International Journal of Colorectal Disease (2019) 34:1283–1293
https://doi.org/10.1007/s00384-019-03315-0

ORIGINAL ARTICLE

Peridural analgesia does not impact survival in patients after colon cancer resection: a retrospective propensity score-adjusted analysis

Elena F. Wurster[1] · Frank Pianka[1] · Rene Warschkow[2,3] · Pia Antony[1] · Thorsten Brenner[4] · Markus A. Weigand[4] · Bruno M. Schmied[2] · Markus W. Büchler[1] · Ignazio Tarantino[2] · Alexis Ulrich[5]

Accepted: 10 May 2019 / Published online: 6 June 2019

Abstract

Purpose To assess the putative impact of peridural analgesia on oncological outcome in patients undergoing resection of stages I–IV colon cancer.

Methods In a single-center study, 876 patients undergoing resection for primary colon cancer (AJCC stages I–IV) between 2001 and 2014 were analyzed. Mean follow-up of the entire cohort was 4.2 ± 3.5 years. Patients who did and did not receive peridural analgesia were compared using Cox regression and propensity score analyses.

Results Overall, 208 patients (23.7%) received peridural analgesia. Patients' characteristics were biased with regard to the use of peridural analgesia (propensity score 0.296 ± 0.129 vs. 0.219 ± 0.108, $p < 0.001$). After propensity score matching, the use of peridural analgesia had no impact on overall (HR 0.81, 95% CI 0.59–1.11, $p = 0.175$), cancer-specific (HR 0.72, 95% CI 0.48–1.09, $p = 0.111$), and disease-free survival (HR 0.89, 95% CI 0.66–1.19, $p = 0.430$). The 5-year overall survival after propensity score matching was 60.9% (95% CI 54.8–67.7%) for patients treated with peridural analgesia compared with 54.1% (95% CI 49.5–59.1%) for patients not treated with peridural analgesia. Cancer-specific and disease-free survival showed similar non-significant results.

Conclusions Peridural analgesia in patients after colon cancer resection was not associated with a better oncological outcome after risk adjusting in multivariable Cox regression and propensity score analyses. Hence, oncological outcome should not serve as a reason for the use of peridural analgesia in patients with colon cancer.

Keywords Peridural analgesia · Colon cancer · Overall survival · Cancer-specific survival

Introduction

Due to its numerous advantages, including better pain control [1], early mobilization, a decrease in the duration of postoperative ileus [2], and a possible reduction in postoperative morbidity and mortality [3, 4], peridural analgesia (PDA) has become a mainstay in colorectal surgery. Furthermore, recent studies of fast-track or enhanced recovery surgery protocols have shown a significant reduction in healthcare-associated infections as well as in the duration of hospital stay compared with classic perioperative care [5]. Although studies have failed to demonstrate a reduction in the duration of

Elena F. Wurster, Frank Pianka, Ignazio Tarantino and Alexis Ulrich contributed equally to this work.

✉ Ignazio Tarantino
ignazio.tarantino@kssg.ch

[1] Department of General, Visceral and Transplantation Surgery, University Hospital Heidelberg, Im Neuenheimer Feld 110, 69120 Heidelberg, Germany

[2] Department of General, Visceral, Endocrine and Transplantation Surgery, Kantonsspital St. Gallen, Rorschacher Straße 95, 9007 St. Gallen, Switzerland

[3] Institute of Medical Biometry and Informatics, University of Heidelberg, Im Neuenheimer Feld 305, 69120 Heidelberg, Germany

[4] Department of Anesthesiology, University Hospital Heidelberg, Im Neuenheimer Feld 110, 69120 Heidelberg, Germany

[5] Surgical Department I, Städtische Kliniken Neuss Lukaskrankenhaus GmbH, Lukaskrankenhaus GmbH, Preußenstraße 84, 41464 Neuss, Germany

hospital stay associated with PDA use, it continues to play a central role in postoperative patient management after colorectal surgery [6].

Interestingly, a recent study postulated that the use of PDA might actually confer a survival benefit to patients undergoing colorectal surgery [7]. Here, a difference in the 5-year survival rate (62% vs. 54%, $p < 0.02$) could be observed in patients treated with or without PDA, respectively. In this study, patients treated with PDA appeared significantly younger and possessed a lower American Society of Anesthesiologists (ASA) score, thereby possibly influencing results. Moreover, the study included patients with both colon and rectum cancers, further distorting the observed outcome as both tumor entities should be investigated separately due to differences in anatomy, clinical behavior, and stage-dependent treatment [8].

