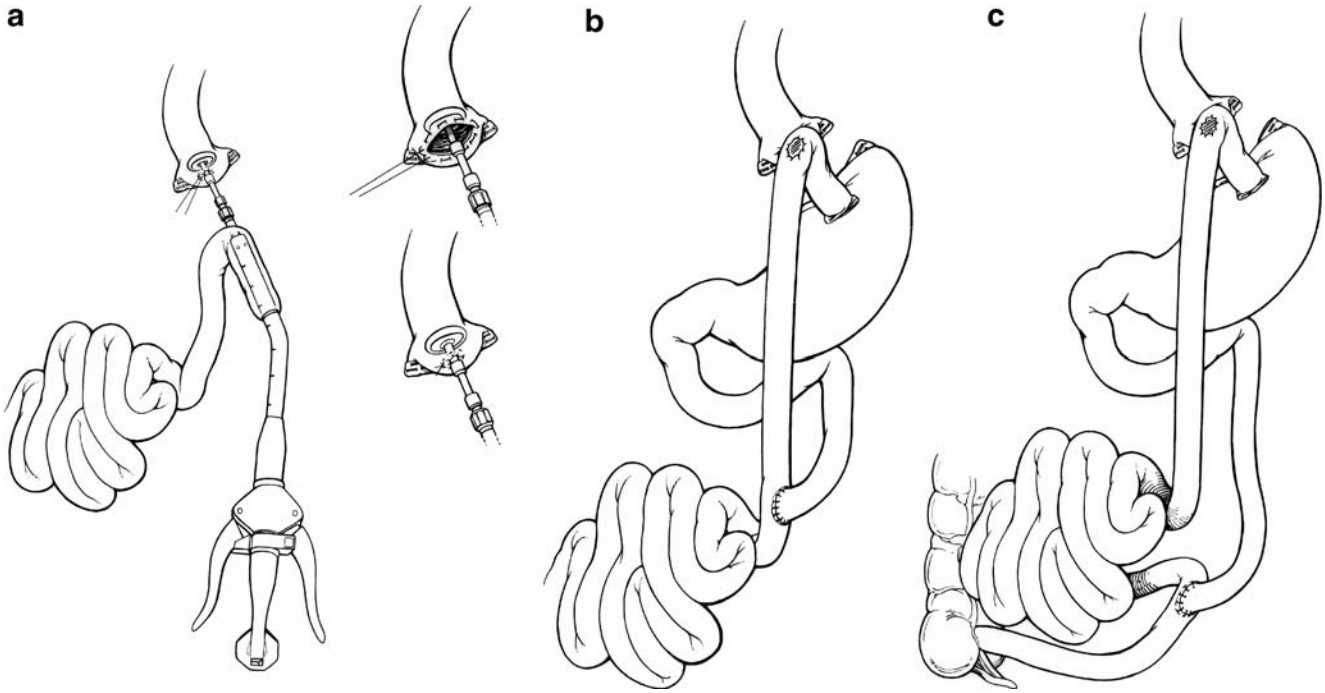


Figure 5 **a** Cardiojejunostomy. The circular stapler is passed into the distal end of the Roux limb; the anvil is placed into the pouch of cardia through a cardiotomy, which is then closed with a purse string suture (inserts). **b** Regular RYGB 150-cm Roux limb. **c** Very, very long limb RYGB with a jejunoileostomy and a 100-cm common channel.

the Roux limb, and the sharp end of the trocar advanced through the Roux limb at this antimesenteric mark. The sharp trocar is removed, the cartridge end is docked with the stem (Fig. 5a), and the stapler is fired, creating the anastomosis. Both donuts should be inspected carefully as should the anastomosis. Any defects should be approximated with transmural sutures. Next, the anastomosis is oversewn circumferentially with interrupted, seromuscular Lembert sutures. The redundant, distal end of the Roux limb is stapled and resected, finishing the proximal anastomosis.

The Enteroenterostomy

The site of the enteroenterostomy depends on the patient's BMI, comorbidities, and whether the procedure is a reoperation for a failed but anatomically intact previous bariatric operation. Our standard RYGB incorporates a 150-cm Roux limb, whereas a malabsorptive RYGB, which we call a very, very long limb RYGB,[2] a form of biliopancreatic diversion, involves a jejunoileostomy 100 cm proximal to the ileocecal junction. This enteroenterostomy is created in end-to-side fashion either via a two-layer, handsewn anastomosis or via a stapled technique. The anastomosis is created on the patient's left lateral side of the Roux limb such that the entire Roux limb lies to the patient's right side of the abdomen (Fig. 5b, c); this prevents kinking/twisting of either the bowel or the mesentery.

Closure of Mesenteric Defects

Three specific mesenteric defects require closure to prevent internal hernias. We use interrupted stitches with a permanent suture material to close these defects. The first is at the enteroenterostomy between the mesentery of the proximal jejunum and the mesentery of the Roux limb (Fig. 6). The second involves obliterating the defect in the transverse mesocolon after pulling any redundancy in the supracolic Roux limb below the mesocolon. The third defect is Petersen's hernia,[3] the previously underappreciated, infracolic defect/space between the mesentery of the Roux limb and the peritoneum overlying the retroperitoneum at the base of the transverse mesocolon.

Adjunctive Concerns

We place a tube gastrostomy routinely for patients undergoing a malabsorptive procedure, because there is a higher risk of complications, and the bypassed stomach offers another enteral avenue for nutritional support if needed. The abdomen is then closed with a running no. 1 braided nylon suture, but we acknowledge that we have no good data to support a running vs an interrupted technique, permanent vs long-acting absorbable suture material, or the use of any other type of bioprosthesis to decrease the 15–20% incidence of incisional hernia.

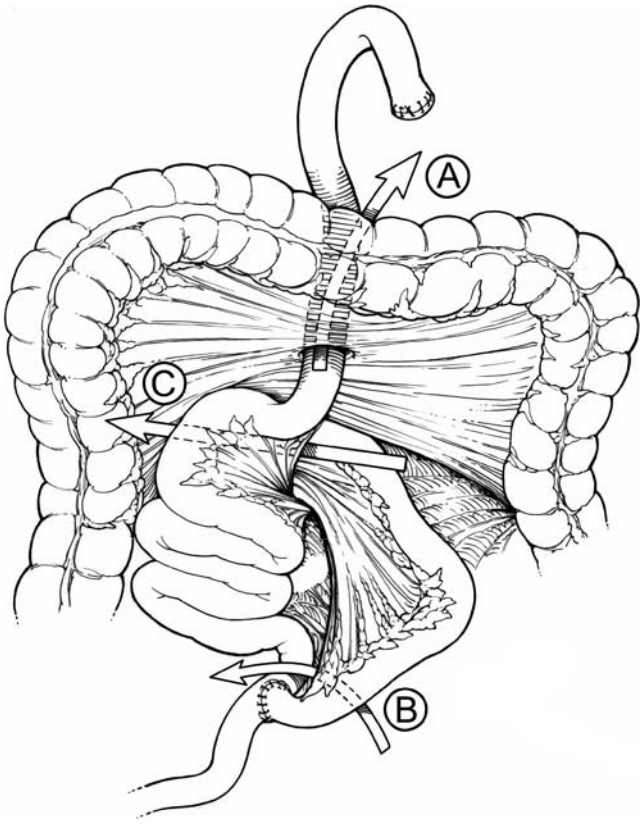


Figure 6 Potential internal hernia defects. **a** Transverse mesocolon. **b** Mesenteric defect at enteroenterostomy. **c** Peterson's infracolic defect.

