The Long-term Results of a Randomized Clinical Trial of Laparoscopy-assisted Versus Open Surgery for Colon Cancer

Antonio M. Lacy, MD, PhD, Salvadora Delgado, MD, PhD,* Antoni Castells, MD, PhD,† Hubert A. Prins, MD, PhD,* Vicente Arroyo, MD, PhD,† Ainitze Ibarzabal, MD, PhD,* and Josep M. Pique, MD, PhD†*

Objective: The aim of this study was to compare the long-term outcome of laparoscopy-assisted colectomy (LAC) and open colectomy (OC) for nonmetastatic colon cancer.

Methods: From November 1993 to July 1998 all patients with adenocarcinoma of the colon were assessed for entry in this single center, clinically randomized trial. Adjuvant therapy and postoperative follow-up were similar in both groups. The primary endpoint was cancer-related survival and secondary endpoints were probability of overall survival and probability of being free of recurrence. Data were analyzed according the intention-to-treat principle.

Results: Two hundred and nineteen patients entered the study (111 LAC group and 108 OC group). The median follow-up was 95 months (range, 77–133). There was a tendency of higher cancer-related survival ($P = 0.07$, NS) and overall survival ($P = 0.06$, NS) for the LAC group. Probability of cancer-related survival was higher in the LAC group ($P = 0.02$) when compared with OC. The regression analysis showed that LAC was independently associated with a reduced risk of tumor relapse (hazard ratio 0.47, 95% CI 0.23–0.94), death from a cancer-related cause (0.44, 0.21–0.92) and death from any cause (0.59, 0.35–0.98).

Conclusions: LAC is more effective than OC in the treatment of colon cancer.

(*Ann Surg* 2008;248: 1–7)

We previously published the results of a single center, randomized clinical trial comparing the efficacy of laparoscopy-assisted colectomy (LAC) and open colectomy (OC) for nonmetastatic colon cancer.[1] This study, with a median length of follow-up of 43 months, demonstrated that laparoscopy-assisted colectomy was more effective for treatment of colon cancer in terms of morbidity, hospital stay, tumor recurrence, and cancer-related survival.

Other major randomized studies, such as the Clinical Outcome of Surgical Therapy,[2] the UK Medical Research Council trial of Conventional versus Laparoscopic-Assisted Surgery in Colorectal Cancer,[3] and the Colon Cancer Laparoscopic or Open Resection (COLOR),[4] confirmed the benefits of LAC with respect to morbidity and hospital stay.

The striking finding of our trial was a higher probability of the cancer-related survival in the LAC group and that LAC was independently associated with a reduced risk of tumor relapse and death from a cancer-related cause when compared with the OC. Interestingly, these differences between the LAC and OC groups were only due to the patients with advanced nonmetastatic cancer (stage III). This advantage of LAC in relation to OC has not been reproduced by any other major randomized clinical trial. Although the long-term oncological data of the Conventional versus Laparoscopic-Assisted Surgery in Colorectal Cancer or the COLOR trials have yet to be published to date, the Clinical Outcome of Surgical Therapy study, which is the largest randomized controlled trial performed so far, showed no differences in terms of recurrence rate, and disease-free and overall survival between the LAC and OC after a median follow-up of 4 years.[2] However, there are reports of uncontrolled studies that show survival benefits LAC for cancer compared with historical series of conventional open surgery.[5–7]

Thus, it is essential to revisit our trial and evaluate the outcome after all included patients accomplished a follow-up of more than 5 years. We report the long-term oncological data comparing LAC and OC for nonmetastatic colon cancer.

PATIENTS AND METHODS

Patients

From November 1993 to July 1998 all patients with an adenocarcinoma of the colon 15 cm above the anal verge admitted in our Unit were evaluated. Exclusion criteria were cancer located at the transverse colon, distant metastasis,

From the Departments of *Surgery and †Gastroenterology, and Centro de Investigaciones Biomédicas Esther Koplowitz, IMDiM, IDIBAPS, Hospital Clínic, University of Barcelona, Spain.

Supported in part by grants from the Instituto de Salud Carlos III (RC03/02 and RC03/10) and the Ministerio de Educación y Ciencia (SAF2004-07190).

Reprints: Dr. Antonio M. Lacy, Department of Surgery, Hospital Clínic, Villarroel 170, 08036 Barcelona, Spain. E-mail: alacy@clinic.ub.es.