As such, the purpose of the present analysis was to assess the possible impact of the use of PDA on oncological outcome in a large homogenous patient population, exclusively colon cancer. In order to increase the statistical power and further elaborate on the possible beneficial impact of PDA use on recurrence and survival, both the Cox proportional hazard regression analyses and propensity scoring methods were applied.

Methods

Data source and cohort definition

The present retrospective analysis is based on a prospectively maintained colorectal database from the Department of General, Visceral and Transplantation Surgery at the University of Heidelberg. Overall, 1252 patients with histologically confirmed colon cancer were identified between January 2001 and June 2014. Exclusionary criteria included patients with missing demographic data, missing information pertaining to the use of PDA, an in-hospital or 30-day mortality, and patients receiving resections due to local recurrence, R1/2 or Rx resection, and patients who were lost to follow-up (Fig. 1). A total of 876 patients remained for further analysis. Those patients were then grouped according to whether or not they received a PDA.

Data collection, operative technique, definitions, and follow-up

Informed consent was acquired from patients scheduled to undergo oncologic colon resection. An independent ethics committee at the University Heidelberg approved the present study (S-649/2012). Predefined data were collected from medical charts and recorded in a prospectively maintained electronic database. Baseline data included age, gender, ASA score, body mass index (BMI), type of colonic resection (right hemicolectomy, resection of transverse colon, left hemicolectomy, subtotal colectomy), American Joint Committee on Cancer (AJCC) stage, number of harvested and positive lymph nodes, tumor grade, and carcinoembryonic antigen (CEA). In addition, the surgical approach (open vs. laparoscopic) and the administration of adjuvant chemotherapy (yes/no) were ascertained.

All operations were performed or supervised by experienced colorectal surgeons using highly standardized techniques. The surgical concept of complete mesocolic excision, in which the undamaged visceral fascia of the mesocolon is sharply resected from the parietal fascia of peritoneum, was performed [9]. In cases where the tumor was located in the right hemicolon, the venous vessels were divided at their inflow into the identified superior mesenteric vein. The corresponding arteries were transected at the same level. A mobilization of the duodenum (Kocher maneuver) was not routinely performed. In patients presenting with a carcinoma in the region of the hepatic flexure, the great curvature of the stomach was freed over a distance of 10 to 15 cm from the right gastroepiploic arcade. The highly standardized surgical techniques were not changed during the study period.