Laparoscopic Approach

The laparoscopic RYGB was first described in 1994 by Wittgrove et al.[4] The authors developed a technique for a retrocolic, retrogastric RYGB using a 21-mm circular stapler for the gastrojejunostomy. Many different approaches to laparoscopic RYGB were described since. The primary variations of the operation include the method of performing the anastomosis and the anatomic route of passage of the Roux limb. Anastomotic techniques include a circular stapled anastomosis, a linear stapled anastomosis, or a handsewn anastomosis. The Roux limb can be passed retrocolic and antegastric as described above for open RYGB, retrogastric and retrocolic, or most recently antegastric and antecolic. All approaches have had demonstrated success.[3,5] There was an increased incidence of internal hernia formation after the retrocolic approach, because this defect and especially Petersen's defect are harder to close technically from a laparoscopic approach.[2] We prefer an antecolic, antegastric anastomosis using a linear stapling

technique fashioned over a 30-Fr endoscope. It is important, however, to be familiar with all the above techniques because in some patients it is necessary to use these technical modifications when complications arise. It may be necessary to use a retrocolic approach if the mesentery is very short, or it maybe helpful to use a circularly stapled anastomosis when an unusually small gastric pouch is created. In the case of a stapler misfire or for revisional surgery, intracorporeal suturing techniques may be required.

Setup and Positioning

The patient is positioned supine on the operating table with the arms positioned outward on arm boards; however, there is some advantage to having the right arm tucked in terms of access to the patient by the surgeon. The patient should be secured adequately on the operating table with a footboard in place with soft gel or foam padding beneath the feet to prevent pressure ulceration or necrosis. We use a self-retaining, liver retractor secured along the right side of the table. In patients with extreme obesity, this retractor may need to be moved up higher toward the rib cage, as the hips of the patient may actually be beyond the limits of the table and prevent placement of the retractor at a more caudal site. We recommend the use of a table rated to support at least 800 lb and capable of steep, reverse Trendelenburg. Before induction of anesthesia, pneumatic compression devices are applied to the lower extremities of the patient. Antibiotics (cefoxitin 2 g intravenously and either low molecular weight heparin (40 mg subcutaneously) or 5,000–10,000 U of heparin subcutaneously are administered before induction of anesthesia.

The surgeon stands on the patient's right side, the assistant on the patient's left, and the camera operator on the right or left side of the patient, depending on which part of the operation is being done. Monitors are positioned at each shoulder height. We prefer to operate in an Endosuite™ (Stryker Endoscopy, San Jose, CA, USA) with four monitors in place and laparoscopic equipment supported on ceiling-mounted booms. This setup improves operating room ergonomics with fewer cables and wires along the floor to minimize tripping and damage to equipment.

Access and Exposure

We obtain access to the abdomen using an extra long, 15-cm Veress needle (Ethicon Endo-Surgery, Inc., Cincinnati, OH, USA). Although many other options exist for gaining access to the peritoneal cavity, we prefer the left upper quadrant, midclavicular line just below the left costal margin. Once the pneumoperitoneum is established, we then place a 5-mm trocar at this site. In the patient who has had previous upper abdominal surgery, we use an open,

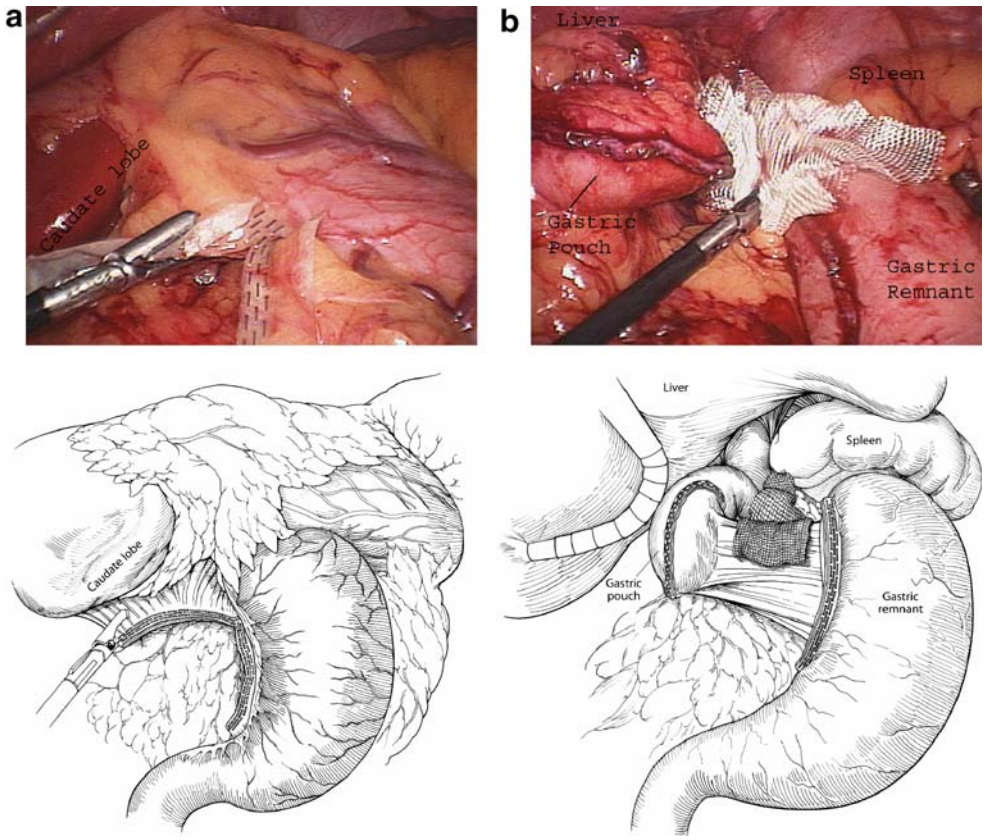


Figure 7 Creation of pouch. **a** Endostapler transection of neurovascular supply along the lesser curvature of stomach; arrows point to bovine pericardial reinforcement of 2.5-mm staples. **b** Final creation of proximal gastric pouch. The bypassed stomach is seen. The Surgicel® helps to minimize oozing from staple lines.

Hasson technique to obtain access; typically, we do this on the right lateral abdomen, 4–5 cm rostral to the umbilicus, so that we can use the port as one of the two working ports. We use a total of six ports for the procedure. Once the pneumoperitoneum is established using the Veress needle, we place a 5-mm bladed port at this site. A camera port (11-mm trocar) is placed about 15–20 cm below the xiphoid just to the left of the midline under vision using a 5-mm laparoscope. We then switch to a 10-mm, 45° laparoscope and place the patient in steep Trendelenburg position. Two working ports are then placed on the patient's right; one is placed just subcostal about the midclavicular line (5-mm trocar) and another just medial to the midclavicular line and slightly rostral to the camera port (12-mm trocar). In some patients, it is necessary to angulate the ports, although in most, it is better to insert them perpendicularly to the fascia. An additional right flank port (5-mm trocar) for the liver retractor is placed and so is a 5-mm trocar in the left flank. The operative procedure is divided into three parts: creation of the gastric pouch, the enteroenterostomy, and the gastrojejunostomy. We perform the procedure in this order, although in some instances there may be an advantage to performing the enteroenterostomy first.

Creation of the Gastric Pouch

The patient is placed in steep, reverse Trendelenburg position. The harmonic scalpel is used to incise the avascular portion of the lesser omentum; the caudate lobe can be identified through a thin, gastrohepatic ligament. Generally speaking, from this dissection point it is easy to identify the location of the left gastric artery. A replaced or accessory left hepatic artery, if present, should be evident just proximal to the thin part of the gastrocolic ligament; care should be taken to not injure this vessel, especially as it is not necessary to go this high up (rostral) in the dissection.