ISSN: 0003-4932/08/24801-0001
DOI: 10.1097/SLA.0b013e31816a9d65

adjacent organ invasion, intestinal obstruction, past colonic surgery, and no consent to participate in the study.[1]

Randomization was performed the day before operation. Patients were stratified in 2 groups according to tumor location (right and left side, with respect to the splenic flexure), and subsequently assigned to LAC or OC by means of sealed opaque envelopes containing computer-generated random numbers. The study was approved by the institutional Ethics of Research Committee, and oral consent was obtained from each patient. No patient was excluded after randomization.

Operative procedures, processing of specimens, postoperative management, and adjuvant treatment have been described previously.[1]

Postoperative Surveillance

All patients were followed according to a preestablished protocol, which includes medical history, physical examination, and laboratory studies including serum carcinoembryonic antigen (CEA) levels 1 month after surgery and every 3 months thereafter. At each visit, symptoms were recorded and wound scars examined for subcutaneous metastasis. Abdominal ultrasonography or computed tomography, and chest x-ray were performed every 6 months, and total colonoscopy was performed every year. When colonoscopy was incomplete, a combination of sigmoidoscopy and barium enema was undertaken.

Recurrences were histologically confirmed and classified as distant metastasis, locoregional relapse (tumor growth restricted to the anastomosis or the region of primary operation), peritoneal seeding, and port-site metastasis. Diagnosis of port-site metastasis required the absence of carcinosis of the peritoneum.

Statistical Methods

The main endpoint of the study was cancer-related survival. Secondary endpoints were probability of overall survival and probability of being free of recurrence.

Categorical variables were compared by means of the χ^2 test, applying the Yates correction when necessary. Continuous variables were compared by means of the Student t test. Calculation of the sample size has been described previously.[1]

Data were analyzed according to the intention-to-treat principle. Survival was calculated from surgical resection of primary tumor to the last visit or death. For cancer-related survival, patients dying by other reasons were censored at the time of death. Probability curves were constructed according to the Kaplan-Meier method and compared with the log-rank test.

A proportional hazards modeling with forward selection was used to determine the influence of baseline characteristics on cancer-related survival and other variables. The surgical procedure and any variable reaching a P value of less than 0.10 in the univariate analysis were introduced in the multivariate analysis to identify independent predictors. Predefined baseline variables for the univariate analysis were sex, age, intervention period (1993–1995 vs. 1996–1998), preoperative serum CEA levels, size, extent and degree of differentiation of primary tumor, and lymph node metastasis. For continuous variables, the cut-off level chosen was their median value. TNM tumor stage was not included as a single covariable but rather decomposed in the corresponding original counterparts (extent of primary tumor and lymph node metastasis). Nevertheless, probability curves were also constructed after classifying the patients according to the TNM system.

Because of the scope of the study was nonmetastatic colon cancer, patients in whom metastasis was detected intraoperatively were not included in the present analysis. Finally, in addition to the intention-to-treat analysis, data were recalculated according to the treatment patients actually received.

All P values were two-sided. A P value of less than 0.05 was considered to indicate a statistically significant difference. All calculations were performed by using the SPSS software package version 12 (SPSS Inc., Chicago, IL).

RESULTS

Characteristics of the Patients

Two hundred and nineteen patients with colon cancer took part in the study (111 in the LAC group and 108 in the OC group). However, metastases were detected intraoperatively in 11 patients (5 from the LAC group and 6 from the OC group) and, consequently, they were not considered in the current analyses of the long-term outcome.

There were no significant differences between groups in baseline characteristics except for age (significantly lower in LAC group) and preoperative serum CEA concentration (significantly higher in LAC group) (Table 1).

LAC intervention was converted to OC in 12 patients (11%) as a result of suspicion of tumor invasion of adjacent organs. Seven of these patients had a stage II tumor and 5 a stage III tumor, according to the TNM classification. No conversion was due to technical problems.

Follow-up

One patient from each group was lost to follow-up 12 months after surgery. All remaining patients were compliant with the proposed postoperative surveillance protocol. The median follow-up was 95 months (range, 77–133) in the whole series, 95 months (range, 77–133) in the LAC group, and 91 months (range, 80–130) in the OC group.