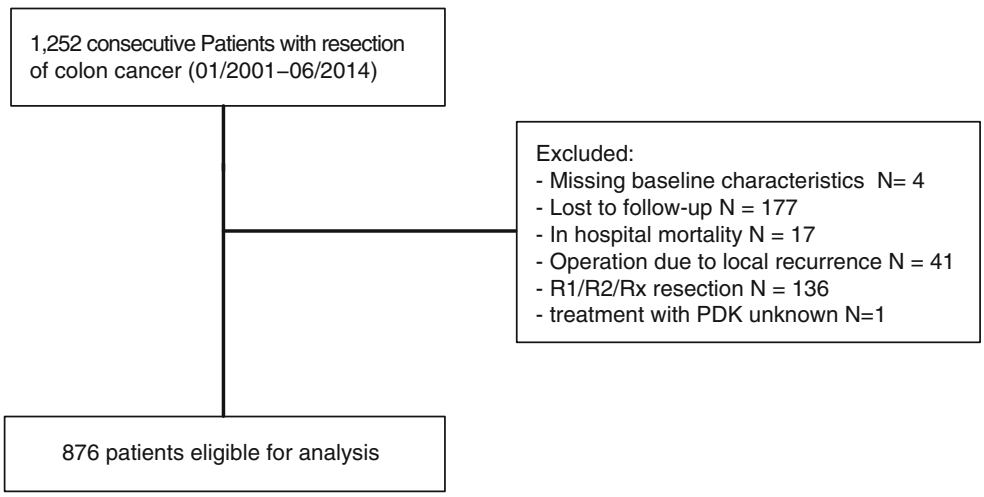

Fig. 1 Patients' cohort definition

Barring other contraindications, the decision for or against the insertion of a peridural catheter was made by the patient in consultation with the attending anesthesiologist. The peridural catheter was placed in the lower thoracic spinal column (thoracic 8/9 ± one level) immediately prior to surgery. Afterwards, general anesthesia was induced via the intravenous administration of sufentanil, propofol, and rocuronium bromide based on a standardized in-house protocol. Anesthesia was then maintained with an inhaled sedative (sevoflurane p.i. or desflurane p.i.). When necessary, rocuronium bromide was administered intravenously in repetitive dosages to ensure for sufficient muscle relaxation. In addition, all patients with PDA received an epidural bolus of 20 μg sufentanil at the beginning of anesthesia. Meanwhile, analgesia was maintained using either intravenous sufentanil or via the peridural catheter using either continuous infusion or bolus injections of ropivacain 0.2–0.5% in combination with sufentanil epidural 10–20 μg. Postoperatively, analgesia was maintained using one of the methods listed as follows: (1) PDA with continuous infusion of both ropivacain 0.2% and sufentanil epidural (initially at a dose of 0.5μg/ml) via the peridural catheter. Dosage reduction was performed according to the standard operating procedure for postoperative pain management. Whenever possible, the administration of sufentanil was reduced and stopped during the first 2 to 3 days postoperatively; (2) patient-controlled analgesia (PCA) with intravenous bolus delivery of oxycodone or piritramide; or (3) nurse-controlled analgesia with intravenous bolus administration of oxycodone or piritramide by the ward staff 4–6 times daily. In addition, peripherally acting analgetics (e.g., paracetamol, metamizol) could be administered in all groups according to individual needs. As soon as patients presented with sufficient gastrointestinal passage, an oral analgetic therapy consisting of a fixed combination of oxycodone/naloxone was initiated 1 or 2 days prior to removal of the peridural catheter and the end of patient-controlled analgesia or the nurse-controlled analgesia, respectively.

Data about the management of patient-controlled intravenous analgesia or PDA in the postoperative period were recorded by members of the Department of Anesthesiology (University Hospital of Heidelberg, Heidelberg, Germany) using a database for the documentation of analgesia (MEDLINQ, Medlinq Softwaresysteme GmbH, Hamburg, Germany). The treatment success was monitored and adjusted if necessary during the daily rounds.

Regular follow-up was performed for 5 years postoperatively at the outpatient clinics of the Surgical Department and at the National Center for Tumor Diseases, Heidelberg, Germany, according to national guidelines [10]. Adjuvant chemotherapy was administered routinely to patients with node-positive disease.

Statistical analysis

Statistical analyses were performed using the R statistical software (www.r-project.org). A two-sided p value < 0.05 was considered statistically significant. Continuous data were expressed as means ± standard deviation. In order to compare proportions, chi-square statistics were used, while the t tests and Mann–Whitney U tests were used for continuous variables as appropriate. Missing data were imputed using the random survival forest method [11].

First, the bias with regard to PDA vs. no PDA was assessed in terms of age, gender, ASA score, BMI, tumor stage, tumor grade, CEA level, type of operation, laparoscopic vs. open surgery, and the need for adjuvant chemotherapy. The same set of covariates including PDA vs. no PDA were then assessed as putative prognostic factors for overall, cancer-specific, and disease-free survival in unadjusted and risk-adjusted Cox regressions including a backward variable selection procedure from the full Cox regression model based on Akaike's information criterion.

Moreover, a propensity score analysis, as a superior and more refined statistical method of adjusting for potential baseline confounding variables, was performed [12–14]. We used the "Matching" R package to perform a bipartite weighting propensity score analysis [15, 16].

Finally, the baseline risk of the matched patients was compared with assurance that no major differences in observed baseline patients' characteristics persisted. The prognostic value of PDA vs. no PDA for overall, cancer-specific, and disease-free survival was assessed in a stratified Cox regression analysis applying the subclasses and the weights obtained by the propensity score analysis.

In sensitivity analysis, first the year of diagnosis was incorporated in univariate, multivariable, and propensity score analysis as a strata term. In addition, in a second step, the AJCC stage was also incorporated as a second strata term in univariate and multivariable Cox regression analysis. The estimates were based on the Huber sandwich estimator.

Reporting of this study was carried out in accordance with the STROBE checklist (www.strobe-statement.org). Rene Warschkow (Master of Science in Medical Biometry) performed the statistical analysis.

Results

Patient population and baseline characteristics

The patient selection is illustrated in a flow chart in Fig. 1. A total of 876 patients that underwent R0 resection for primary colon cancer (AJCC stages I–IV) were analyzed. Of these, 208 patients (23.7%) received a PDA and 668 patients (76.3%) received conventional intraoperative general and

postoperative intravenous and oral analgesia. The application of PDA decreased over time. The last PDA was applied end of 2010, and thereafter, PDA was no longer used in patients with colon cancer. The mean duration of PDA was 4.5 ± 2.1 days after surgery.