We begin by transecting the neurovascular fat along the lesser curvature (including the vagal nerves) using a 60-mm linear endostapler (AutoSuture Corporation) with a white "vascular" cartridge of 2.5-mm staples; bleeding from the staple lines is reduced using reinforcements such as bovine pericardium (Fig. 7a). The stapler is oriented transversely approximately 2 cm distal to the gastroesophageal junction distal to the origin of the left gastric artery. We have not found any adverse sequelae to this technique of transecting the neurovascular pedicle along the lesser curvature. A small, vertically oriented, lesser curvature gastric pouch, approximately 15–20 ml in size, is created using blue cartridges with staples of 3.5 mm in height. Initially, we sized our pouch with a 20-ml balloon; however, this technique was not necessary in our last several hundred cases. The first application of the endostapler is oriented transversely, after which subsequent applications are carried up to the angle of His parallel to the lesser curvature (Fig. 7b). Care is taken to avoid incorporation of any fundus in the pouch and to ensure complete division of the stomach; this approach avoids worry of a retained gastro-gastric communication. Troublesome bleeding on the gastric pouch side is sometimes found; however, generally, this can be controlled easily using a piece of topical hemostatic agent, such as Surgicel® (Johnson & Johnson, Brunswick, NJ, USA). Any defects found in the staple lines are closed either using intracorporeal suturing techniques with braided nylon suture or by using the Endostitch® device (AutoSuture Corporation). It is important to repair corresponding defects in the staple lines in both the gastric pouch and the gastric remnant. Staple line leaks from the gastric remnant are notoriously difficult to diagnose and can lead to morbidity and mortality during the postoperative period. There are often multiple, "congenital" adhesions posterior to the gastric remnant, which usually do not need to be taken down, because we perform an antecolic,

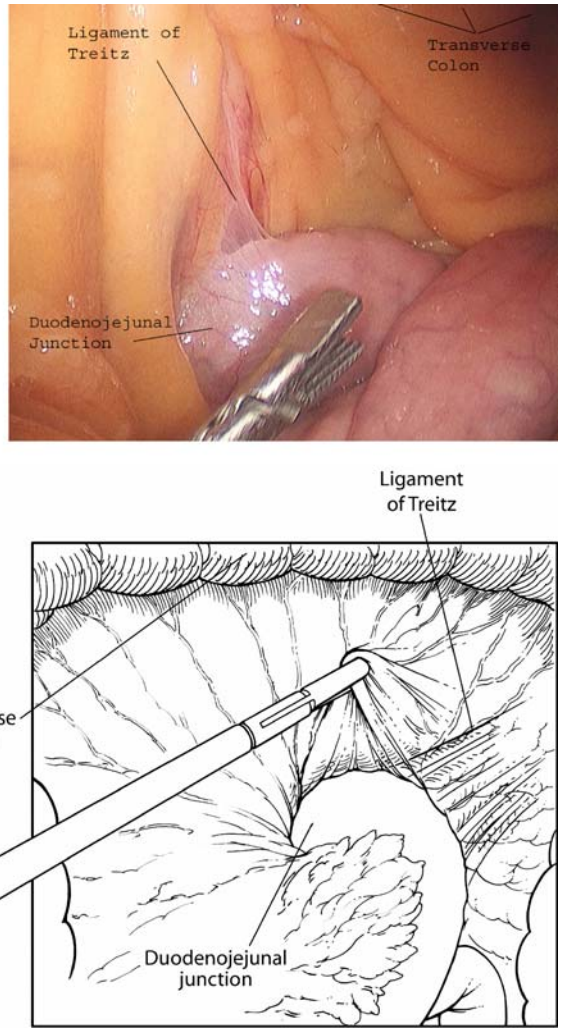


Figure 8 By grasping and elevating the left transverse mesocolon, the proximal jejunum at the ligament of Treitz is seen (arrow).

antegastric Roux limb. If retrogastric passage of the Roux limb is anticipated for a posterior gastrojejunostomy, then these adhesions must be lysed, and a 1-cm area from the posterior aspect of the proximal gastric pouch for the gastrojejunal anastomosis must be cleared.

Special considerations are needed when a large sliding hiatus or paraesophageal hernia is encountered. In these patients, during the mobilization and reduction of the hernia, the vascular pedicle (blood supply) to the proximal lesser curvature of the stomach arising from the left gastric artery must be preserved. Once the crura were approximated, the pouch can be formed. If there are any concerns, conversion to an open procedure is encouraged.

Creation of the Enteroenterostomy

The operating table is made horizontal. The transverse colon is elevated rostrally, and the omentum is flipped over the transverse colon to expose the left transverse mesocolon. The area above the ligament of Treitz is grasped, and the proximal jejunum at the ligament of Treitz is identified clearly (Fig. 8). Great care must be taken to identify confidently the relevant anatomic landmarks before proceeding with jejunal division. The jejunal mesentery is inspected at this time. If the mesentery is foreshortened, as is often the case in patients with central, "truncal" obesity, it may be best to perform the procedure placing the Roux limb in a retrocolic fashion. We divide the jejunum 50 cm distal to the ligament of Treitz using a double linear cutting stapler. We originally used a bowel grasper with a 10 cm marker to measure bowel length, but we are now comfortable estimating bowel length. A white staple load 60 mm in length is used to divide the jejunum. We use a gray staple cartridge with 2.0-mm staple height for dividing the mesentery. One application of the stapler will suffice for a retrocolic Roux limb; however, two staple loads are generally needed for an antecolic Roux limb, especially with a thicker mesentery. A Penrose drain is sewn to the proximal end of the Roux limb using the Endostitch® device.

Next, the Roux limb is measured. We prefer a Roux limb of 75 cm for patients with a BMI <50 kg/m^2 and 150 cm for patients with a BMI of 50–60 kg/m^2 and for those with insulin-resistant diabetes mellitus regardless of BMI. For the superobese (BMI >60), we make the Roux limb 200–250 cm. After measuring the Roux limb, the proximal end of the Roux limb (with attached Penrose drain) is positioned such that it points rostrally to the midtransverse colon, and the mesentery of the Roux limb points to the left side of the abdomen. The Roux limb thus lies in a "C"