Overall Survival

There was a trend of a higher overall survival in the LAC group when compared with the OC group, although the difference was not statistically significant ($P = 0.06$). Thirty-eight patients (36%) of the LAC group and 50 patients (49%) of the OC group died (Table 2). Similarly, there was a tendency of a higher probability of overall survival in favor of the LAC group compared with OC group (Fig. 1), but difference did not reach statistical significance ($P = 0.07$). However, the Cox regression analysis identified the surgical procedure ($P = 0.04$) together with the presence of lymph node metastasis ($P = 0.02$) as independent predictors of overall survival (Table 3).

Cancer-related Survival

There was a tendency of higher cancer-related survival in the LAC group, but it was not statistically different ($P = 0.07$).

TABLE 1. Baseline Characteristics of Patients With Nonmetastatic Colon Cancer

	Laparoscopic-assisted Colectomy (n = 106)	Open Colectomy (n = 102)	*P*
Age (yr)	68 ± 12	71 ± 11	0.02
Sex (male/female)	53/53	46/56	0.48
Preoperative serum CEA (ng/mL)	17 ± 43	6 ± 12	0.04
Tumor location			0.21
Cecum	32	21	
Ascending colon	7	15	
Hepatic flexure	8	10	
Descending colon	7	9	
Sigmoid colon	52	47	
Intervention			0.43
Right colectomy	47	46	
Left colectomy	4	1	
Sigmoidectomy	49	44	
High anterior resection	3	8	
Subtotal colectomy	1	2	
Hartmann procedure	2	1	
Lymph nodes in the resected specimen (number)	11.1 ± 7.9	10.7 ± 7.3	0.70
Extent of primary tumor*			0.41
1	16	10	
2	14	12	
3	73	74	
4	3	6	
Lymph-node metastasis			0.70
No	68	67	
Yes	38	35	
Tumor stage*			0.28
I	27	18	
II	42	48	
III	37	36	

*According to the TNM classification (International Union against Cancer).
CEA indicates carcinoembryonic antigen.

TABLE 2. Tumor Recurrence and Survival

	Laparoscopic-assisted Colectomy (n = 106)	Open Colectomy (n = 102)	*P*
Overall mortality (%)	38 (36%)	50 (49%)	0.06
Cancer-related mortality (%)	17 (16%)	28 (27%)	0.07
Causes of death			0.49
Perioperative mortality	1	3	
Tumor progression	16	25	
Others	21	22	
Tumor recurrence (%)	19 (18%)	29 (28%)	0.07
Type of recurrence			0.65
Distant metastasis	7	10	
Locoregional relapse	8	14	
Peritoneal seeding	3	5	
Port site metastasis	1	0	

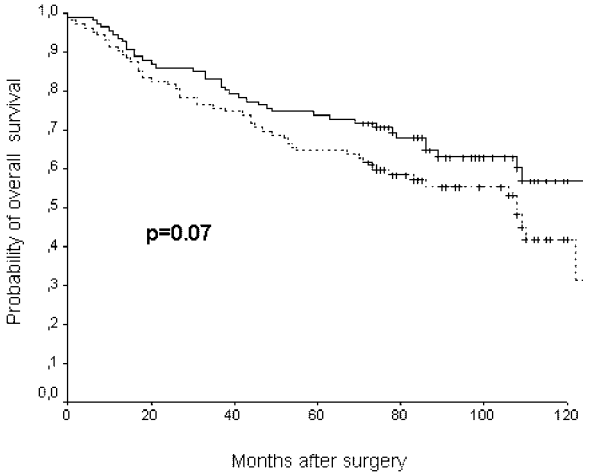

FIGURE 1. Kaplan-Meier estimates of overall survival. Laparoscopic-assisted colectomy group is represented by a continuous line and open colectomy group is represented by a dotted line.

TABLE 3. Results of Cox Regression Analysis

	Hazard Ratio (95% CI)	*P*
Overall survival		
Lymph node metastasis (presence vs. absence)	0.54 (0.32–0.92)	0.02
Surgical procedure (OC vs. LAC)	0.59 (0.35–0.98)	0.04
Cancer-related survival		
Lymph node metastasis (presence vs. absence)	0.33 (0.16–0.68)	0.002
Preoperative serum CEA levels (≥2.8 ng/mL vs. <2.8 ng/mL)	0.43 (0.19–0.94)	0.04
Surgical procedure (OC vs. LAC)	0.44 (0.21–0.92)	0.03
Probability of being free of recurrence		
Lymph node metastasis (presence vs. absence)	0.39 (0.19–0.77)	0.007
Surgical procedure (OC vs. LAC)	0.47 (0.23–0.94)	0.03

95% CI indicates 95% confidence interval; OC, open colectomy; LAC, laparoscopic-assisted colectomy; CEA, carcinoembryonic antigen.