Mean follow-up of the entire cohort was 4.2 ± 3.5 years. The follow-up for patients receiving a PDA was significantly longer than in patients not receiving a PDA (5.2 ± 3.3 vs. 3.9 ± 3.5, $p < 0.001$). The distribution of baseline characteristics was similar concerning age, gender, BMI, tumor grade, and CEA (Table 1). There was no statistically significant difference in the rate of patients receiving adjuvant chemotherapy. In contrast, ASA scores and AJCC stages differed markedly between the two analyzed groups. Patients treated with PDA were significantly more often classified as ASA III and IV and presented with significantly more advanced tumor stages. In addition, patients operated in minimally invasive laparoscopic technique were significantly less often treated with PDA. Although without clinical relevance, the number of harvested and the number of positive lymph nodes (median number and interquartile range of harvested lymph nodes in patients with and without PDA 18 (13–24) vs. 20 (15–25); mean number and standard deviation of positive lymph nodes 2.0 ± 3.3 vs. 1.6 ± 3.2) also differed significantly ($p = 0.032$ and $p = 0.015$). In patients with PDA, a right hemicolectomy, transverse colon resection, left hemicolectomy, and subtotal colon resection was performed in 102 (49%), 14 (6.7%), 67 (32.2%), and 25 (12%) patients, respectively. In patients without PDA, right hemicolectomy, transverse colon resection, left hemicolectomy, and subtotal colon resection were performed in 296 (44.3%), 48 (7.2%), 273 (40.9%), and 51 (7.6%) patients, respectively.

PDA as a prognostic factor for overall, cancer-specific, and disease-free survival

In an unadjusted Cox proportional hazards regression analysis, a PDA had no significant impact on overall survival (hazard ratio (HR) of death 1.28, 95% confidence interval (CI) 0.99–1.65, $p = 0.065$) (Fig. 2a). The 5-year overall survival for patients treated with a PDA was 60.0% (95% CI 53.6–67.3%) compared with 70.6% (95% CI 66.4–75.1%) for patients not treated with PDA ($p = 0.065$). Overall survival was, however, dependent on age, gender, ASA stage, AJCC stage, differentiation of the tumor, CEA, surgical approach (open vs. laparoscopic), and the administration of adjuvant chemotherapy (Table 2). Even after risk adjusting the Cox regression analysis, PDA had no influence on overall survival (HR 0.87, 95% CI 0.67–1.14, $p = 0.319$). In this adjusted multivariable analysis, age, male gender, ASA classification, advanced AJCC stage, and elevated CEA were shown to be independent predictors for increased mortality. Meanwhile, surgical approach (open vs. laparoscopic), tumor differentiation, and the need for adjuvant chemotherapy did not reach statistical significance. The results were then confirmed using a backward variable selection.

In an unadjusted Cox proportional hazards regression analysis, the risk of cancer-specific mortality was significantly increased by 63% (HR 1.63, 95% CI 1.13–2.34, $p = 0.010$) in patients treated with PDA (Table 2). Indeed, the 5-year cancer-specific survival for patients treated with PDA was 75.2% (95% CI 69.1–81.7%) compared with 84.6% (95% CI 81.2–88.2%) for patients not treated with PDA ($p < 0.001$) (Fig. 2b). After risk adjusting for a variety of confounding factors in multivariable Cox regression analysis, it appeared that PDA did not significantly increase the risk of cancer-specific mortality (HR 0.94, 95% CI 0.64–1.39, $p = 0.762$) (Table 2).

Similarly, in unadjusted Cox proportional hazards regression analysis, the use of PDA was a statistically significant negative prognostic factor for disease-free survival (HR 1.34, 95% CI 1.06–1.69, $p = 0.017$) (Fig. 2c). The 5-year disease-free survival for patients treated with PDA was 55.9% (95% CI 49.4–63.3%) compared with 67.9% (95% CI 63.6–72.4%) for patients not treated with PDA ($p = 0.017$). However, after risk adjustment using multivariable Cox regression, PDA was no longer associated with an increased risk for recurrence (HR 0.96, 95% CI 0.76–1.23, $p = 0.765$).