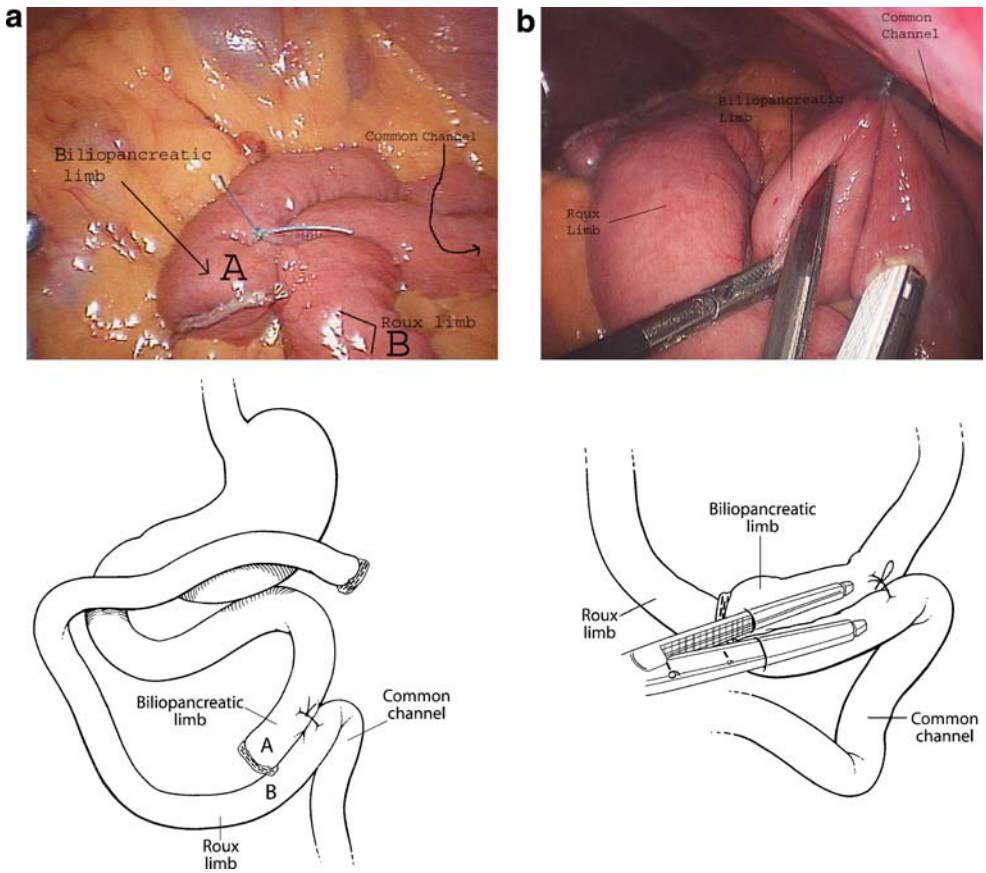
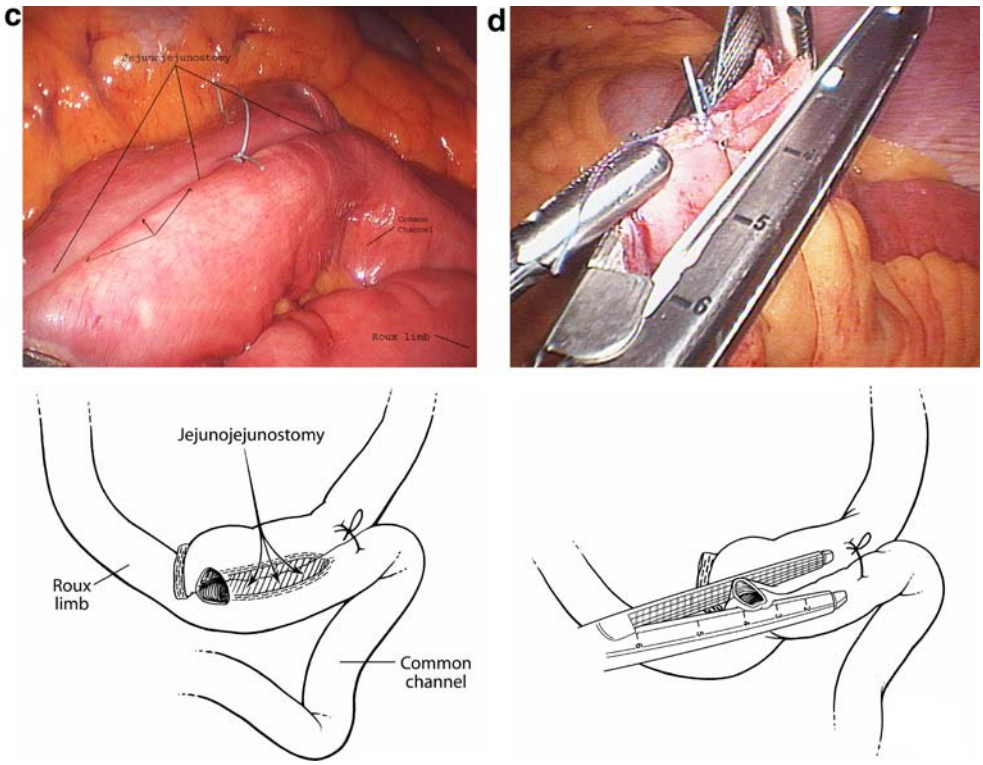


Figure 9 Enteroenterostomy. **a** Approximation of biliopancreatic limb with Roux limb. **b** Enterotomies in biliopancreatic limb (apex of staple line excised) and Roux limb with stapler being inserted. **c** Side-to-side anastomosis completed. **d** Transverse closure of the now common enterotomy also with an endostapler.

Figure 9 (continued)



shaped position. The more distal part of the Roux limb is allowed to fall into the right lower quadrant so that it does not interfere with access to the biliopancreatic limb. Next, at the site for the planned enteroenterostomy, we sew the distal (stapled) end of the proximal jejunum (biliopancreatic limb) to the Roux limb at their antimesenteric borders to allow a functional end-to-side stapled anastomosis (Fig. 9a). The harmonic scalpel is used to make an enterotomy in the Roux limb and in the proximal jejunum of the biliopancreatic limb. For the biliopancreatic limb, we

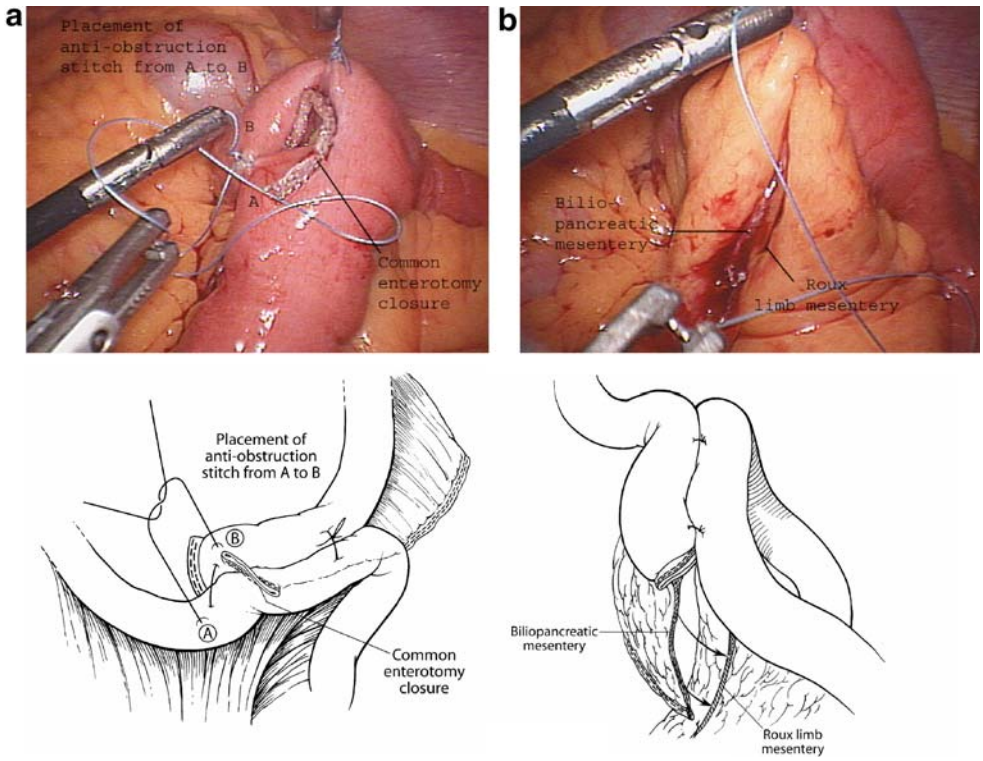
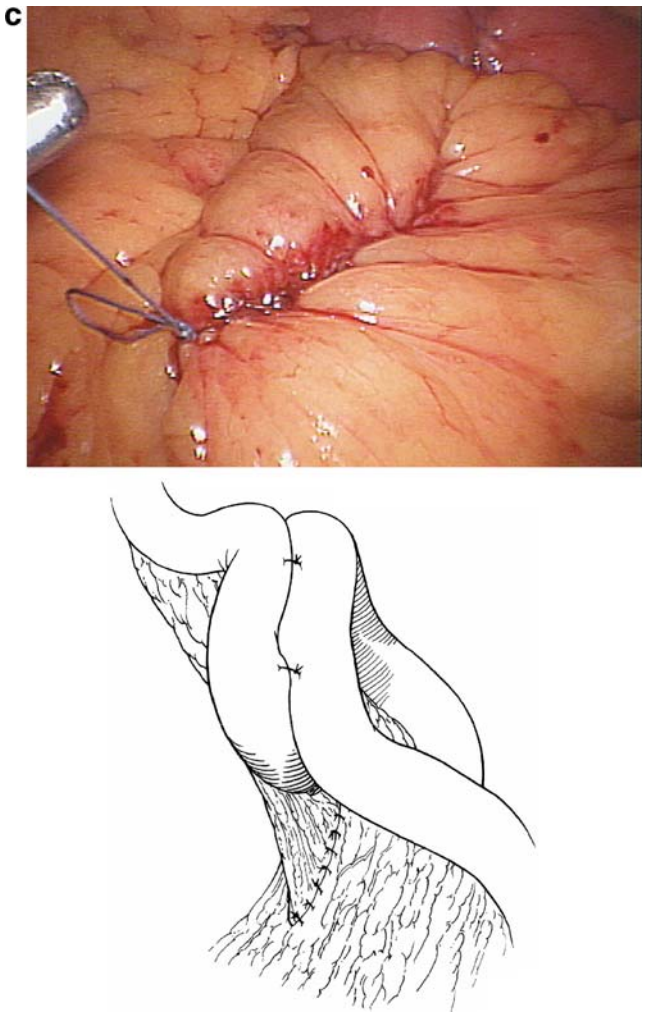