Seventeen patients (16%) of the LAC group and 28 patients (27%) of the OC group died from causes related to cancer (Table 2). Figure 2 shows the higher probability of cancer-related survival ($P = 0.02$) in the LAC group when compared with OC. In addition, the multivariate analysis confirmed that the surgical procedure ($P = 0.03$), the presence of lymph node metastasis ($P = 0.002$), and preoperative serum CEA levels ($P = 0.04$) were independent predictors of cancer-related survival (Table 3).

Tumor Recurrence

Although the difference was not statistically significant, tumor recurrence tended to be lower in the LAC

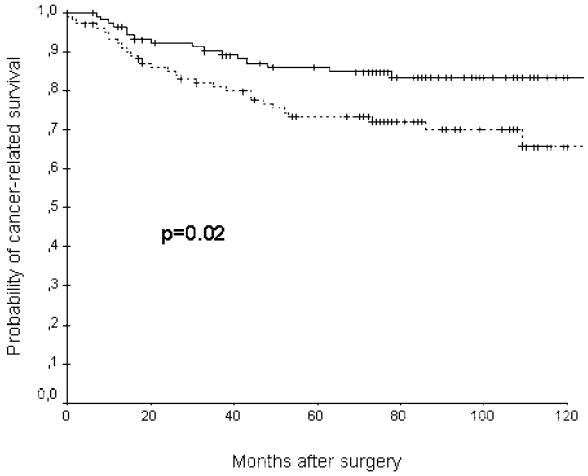

FIGURE 2. Kaplan-Meier estimates of cancer-related survival. Laparoscopic-assisted colectomy group is represented by a continuous line and open colectomy group is represented by a dotted line.

group ($P = 0.07$). Nineteen patients (18%) of the LAC group and 29 patients (28%) of the OC group developed tumor recurrence (Table 2). Importantly, there is no difference in the type of recurrence in both treatment groups (Table 2). As shown in Figure 3, the probability of being free of recurrence was $P = 0.054$ (Fig. 3) in the LAC group when compared with the OC group. Moreover, the surgical procedure ($P = 0.03$), the presence of lymph node metastasis ($P = 0.007$), and the preoperative serum levels of CEA were found to be independent predictors of tumor recurrence (Table 3).

Survival and Tumor Recurrence Analysis According to the TNM Classification

When patients were stratified according to the tumor stage, the probabilities of overall survival ($P = 0.048$), cancer-related survival ($P = 0.02$), and being free of recurrence ($P = 0.048$) were significantly higher in the LAC group compared with OC for stage III tumors. The superiority of LAC over OC regarding these variables was only due to significant differences in patients with stage III tumors; probability curves in patients with stage I and II tumors were identical for both therapeutic approaches overall survival, cancer-related survival, and being free of recurrence ($P =$ NS, stage I and II).

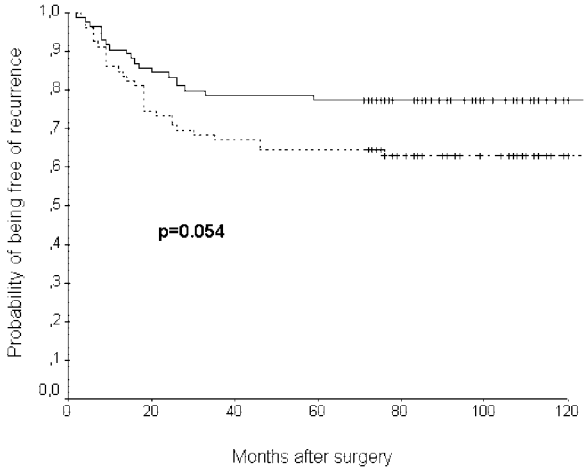

FIGURE 3. Kaplan-Meier estimates of probability of being free of recurrence. Laparoscopic-assisted colectomy group is represented by a continuous line and open colectomy group is represented by a dotted line.

Survival and Tumor Recurrence Analysis According to the Actual Treatment

When the analysis of the results was performed on an actual treatment basis, the probabilities of overall survival ($P = 0.01$), cancer-related survival ($P = 0.0002$), and being free of tumor recurrence ($P = 0.0015$) were significantly higher in the LAC group when compared with the OC group (Fig. 4).