Propensity score adjustment for patients' characteristics

To more optimally adjust for the significant imbalances in patients' characteristics in those treated with and without PDA in the univariate- and multivariate-adjusted Cox regression analysis, a propensity score was estimated. The propensity score for patients treated with PDA was 0.296 ± 0.129 compared with 0.219 ± 0.108 in those without PDA ($p < 0.001$), thus revealing a strong bias regarding patients' characteristics between the 2 groups (Table 1). During the propensity score matching procedure, one patient treated with PDA and 45 patients treated without PDA had to be excluded because their characteristics could not be matched with patients from the other group. After the matching procedure, the propensity score was virtually the same in the two patient groups (0.294 ± 0.126 for patients treated with PDA compared with 0.294 ± 0.127 for patients treated without PDA [$p = 0.976$]), thus indicating no persisting bias regarding the observed patients' characteristics in the two groups. Patients' characteristics after propensity score matching with the corresponding p values are listed in Table 1.

Table 1 Baseline characteristics, logistic regression for treatment with peridural analgesia, and patients' characteristics after propensity score matching

	Patients characteristics				Logistic regression for PDA		Patients' characteristics after propensity		
							Score matching		
	Total $n = 876$	PDA $n = 208$	Ø PDA $n = 668$	P	OR (95% CI)	P^D	PDA $n = 207$	Ø PDA $n = 623$	P
Age				0.456[A]					0.580[E]
< 65	393 (44.9%)	98 (47.1%)	295 (44.2%)		Reference		98 (47.3%)	308.8 (49.6%)	
> 65	483 (55.1%)	110 (52.9%)	373 (55.8%)		1.33 (0.93–1.92)	0.122	109 (52.7%)	314.2 (50.4%)	
Gender				0.058[A]					0.627[E]
Male	502 (57.3%)	131 (63.0%)	371 (55.5%)		Reference		130 (62.8%)	379.4 (60.9%)	
Female	374 (42.7%)	77 (37.0%)	297 (44.5%)		1.29 (0.92–1.80)	0.138	77 (37.2%)	243.6 (39.1%)	
ASA				0.032[A]					0.550[E]
II	507 (57.9%)	107 (51.4%)	400 (59.9%)		Reference		107 (51.7%)	336.9 (54.1%)	
III/IV	369 (42.1%)	101 (48.6%)	268 (40.1%)		0.75 (0.53–1.07)	0.116	100 (48.3%)	286.1 (45.9%)	
BMI				0.386[A]					0.263[E]
< 30	737 (84.1%)	171 (82.2%)	566 (84.7%)		Reference		171 (82.6%)	534.6 (85.8%)	
> 30	139 (15.9%)	37 (17.8%)	102 (15.3%)		0.88 (0.57–1.37)	0.568	36 (17.4%)	88.4 (14.2%)	
AJCC stage				< 0.001[B]					0.376[E]
I	194 (22.1%)	38 (18.3%)	156 (23.4%)		Reference		38 (18.4%)	94.4 (15.2%)	
II	274 (31.3%)	52 (25.0%)	222 (33.2%)		1.03 (0.63–1.65)	0.913	52 (25.1%)	177.8 (28.5%)	
III	251 (28.7%)	59 (28.4%)	192 (28.7%)		0.57 (0.34–0.95)	0.029	59 (28.5%)	154.5 (24.8%)	
IV	157 (17.9%)	59 (28.4%)	98 (14.7%)		0.25 (0.14–0.45)	< 0.001	58 (28.0%)	196.3 (31.5%)	
Grading				0.914[A]					0.956[E]
G1/2	667 (76.1%)	159 (76.4%)	508 (76.0%)		Reference		158 (76.3%)	474.4 (76.1%)	
G3/4	209 (23.9%)	49 (23.6%)	160 (24.0%)		1.29 (0.87–1.93)	0.206	49 (23.7%)	148.6 (23.9%)	
CEA				0.129[C]					0.931[E]
C0 stage < 5 ng/ml	562 (64.2%)	124 (59.6%)	438 (65.6%)		Reference		124 (59.9%)	364.7 (58.5%)	
C1 stage > 5 ng/ml	219 (25.0%)	63 (30.3%)	156 (23.4%)		1.00 (0.67–1.49)	0.988	62 (30.0%)	190.8 (30.6%)	
Unknown	95 (10.8%)	21 (10.1%)	74 (11.1%)		1.19 (0.70–2.11)	0.526	21 (10.1%)	67.5 (10.8%)	
Operation				0.059[C]					0.856[E]
Right colectomy	398 (45.4%)	102 (49.0%)	296 (44.3%)		Reference		102 (49.3%)	302.3 (48.5%)	
Transverse colectomy	62 (7.1%)	14 (6.7%)	48 (7.2%)		1.41 (0.75–2.81)	0.297	14 (6.8%)	33 (5.3%)	
Left colectomy	340 (38.8%)	67 (32.2%)	273 (40.9%)		1.50 (1.04–2.19)	0.031	67 (32.4%)	212.4 (34.1%)	
Subtotal colectomy	76 (8.7%)	25 (12.0%)	51 (7.6%)		0.70 (0.40–1.23)	0.208	24 (11.6%)	75.3 (12.1%)	
Surgery				< 0.001[A]					0.804[E]
Open	810 (92.5%)	204 (98.1%)	606 (90.7%)		Reference		203 (98.1%)	612.6 (98.3%)	
Laparoscopic	66 (7.5%)	4 (1.9%)	62 (9.3%)		3.56 (1.47–11.16)	0.003	4 (1.9%)	10.4 (1.7%)	
Adjuvant CHT				0.103[A]					0.532[E]
No	631 (72.0%)	159 (76.4%)	472 (70.7%)		Reference		158 (76.3%)	461.9 (74.1%)	
Yes	245 (28.0%)	49 (23.6%)	196 (29.3%)		2.30 (1.48–3.60)	< 0.001	49 (23.7%)	161.1 (25.9%)	