Figure 10 **a** The antiobstruction stitch placed from the distal end of the biliopancreatic limb (**a**) to the Roux limb (**b**) at the anastomosis. **b** Closure of the mesenteric defect at the jejunojejunostomy to prevent an internal hernia. **c** Closure of mesenteric defect at jejunojejunostomy.

Figure 10 (continued)



excise the top corner of the staple line; for the Roux limb, however, it is important to make the enterotomy antimesenteric and approximately 1 cm or so proximal to the corresponding enterotomy on the biliopancreatic limb (Fig. 9b). Using a 60-mm white cartridge load, the double linear cutting stapler is inserted to its full length and fired (Fig. 9c); we also place one suture of braided nylon to reinforce the heel of the anastomosis. Next, at the proximal aspect of the staple line, we approximate the edges of the now common enterotomy with an Endostitch®, rotate the enterotomy of the jejunojejunostomy by 90° so that the enterotomy points to the left upper quadrant, and close it with an additional firing of the endostapler using a 60-mm white load (Fig. 9d); care must be taken to minimize the amount of bowel in the staple line such that one limb of the bowel is not narrowed. In some situations, such as a more distal anastomosis with smaller caliber bowel, it may be preferential to sew the enterotomy closed with braided nylon. We inspect carefully all the staple lines to make sure that no mucosa is visible and that there is no separation of the serosa. If there is any questionable area, we imbricate the area carefully by suturing the serosal edges together with a Lembert-type stitch. Fibrin glue is then applied to the staple lines to minimize adhesions and bleeding.

The final step of the enteroenterostomy is to close the mesenteric defect. We begin with an "anti-obstruction stitch"[6] from the Roux limb to the biliopancreatic limb to prevent kinking (Fig. 10a). Next, the mesenteric defect is closed with a running nylon suture by carefully opposing the transected edge of the mesentery of the biliopancreatic limb to the mesentery of the Roux limb (Fig. 10b, c).

Creation of Gastrojejunostomy

Using the harmonic scalpel and beginning at the midpoint of the transverse colon, the greater omentum is split vertically into two halves to allow ease of passage of the Roux limb to the upper abdomen (Fig. 11a). After grasping the Penrose drain and making certain there is no mesenteric twist, the Roux limb is pushed up to the gastric pouch in an antecolic position. This maneuver is best done with the patient out of reverse Trendelenburg to maximize mobility.

If it appears that there will be undue tension on the anastomosis, then a retrocolic, retrogastric passage should be planned. Before beginning creation of a defect in the transverse mesocolon, any remaining retrogastric adhesions are lysed using the harmonic scalpel by opening the gastrocolic ligament, grasping and retracting the stomach anteriorly, and visualizing these adhesions. Then, the mesentery of the transverse colon 2–3 cm anterior to the ligament of Treitz is grasped and incised using the harmonic scalpel. The stomach is identified and pulled to the left, and the Penrose drain is passed into the lesser sac using an articulating grasper. After completion of the procedure, this mesenteric defect must be closed very carefully using a permanent suture after reduction of redundant Roux limb with additional attention placed on closing Petersen's defect (see "Open RYGB" section). Any twist of the Roux limb below the gastric remnant must be excluded.

In both the antecolic and retrocolic/retrogastric approaches after the Roux limb is brought up to the proximal gastric pouch, the patient is next placed slowly in a steep, reverse Trendelenburg position. The gastrojejunostomy is constructed in three layers. Posteriorly, there is a handsewn seromuscular layer between the pouch and the Roux limb. In the middle, there is a stapled gastrojejunostomy and a common gastroenterotomy which is sewn closed. For the third layer, the entire pouch staple line, the gastroenterotomy closure site, and the stapled gastrojejunostomy are oversewn with a final suture line of seromuscular sutures. We begin this anastomosis with a back row of running, seromuscular sutures using 3-0 braided nylon. The suture line is started at the angle of