DISCUSSION

This is the first randomized controlled trial with a long-term follow-up (median of 95 months) comparing LAC and OC for nonmetastatic colon cancer. The overall survival and recurrence rate favored the LAC group, but the differences did not reach statistical significance. However, disease-related survival was significantly higher in the LAC group. This survival advantage was more pronounced in patients with stage III tumors, in which LAC was associated with a significantly lower probability of recurrence and higher disease-free and overall survival, whereas no differences were observed in patients with stage I and II colon cancer. These results are in line with the original article,[1] with a median follow-up of 43 months.

The presented data do raise some questions that have to be addressed. The first one is how LAC could achieve a better survival rate compared with the OC group. The second question is why this advantage in survival is limited to patients with stage III colon cancer and, third, why these results have not been reproduced by other trials. Supporting evidence of the beneficial oncological role of LAC includes its impact on surgical stress response, cellular immunity, cytokine release, intraoperative tumor manipulation, complication rate, and blood transfusion factors. The stress response after colorectal surgery for cancer is less pronounced and consequently results in better preservation of the early postoperative cellular immune function and attenuated disturbance of inflammatory mediators when the laparoscopic approach is chosen.[8,9]

The interrelationship between the extent of the stress response after the trauma of surgery and the host resistance to cancer was clearly described in an animal model of intraperitoneal injection of tumor cells after laparotomy or an incision of the skin. The laparotomy group resulted in augmented tumor growth and a significant reduction in interleukin-2 and lymphocyte-activated killer cells, although a similar surgical wound on the animal's back did not promote any tumor growth and did not have these immunomodulatory effects.[10] Carter et al[11] showed that the less invasive laparoscopy-assisted cecectomy is associated with decreased formation of postoper-

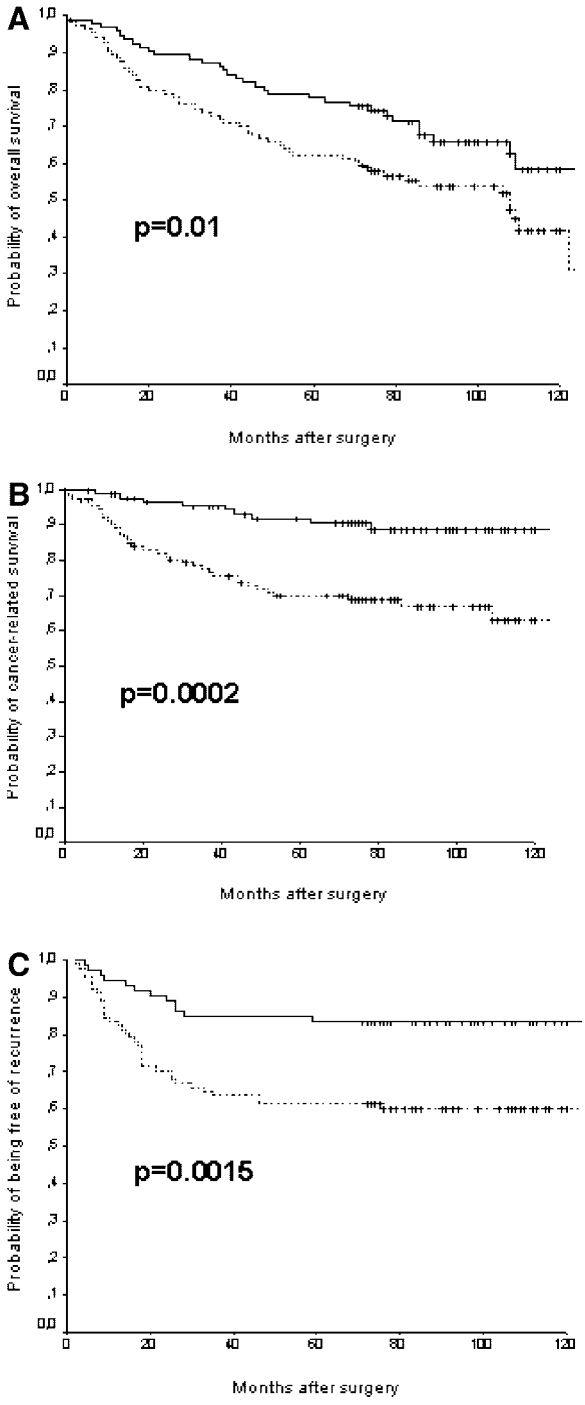

FIGURE 4. Kaplan-Meier estimates of probability of overall survival (A), cancer-related survival (B), and being free of recurrence (C) according to the treatment patients actually received. Laparoscopic-assisted colectomy group is represented by a continuous line and open colectomy group is represented by a dotted line.