AJCC American Joint Committee on Cancer, *ASA* American Society of Anesthesiologists score, *BMI* body mass index, *CEA* carcinoembryonic antigen, *CHT* chemotherapy, *CI* confidence interval, *PDA* peridural analgesia, *OR* odds ratio

N (%)

[A] Mid-p test

[B] Chi-square test

[C] Chi-square test, MC simulated

[D] Likelihood ratio tests

[E] Weighted chi-square test

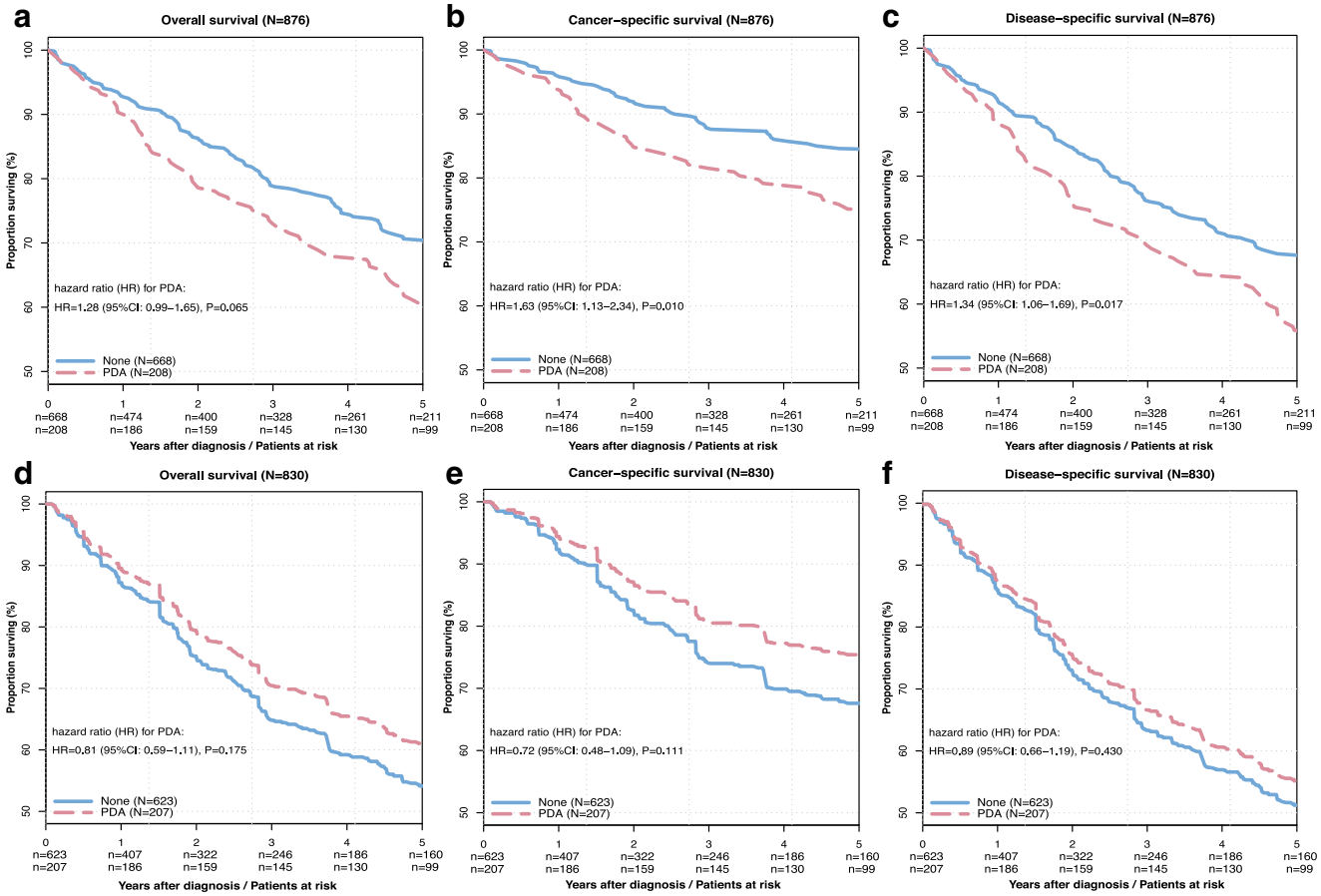

Fig. 2 Comparisons of Kaplan–Meier curves between patients with and without PDA with regard to overall, cancer-specific, and disease-free survivals after resection for colon cancer in unadjusted and propensity score-adjusted analysis

PDA as a prognostic factor for overall, cancer-specific, and disease-free survival after propensity score matching

After adjusting the data according to the propensity score analysis, it was revealed that the use of PDA had no impact on overall survival (HR 0.81, 95% CI 0.59–1.11, $p = 0.175$) (Fig. 2d), cancer-specific survival (HR 0.72, 95% CI 0.48–1.09, $p = 0.111$) (Fig. 2e), and disease-specific survival (HR 0.89, 95% CI 0.66–1.19, $p = 0.430$) (Fig. 2f). The 5-year overall survival after propensity score matching was 60.9% (95% CI 54.8–67.7%) for patients treated with PDA compared with 54.1% (95% CI 49.5–59.1%) for patients not treated with PDA. Similarly, 5-year cancer-specific survival was 75.4% (95% CI 69.5–81.8%) for patients treated with PDA and 67.6% (95% CI 63.0–72.5%) for patients not treated with PDA, respectively. The 5-year disease-free survival was 55.2% (95% CI 49.1–61.9%) for patients treated with PDA and 51.2% (95% CI 46.7–56.2%) for patients not treated with PDA, respectively.

Auxiliary analyses

To account for the decreasing application of PDA over time, several sensitivity analyses were performed.

The survival of patients operated during the period when PDA was not applied (2011–2014, $N = 240$) was compared with the survival of patients operated earlier when PDA was used (2001–2010, $N = 636$). There was a non-significant trend for worse overall (HR 1.46, 95% CI 0.82–2.60, $p = 0.170$) cancer-specific (HR 1.08, 95% CI 0.56–2.11, $p = 0.812$), and disease-free survival (HR 1.61, 95% CI 0.93–2.80, $p = 0.070$).

Furthermore, analyses were stratified over three time periods when PDA was applied (2001–2010) excluding patients operated between 2011 and 2014 when PDA was not used. Of the included 636 patients in this analysis, 208 (32.7%) received a PDA and 428 patients (67.3%) received conventional intraoperative general and postoperative intravenous and oral analgesia. In an unadjusted Cox proportional hazards regression analysis, a PDA had no significant impact on overall (HR 1.20, 95% CI 0.91–1.59, $p = 0.196$) and on disease-free survival (HR 1.26, 95% CI 0.97–1.63, $p = 0.082$). In contrast, the

Table 2 Cox regression analysis for overall, cancer-specific, and disease-specific survival