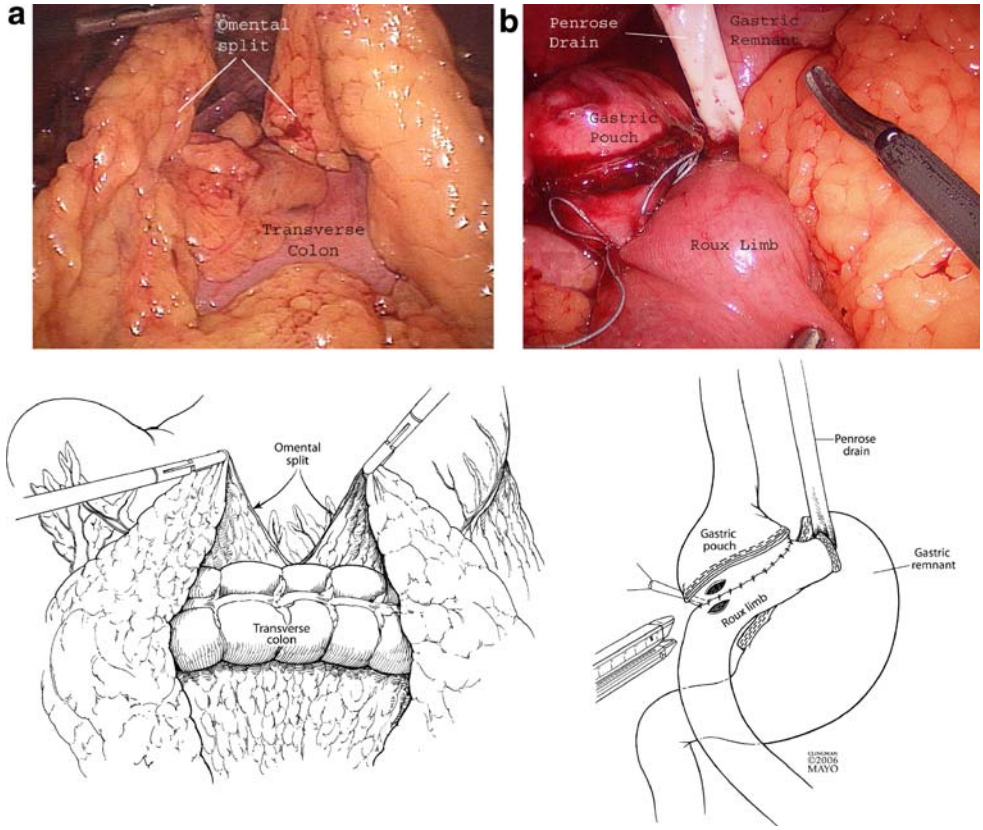
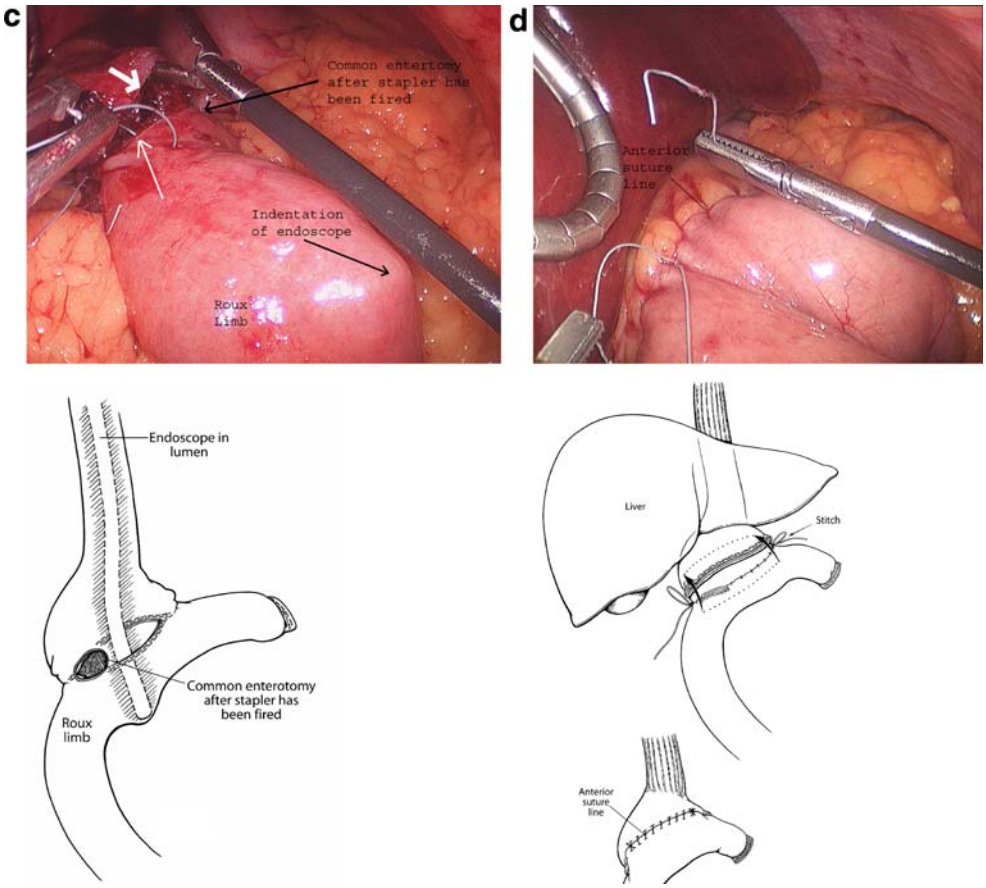


Figure 11 Gastrojejunostomy. **a** The greater omentum is "split" in its midportion to allow the Roux limb to pass antecolic up to the gastric pouch. **b** The anastomosis begins by approximating the posterior aspect of the proximal gastric pouch to the end of the Roux limb with a layer of seromuscular sutures. The gastrostomy and jejunotomy are not shown in the intraoperative photo. **c** The "inner" layer of the anastomosis was created with a linear endostapler passed through a gastrostomy and enterotomy, forming a now common gastroenterotomy. The thick white arrow indicates the edge of the gastrostomy of the stomach and the thin arrow is the edge of the enterotomy in the Roux limb. Note the jejunal bulge produced by the end of the luminal endoscope passed per os. **d** The completed gastrojejunostomy, the anterior "outer layer" seromuscular suture line covers the gastrojejunostomy closure and the staple line of the gastric pouch.

Figure 11 (continued)



His at the rostral aspect of the staple line of the gastric pouch with the end of the Roux limb where the Penrose drain is attached. This row of sutures is continued all the way along the posterior aspect of the pouch obliquely and along the Roux limb parallel and close to the mesentery (Fig. 11a, b). A gastrostomy is made at the patient's right inferior corner of the pouch and at a corresponding point on the Roux limb, an enterotomy is made. A linear stapler with a blue cartridge is inserted no more than 1.5 cm and fired, effectively accomplishing the gastrojejunostomy and creating a now common gastroenterotomy (Fig. 11c). We then insert an Olympus 30-Fr endoscope per os through the anastomosis and into the Roux limb as a stent and then oversew the remaining defect in two layers over the endoscope using the Endostitch® device (Fig. 11c), thereby completing the anastomosis (Fig. 11d). After placing a bowel clamp about 5 cm distal to the gastrojejunostomy, air is insufflated via the endoscope into the bowel lumen at the gastrojejunostomy with the anastomosis under saline to look for any leakage or bubbling. Any sites of bubbling must be identified and oversewn, after which the air insufflation maneuver is then repeated. If the site of bubbling either cannot be found or oversewn successfully, the procedure should be converted to an open approach. Finally, for an antecolic approach, any generous Petersen's defect should be closed.

We do not place a drain routinely in the area of the anastomosis; however, for difficult anastomoses, if any bubbling was found at the time of testing the anastomosis that required oversewing or in the patient with supermorbid obesity with comorbid conditions, we will use a drain. We do not use a nasogastric tube. Because we use noncutting, dilating trocars, we typically do not close the fascia for trocar sites above the umbilicus but simply staple the skin incisions. Skin staples are removed before discharge on postoperative day 2 and replaced with Steristrips (3M Corporation, St Paul, MN, USA). We have a very low incidence of wound infection using this approach (less than 2%).

Postoperative Care

Patients undergo a routine, upper gastrointestinal contrast study on postoperative day 1, which is helpful to diagnose leak or bowel obstruction. Most radiographic leaks necessitate return to the operating room for a laparoscopic repair of the site of leak. Clear liquids are started immediately after this point. Patients are discharged home on the second or third postoperative day and asked to visit in 1 week. If there is a drain, it is removed at this time.

References

1. Sarr MG. How I do it: vertical disconnected Roux-en-Y gastric bypass. Dig Surg 1996;13:45–49.
2. Murr MM, Balsiger BM, Kennedy FP, Mai JL, Sarr MG. Malabsorptive procedures for severe obesity: comparison of pancreaticobiliary bypass and very very long limb Roux-en-Y gastric bypass. J Gastrointest Surg 1999;3:607–612.
3. Schauer PR, Ikramuddin S, Hamad G, Eid GM, Mattar S, Cottam D, Ramanathan R, Gourash W. Laparoscopic gastric bypass surgery: current technique. J Laparoendosc Adv Surg Tech, Part A 2003;13:229–239.
4. Wittgrove AC, Clark GW, Tremblay LJ. Laparoscopic gastric bypass, Roux-en-Y: preliminary report of five cases. Obes Surg 1994;4:353–357.
5. Higa KD, Ho T, Boone KB. Laparoscopic Roux-en-Y gastric bypass: technique and 3-year follow-up. J Laparoendosc Adv Surg Tech A 2001;11;377–382.
6. Brolin RE. The antiobstruction stitch in Roux en Y enteroenterostomy. Am J Surg 1995;3:355–357.