ative pulmonary metastases compared with open cecectomy in a murine model. In a similar murine model, open-surgery–related immunosuppression resulted in an increased tumor growth when compared with laparoscopy-assisted procedures.[12] In that sense, it has been suggested that a decrease in plasma insulin-like growth factor binding protein-3 concentration as seen in open colon cancer surgery may be, at least in part, responsible for these tumor stimulatory effects.[13,14]

A similar pattern on cytokine release is also seen in patients,[9,15–17] and it has been postulated that the less attenuated cytokine response after laparoscopic surgery is due to the reduced surgical trauma. Several researchers have studied the influence of attenuated cytokines release after laparoscopic procedures on tumor recurrence. Pera et al[18] reported in a randomized controlled trial the influence of postoperative acute phase response on angiogenesis and tumor growth. This group found that postoperative serum interleukin-6 and vascular endothelial growth factor (VEGF) levels were significantly higher in the laparotomy group than in the laparoscopy group. These increased levels of systemic proinflammatory cytokines and VEGFs were associated with increased angiogenesis and tumor growth. In a report by Belizon et al[19] it was shown that colon cancer patients before surgery have higher VEGF levels than patients without tumors. Also, both open and closed colorectal resection are associated with significantly elevated plasma VEGF levels early after surgery, however, this elevation is significantly increased and occurs earlier in open surgery patients. VEGF is the most potent inducer of angiogenesis that is necessary for wound healing but also promotes tumor growth. In a recent report, Chen et al[20] described that in patients with gastric cancer, the expression of VEGF correlated with the infiltration of the gastric tumor, metastatic spread, and prognosis. Also in colon cancer, VEGF is the predominant angiogenic factor and is associated with tumor recurrence, formation of metastases, and poor prognosis. Interestingly, bevacizumab (Avastin; Genentech Inc., South San Francisco, CA), a recombinant humanized anti-VEGF monoclonal antibody that inhibits tumor angiogenesis, has demonstrated survival benefit in patients with previously untreated metastatic colorectal cancer when combined with irinotecan/fluorouracil.[21] These data not only stress the important role for VEGF in colorectal cancer, but also may explain why a less invasive procedure for colorectal resection might have an improved outcome.

Restricted access to the abdominal cavity with the laparoscopic approach raises the likelihood of minimal tumor handling and an accurate application of nontouch techniques, both favoring the crucial oncology principle to avoid dissemination of tumor cells during the operation. In fact, there is large evidence to support the hypothesis that surgical manipulation may promote colorectal cancer cell detachment and mobilization.[19,22,23] Weitz et al[22] demonstrated the presence of circulating cancer cells in up to 20% of baseline negative patients during conventional OC. In most cases, it was a transient phenomenon, and neoplastic cells usually became undetectable shortly after surgery. Using a nontouch technique, Sales et al[23] reported a lower rate of tumor cell detection in the draining veins, which was confirmed by Hayashi et al[24] who showed a significantly lower rate of intraoperative cancer cell mobilization in the portal venous system. Nonetheless, one can easily argue that a nontouch technique can also be applied in open surgery and, indeed, an

early pilot study did not show differences between the 2 surgical approaches on intraoperative tumor dissemination when a nontouch technique was used.[25]

Finally, another possible explanation for the oncological advantages of LAC observed in our study might possibly rely on the difference in complication rate (LAC vs. OC: 12 vs. 31; $P < 0.001$) and blood loss (LAC vs. OC; 105 vs. 193 mL; $P < 0.001$).[1] The transfusion of red blood cell enhances the angiogenesis of tumor and increases recurrent disease.[26] However, the reported minimal blood losses in both groups did not lead to differences in red blood cell transfusion regimen between the groups. Furthermore, conventional surgery can also be performed with minimal morbidity with avoidance of transfusion. Although this target can be achieved with ease in open surgery, it requires a substantial learning curve in the more technically demanding laparoscopic surgery. In our study, differences in postoperative morbidity may have contributed to the long-term benefits of LAC. Chance finding cannot definitely be excluded.

We could only speculate why the survival benefit of LAC is predominantly seen in patients with stage III tumors. The effector cells of nonspecific immune response, that is, natural-killer cells, which are thought to be crucial in tumor cell immunosurveillance, were suppressed to a greater extent in the early postoperative period following OC compared with LAC.[9,27] Furthermore, it has been shown that natural-killer cells are critical in controlling metastatic tumor growth in colorectal cancer whereby low levels of in vitro NK-cell cytotoxicity may identify a subgroup of patients at high risk for recurrence.[28] Consequently, it might be that in cases with low levels of in vitro NK-cell cytotoxicity, that is, stage III colorectal cancer, the surgical procedure should not further deteriorate to this cell population and, thus, the laparoscopic approach is preferred.

Variations in case volume and surgeon's experience may also account for differences in the surgical outcome observed in single dedicated centers and large multicenter trials.[29] Birkmeyer et al[30] observed that associations between hospital volume and operative mortality are largely mediated by surgeon volume and surgeons' experience. They concluded that patients could substantially improve their chances of survival by selecting surgeons who perform the operations frequently, even at high-volume hospitals.[31] The impact of hospital case volume on short-term outcome after LAC for colon cancer has been studied in the COLOR trial and showed that performance of such a procedure at hospitals with high-case loads seems to be associated with improved short-term results.[30] If we accept that the surgical outcome relies exclusively on surgeons with large training and substantial experience, and the fact that all large randomized trials were carried out in multicenters with variable experience, one could expect that a single, laparoscopy-devoted center would achieve a better outcome.

In conclusion, in a dedicated laparoscopic center, LAC may result in a long-term survival benefit compared with OC, particularly in advanced cases. This oncological advantage can be explained by a preserved cellular immunity, attenuated stress and inflammatory response, minimal tumor handling, and lower complication rate in patients treated by LAC.

REFERENCES

1. Lacy AM, García-Valdecasas JC, Delgado S, et al. Laparoscopy-assisted colectomy versus open colectomy for treatment of non-metastatic colon cancer: a randomised trial. *Lancet*. 2002;359:2224–2229.
2. The Clinical Outcome of Surgical Therapy (COST) Study Group. A comparison of laparoscopically assisted and open colectomy for colon cancer. *N Engl J Med*. 2004;350:2050–2059.
3. Guillou PJ, Quirke P, Thorpe H, et al. Short-term endpoints of conventional versus laparoscopic assisted surgery in patients with colorectal cancer (MRC CLASICC trial): multicentre, randomised controlled trial. *Lancet*. 2005;365:1718–1726.
4. Veldkamp R, Kuhry E, Hop WC, et al. Colon Cancer Laparoscopic or Open Resection Study Group. Laparoscopic surgery versus open surgery for colon cancer: short-term outcomes of a randomised trial. *Lancet Oncol*. 2005;6:477–484.
5. Poulin EC, Mamazza J, Schlachta CM, et al. Laparoscopic resection does not adversely affect early survival curves in patients undergoing surgery for colorectal adenocarcinoma. *Ann Surg*. 1999;229:487–492.
6. Jacob BP, Salky B. Laparoscopic colectomy for colon adenocarcinoma: an 11-year retrospective review with 5-year survival rates. *Surg Endosc*. 2005;19:643–649.
7. Law WL, Lee YM, Chol HK, et al. Impact of laparoscopic resection for colorectal cancer on operative outcomes and survival. *Ann Surg*. 2007; 245:1–7.
8. Whelan RL, Franklin M, Holubar SD, et al. Postoperative cell mediated immune response is better preserved after laparoscopic vs open colorectal resection in humans. *Surg Endosc*. 2003;17:972–978.
9. Wichmann MW, Huttl TP, Winter H, et al. Effects of laparoscopic vs open colorectal surgery, a prospective clinical study. *Arch Surg*. 2005; 140:692–697.
10. Eggermont AM, Steller EP, Sugarbaker PH. Laparotomy enhances intraperitoneal tumour growth and abrogates the antitumour effects of interleukin-2 and lymphokine-activated killer cells. *Surgery*. 1997;102: 71–78.
11. Carter JJ, Feingold DL, Kirman I, et al. Laparoscopic-assisted cecectomy is associated with decreased formation of postoperative pulmonary metastases compared with open cecectomy in a murine model. *Surgery*. 2003;134:432–436.
12. Allendorf JDF, Bessler M, Horvath KD, et al. Increased tumor establishment and growth after open vs laparoscopic surgery in mice may be related to differences in postoperative T-cell function. *Surg Endosc*. 1999;13:223–235.
13. Kirman I, Cekic V, Poltaraskaia N, et al. Open surgery induces a dramatic decrease in circulating intact IGFBP-3 in patients with colorectal cancer not seen with laparoscopic surgery. *Surg Endosc*. 2005;19: 55–59.
14. Kirman I, Cekic V, Poltaraskaia M, et al. Plasma from patients undergoing major open surgery stimulates in vitro tumor growth: lower insulin-like growth factor binding protein 3 levels may, in part, account for this change. *Surgery*. 2002;132:186–192.
15. Delgado S, Lacy AM, Filella X, et al. Acute phase response in laparoscopic and conventional colectomy in colon cancer. A randomized study. *Dis Colon Rectum*. 2001;44:638–646.
16. Wu FPK, Sietses C, von Blomberg BME, et al. Systemic and peritoneal inflammatory responses after laparoscopic or conventional colon resection in cancer patients. *Dis Colon Rectum*. 2003;46:147–155.
17. Leung KL, Lai PBS, Ho RLK, et al. Systemic cytokines response after laparoscopic assisted resection of rectosigmoid carcinoma: a prospective randomized trial. *Ann Surg*. 2000;231:506–511.
18. Pera M, Nelson H, Rajkumar SV, et al. Influence of postoperative acute-phase response on angiogenesis and tumor growth: open vs. laparoscopic-assisted surgery in mice. *J Gastrointest Surg*. 2003;7:783–790.
19. Belizon A, Balik E, Feingold DL, et al. Major abdominal surgery increases plasma levels of vascular endothelial growth factor: open more so than minimally invasive methods. *Ann Surg*. 2006;244:792–798.
20. Chen W, Ouyang XN, Lin QC. Study on the relationship between