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An Ileal Endometrioma: Of Carcinoids and Cadherin

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Abstract A 38-year-old woman with history of prior adrenalectomy for Cushing's syndrome presented with intermittent right lower quadrant (RLQ) abdominal pain, nausea, bloating, and non-bloody diarrhea for 2 months. Symptoms were not related to her menstrual periods. Examination revealed only an ill-defined mass in the RLQ. Investigations for infectious causes, inflammatory bowel disease, and carcinoid tumor were negative. Computed tomography (CT) demonstrated a terminal ileal mass with mesenteric stranding and dilatation of the proximal bowel. At laparotomy, a fibrotic, terminal ileal mass with matted adhesions involving the mesentery and retroperitoneum was resected. Histopathological examination identified multiple foci of endometriosis extending from the serosal surface into the mucosa of the terminal ileum. Immunostaining revealed E- and P-cadherin, but not N-cadherin immunopositivity. Mucosal involvement without cyclical menstrual symptoms and intestinal obstruction is an unusual presentation of intestinal endometriosis. Although the mechanism of endometriosis is not clear, the role of cell adhesion molecules such as cadherins has received attention. Increased expression of E- and P-cadherin and decreased N-cadherin expression in our patient demonstrates differential expression of these cadherins in endometriotic tissue. Future studies may investigate patterns of differential expression of these cadherins in a series of cases to elucidate the mechanisms of migration of endometriotic tissue.

Keywords Endometriosis · Ileum · Cadherin

A 38-year-old nulliparous African-American woman with a history of adrenalectomy for Cushing's syndrome, 9 years ago, presented with right lower quadrant (RLQ) abdominal pain, intermittent nausea, bloating, and non-bloody diarrhea for approximately 2 months. There was no association between the abdominal pain and bowel movements. She denied fever, chills, or weight loss. Her menstrual periods were regular, and her symptoms were not related to her menses. Clinical examination was remarkable only for an ill-defined, mildly tender mass in the RLQ.

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Complete blood count, chemistries, and liver tests were normal. Cultures for *Chlamydia trachomatis* and *Neisseria gonorrhoeae*, a Papanicolaou smear, and a tuberculin skin test showed negative results. A small bowel series demonstrated a mesenteric mass with scarring and infiltration in the RLQ and splaying of the adjacent ileal loops (Fig. 1). Axial computed tomography (CT) scan revealed a terminal ileal mass just proximal to the ileocecal junction associated with proximal dilatation of the small bowel loops. Colonoscopy demonstrated a normal colon and cecum, but attempts to enter the terminal ileum were unsuccessful. Neuroendocrine investigations including adrenal function parameters, chromogranin A, substance P, serotonin, gastrin, and pancreatic polypeptide were normal. An Octreoscan and Meckel's scan were also unremarkable. An exploratory laparotomy was performed for tissue diagnosis and possible excision of the mass. At laparotomy, a fibrotic, intraluminal, terminal ileal mass, measuring approximately 7 cm was identified. The mass was located about 10 cm from the ileocecal valve and displayed matted adhesions involving the mesentery and retroperitoneum.

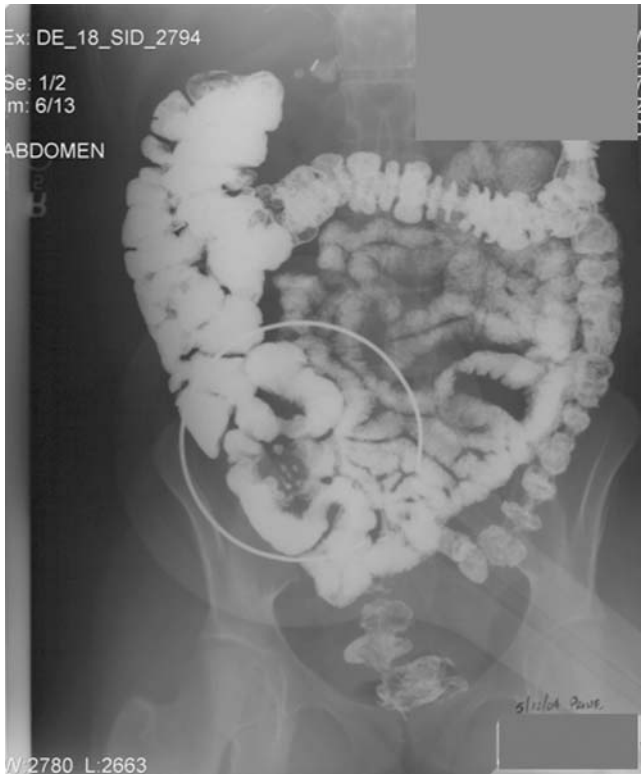

Figure 1 Small bowel series showing a mesenteric mass with scarring, tethering, and infiltration in the right lower quadrant.

The ileal surface exhibited some punctate brown dots, and the ileal mesentery was contracted and adherent to the retroperitoneum. The liver appeared normal. Multiple uterine, ovarian, and fallopian endometrial implants with adhesions were noted in the pelvis. A right hemicolectomy, including the removal of the mass and approximately 30 cm of the terminal ileum, was undertaken.

Gross pathological examination revealed a tan-white irregular ileal mass about 10 cm from the ileocecal valve extending through the serosa into the bowel wall. Histopathological examination demonstrated multiple foci of endometriosis, extending from the serosal surface into the mucosa of the terminal ileum (Fig. 2), and endometriosis in the appendix. Immunostaining of ileal endometriotic tissue sections revealed the presence of E-cadherin in all foci, absence of N-cadherin in all foci, and variable staining of P-cadherin (Fig. 3). All samples were cytokeratin-positive. Postoperatively, the patient had complete resolution of her symptoms. She was prescribed oral contraceptive therapy as long-term management of her pelvic endometriosis.

Discussion

Endometriosis is defined as the presence of endometrial tissue outside the uterine cavity, and such lesions were reported in numerous sites including the fallopian tubes, ovaries, pelvic cavity, and the gastrointestinal tract[1]. In this patient, the preoperative diagnosis of endometriosis was difficult, as abdominal symptoms were not associated with menses. Although uncommon, adrenal tumors may be metachronously or synchronously associated with other neuroendocrine tumors[2]. However, in our patient, neuroendocrine markers and Octreoscan were negative. The diagnostic conundrum, symptoms of subacute intestinal obstruction, and concern for malignancy prompted an explorative laparotomy.

Gastrointestinal endometriosis affects 3–37% of women with endometriosis[1]. In a series of 292 cases of intestinal endometrioisis, the most commonly affected areas were the sigmoid colon and rectum (85%), small bowel (7%), cecum (4%), and the appendix (3%)[3]. In a series of 7,200 cases of endometriosis, small bowel involvement was noted in only 0.5% of cases[4]. Interestingly, ileal endometriosis is most commonly seen within 10 cm of the ileocecal valve, and in this respect, closely resembles the distribution of carcinoid lesions of the ileum[5].

The clinical presentation of intestinal endometriosis can be very similar to disorders such as Crohn's disease, appendicitis, diverticulitis, irritable bowel syndrome, intestinal malignancies, and a host of gynecologic diagnoses[6]. In some patients, the coexistence of these diseases with intestinal endometriosis makes preoperative diagnosis even more challenging[7]. Characteristically, the fibrotic nature of ileal endometriosis may result in both acute and subacute intestinal obstruction, and therefore, may mimic the presentation of an ileal carcinoid[8,9]. Given the propensity to present with obstructive symptomatology, definitive management usually involves surgical resection[10].

On laparotomy, intestinal endometriosis usually appears as subserosal and muscularis propria implants[10]. Mucosal

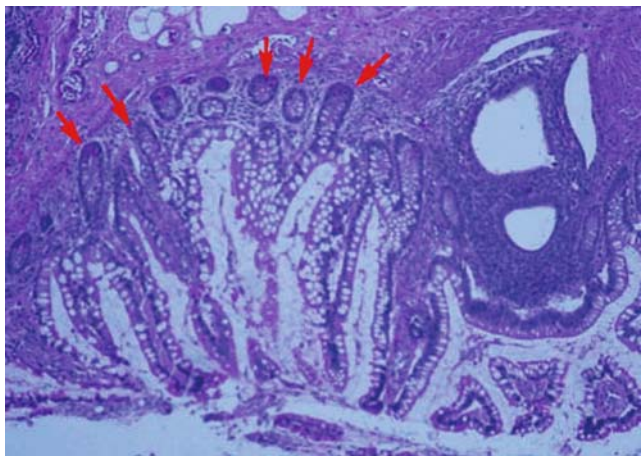

Figure 2 Pathological examination of the resected specimens reveals multiple foci of endometriosis (*red arrows*) extending from the serosal surface focally into the mucosa.

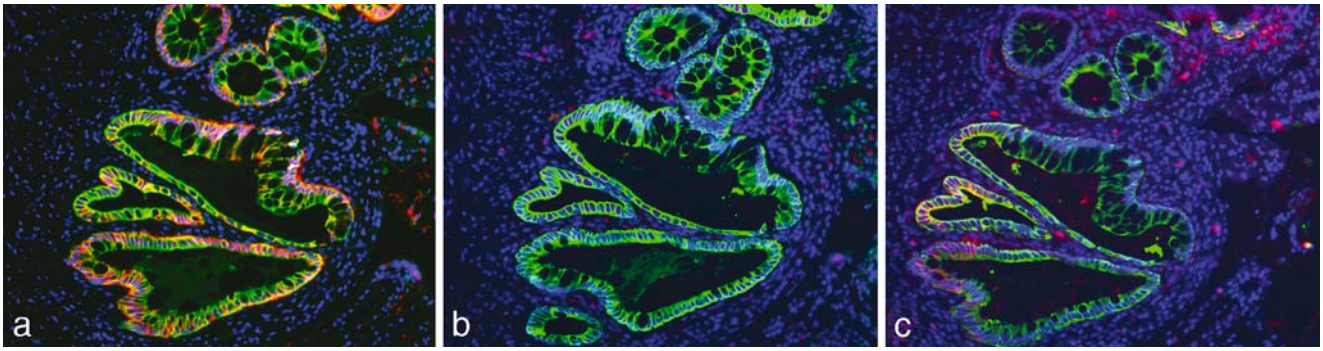

Figure 3 Triple color immunostaining of serial tissue sections reveals significant E-cadherin staining (**A**), no N-cadherin staining (**B**) and occasional P-cadherin staining (**C**). Immunostaining of cadherins was invariably membrane localized. *Blue*—nuclei [4',6-Diamidino-2-phenylindole (DAPI)], *green*—cytokeratin (Alexa488), *red*—cadherin (Cy5). Dual membrane staining (*red and green*) results in *yellow* (×400 magnification).

involvement resulting in acute intestinal obstruction and perforation is very rare[6]. Our patient presented with partial small bowel obstruction from an ileal mass that was clinically and radiographically suspicious for a carcinoid tumor, but at laparotomy, it was identified as ileal endometriosis. This case is unusual for two reasons. Firstly, intestinal endometriosis in this instance presented as a mass lesion mimicking a neuroendocrine tumor (location and topographic studies) in a patient who had already undergone a right adrenalectomy for Cushing's disease. Secondly, pathological studies demonstrated extensive mucosal invasion without intestinal perforation or without any history of cyclical menstrual symptoms.

Of the various hypotheses that attempt to explain the migration of endometrial tissue to extrauterine sites, the implantation theory (retrograde menstruation through the fallopian tubes resulting in the deposition of endometrial tissue in various sites) and the coelomic metaplasia theory (endometrial transformation of peritoneal and other cells) are the most widely accepted[6]. More recently, the putative role of cell adhesion molecules in tumor metastases has prompted interest in their possible role in the migration of endometriotic tissue to extrauterine sites[11]. In particular, attention was directed to the role of E-cadherin, a cellular adhesion molecule that suppresses invasion in carcinomas. Gaetje et al.[12] reported that E-cadherin-negative epithelial cells were increased in number in endometriotic tissue compared to eutopic endometrial tissue. Cell lines derived from peritoneal endometriosis appear to exhibit two types of cells: stromal (cytokeratin- and E-cadherin-negative) and epithelial-like cells (cytokeratin-positive and E-cadherin-negative)[13]. The endometriotic epithelial-like cells were demonstrated to display invasive capabilities similar to cells derived from metastatic carcinomas. Interestingly, these endometriotic cells expressed N-cadherin, a ligand that may be responsible for cell migration, invasion, and metastases[14]. Chen et al.[15] demonstrated that mRNA levels of P-cadherin, a cadherin subtype that may be involved in the progression of cancer, are significantly higher in endometriotic lesions compared to eutopic endometrial tissue. In our patient, endometriotic cells expressing cytokeratin were E-cadherin- (and occasionally P-cadherin-) positive but N-cadherin-negative. This suggests that the role of differential expression of cadherin proteins in endometrial cell migration may be more complex than previously thought. Further elucidation of the respective role(s) of these and other cellular adhesion molecules is necessary to define the pathogenesis of endometriosis and allow prediction of the behavior of such itinerant cell types.

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References

1. Croom RD, Donovan ML, Shwesinger WH. Intestinal endometriosis. Am J Surg 1984;148:660–667.
2. Kaltsas GA, Besser GM, Grossman AB. The diagnosis and medical management of advanced neuroendocrine tumors. Endocr Rev 2004;25:458–511.
3. Macafee CHG, Greer HLH. Intestinal endometriosis: a report of 292 cases and survey of the literature. J Obstet Gynaecol Br Commonw 1960;67:539–555.
4. Martinbeau PW, Pratt JH, Gaffey TA. Small bowel obstruction secondary to endometriosis. Mayo Clin Proc 1975;50:239–243.
5. Kinkel K, Chapron Q, Balleyguier C, Fritel X et al. Magnetic resonance imaging characteristics of deep endometriosis. Hum Reprod 1999;14:1080–1086.
6. Decker D, Konig J, Wardelmann E et al. Terminal ileitis with sealed perforation—A rare complication of intestinal endometriosis: case report and short review of the literature. Archives of Gynecol Obstet 2004;269:294–298.

7. Craninx M, D'Haens G, Cokelaere K et al. Crohn's disease and intestinal endometriosis: an intriguing coexistence. Eur J Gastroenterol Hepatol 2000;12(2):217–221.
8. Wickramasekera D, Hay DJ, Fayz M. Acute small bowel obstruction due to ileal endometriosis: a case report and literature review. J R Coll Surg Edinb 1999;44:59–60.
9. Ray R, Choudhury SR, Bose SM, Kaushik SP. Intestinal endometriosis. Indian J Gastroenterol 1992;11(2):92–93.
10. Ridha JR, Cassaro S. Acute small bowel obstruction secondary to ileal endometriosis: report of a case. Surg Today 2003; 33:944–947.
11. Starzinski-Powitz A, Handrow-Metzmacher H, Kotzian S. The putative role of cell adhesion molecules in endometriosis: can we learn from tumour metastasis? Mol Med Today 1999;5(7): 304–309, Jul.
12. Gaetje R, Kotzian S, Herrmann G et al. Nonmalignant epithelial cells, potentially invasive in human endometriosis, lack the tumor suppressor molecule E-cadherin. Am J Pathol 1997;150(2): 461–467, Feb.
13. Zeitvogel A, Baumann R, Starzinski-Powitz A. Identification of an invasive, N-cadherin-expressing epithelial cell type in endometriosis using a new cell culture model. Am J Pathol 2001;159 (5):1839–1852.
14. Derycke LD, Bracke ME. N-cadherin in the spotlight of cell–cell adhesion, differentiation, embryogenesis, invasion and signalling. Int J Dev Biol 2004;48(5–6):463–476.
15. Chen TCG, Tai CT, Yeh LS et al. Identification of the cadherin subtypes present in the human peritoneum and endometriotic lesions: potential role for P-cadherin in the development of endometriosis. Mol Reprod Dev 2002;62:289–294.