vascular endothelial growth factor and syndrome type of traditional Chinese medicine in patients with gastric carcinoma. *Zhongguo Zhong Xi Yi Jie He Za Zhi*. 2007;27:127–130.
21. Kabbinavar FF, Hambleton J, Man RD, et al. Combined analysis of efficacy: the addition of bevacizumab to fluorouracil/leucovorin improves survival for patients with metastatic colorectal cancer. *J Clin Oncol*. 2005;23:3706–3712.
22. Weitz J, Jienle P, Lacroix J, et al. Dissemination of tumour cells in patients undergoing surgery for colorectal cancer. *Clin Cancer Res*. 1998;4:343–348.
23. Sales JP, Wind P, Douard R, et al. Blood dissemination of colonic epithelial cells during non-touch surgery for rectosigmoid cancer. *Lancet*. 1999;354:392.
24. Hayashi N, Egami Hi, Kai M, et al. Non-touch isolation technique reduces intraoperative shedding of tumour cells into portal vein during resection of colorectal cancer. *Surgery*. 1999;125:369–374.
25. Bessa Z, Castells A, Lacy AM, et al. Laparoscopic-assisted vs. open colectomy for colorectal cancer: influence on neoplastic cell mobilization. *J Gastointest Surg*. 2001;5:66–73.
26. Patel HB, Nasir FA, Nash GF, et al. Enhanced angiogenesis following allogeneic blood transfusion. *Clin Lab Haematol*. 2004;26:129–135.
27. Takeuchi I, Ishida H, Mori T, et al. Comparison of the effects of gasless procedure, CO_2-pneumoperitoneum and laparotomy on splenic and hepatic natural killer activity in a rate model. *Surg Endosc*. 2004;18:255–260.
28. Tartter PI, Steinberg B, Barron D, et al. The prognostic significance of natural killer cytotoxicity in patients with colorectal cancer. *Arch Surg*. 1987;122:1264–1268.
29. Porter GA, Soskolne CL, Yakimets WW, et al. Surgeon-related factors and outcome in rectal cancer. *Ann Surg*. 1998;227:157–167.
30. Birkmeyer JD, Stukel TA, Siewers AE, et al. Surgeon volume and operative mortality in the United States. *N Engl J Med*. 2003;349:2117–2127.
31. Kuhry E, Bonjer HJ, Haglind E, et al. COLOR Study Group. Impact of hospital case volume on short-term outcome after laparoscopic operation for colonic cancer. *Surg Endosc*. 2005;19:687–692.