	Overall survival				Cancer-specific survival				Disease-free survival			
	Univariate[A]		Multivariate[B]		Univariate[A]		Multivariate[B]		Univariate[A]		Multivariate[B]	
	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C
Treatment		0.065		0.319		0.010		0.762		0.017		0.765
Ø PDA	Reference		Reference		Reference		Reference		Reference		Reference	
PDA	1.28 (0.99–1.65)		0.87 (0.67–1.14)		1.63 (1.13–2.34)		0.94 (0.64–1.39)		1.34 (1.06–1.69)		0.96 (0.76–1.23)	
Age		0.001		0.008		0.749		0.460		0.004		0.022
< 65	Reference		Reference		Reference		Reference		Reference		Reference	
> 65	1.52 (1.18–1.96)		1.46 (1.10–1.94)		0.94 (0.66–1.35)		1.16 (0.78–1.75)		1.40 (1.11–1.76)		1.34 (1.04–1.73)	
Sex		0.047		0.023		0.059		0.204		0.022		0.016
Male	Reference		Reference		Reference		Reference		Reference		Reference	
Female	0.77 (0.60–1.00)		0.74 (0.57–0.96)		0.70 (0.48–1.02)		0.78 (0.53–1.15)		0.76 (0.60–0.96)		0.74 (0.58–0.95)	
ASA		< 0.001		< 0.001		0.003		0.006		< 0.001		< 0.001
II	Reference		Reference		Reference		Reference		Reference		Reference	
III/IV	2.45 (1.91–3.14)		2.26 (1.73–2.96)		1.72 (1.21–2.46)		1.74 (1.18–2.58)		2.03 (1.62–2.55)		1.93 (1.51–2.46)	
BMI		0.453		0.845		0.816		0.155		0.277		0.698
< 30	Reference		Reference		Reference		Reference		Reference		Reference	
> 30	0.88 (0.64–1.23)		1.03 (0.73–1.46)		1.06 (0.67–1.66)		1.43 (0.89–2.30)		0.85 (0.62–1.15)		0.94 (0.68–1.29)	
AJCC stage		< 0.001		< 0.001		< 0.001		< 0.001		< 0.001		< 0.001
I	Reference		Reference		Reference		Reference		Reference		Reference	
II	1.45 (0.93–2.25)		1.16 (0.74–1.82)		0.98 (0.41–2.38)		0.87 (0.36–2.13)		1.33 (0.92–1.93)		1.15 (0.78–1.68)	
III	2.11 (1.39–3.20)		1.93 (1.24–3.00)		2.58 (1.21–5.48)		2.50 (1.14–5.47)		1.63 (1.14–2.34)		1.50 (1.02–2.22)	
IV	7.45 (4.95–11.22)		7.10 (4.44–11.34)		17.01 (8.48–34.11)		18.48 (8.49–40.25)		5.69 (3.99–8.12)		5.11 (3.38–7.74)	
Grading		0.002		0.205		0.025		0.937		0.004		0.129
G1/2	Reference		Reference		Reference		Reference		Reference		Reference	
G3/4	1.54 (1.18–2.01)		1.20 (0.91–1.59)		1.57 (1.07–2.31)		1.02 (0.67–1.54)		1.45 (1.13–1.86)		1.23 (0.95–1.59)	
CEA		< 0.001		< 0.001		< 0.001		0.019		< 0.001		< 0.001
C0 stage < 5 ng/ml	Reference		Reference		Reference		Reference		Reference		Reference	
C1 stage > 5 ng/ml	3.03 (2.31–3.96)		1.87 (1.40–2.49)		3.74 (2.53–5.52)		1.69 (1.11–2.58)		2.53 (1.98–3.24)		1.67 (1.28–2.18)	
Unknown	2.44 (1.72–3.45)		1.83 (1.28–2.62)		2.33 (1.35–4.03)		1.84 (1.05–3.24)		1.92 (1.39–2.67)		1.56 (1.12–2.19)	
Operation		0.395		0.075		0.359		0.002		0.353		0.058
Right colectomy	Reference		Reference		Reference		Reference		Reference		Reference	
Transverse colectomy	0.89 (0.57–1.39)		0.84 (0.53–1.32)		0.83 (0.42–1.62)		0.65 (0.32–1.32)		0.89 (0.59–1.35)		0.86 (0.57–1.32)	
Left colectomy	0.82 (0.62–1.08)		0.78 (0.58–1.04)		0.76 (0.51–1.15)		0.58 (0.38–0.89)		0.84 (0.65–1.08)		0.80 (0.61–1.04)	
Subtotal colectomy	1.12 (0.73–1.69)		1.38 (0.90–2.12)		1.25 (0.71–2.21)		1.96 (1.08–3.56)		1.16 (0.79–1.71)		1.40 (0.95–2.07)	

Table 2 (continued)

	Overall survival				Cancer-specific survival				Disease-free survival			
	Univariate[A]		Multivariate[B]		Univariate[A]		Multivariate[B]		Univariate[A]		Multivariate[B]	
	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C	HR (95% CI)	P^C
Surgery		0.001		0.097		0.061		0.944		0.003		0.171
Open	Reference		Reference		Reference		Reference		Reference		Reference	
Laparoscopic	0.32 (0.14–0.73)		0.53 (0.24–1.21)		0.44 (0.16–1.18)		0.96 (0.35–2.67)		0.42 (0.21–0.81)		0.64 (0.33–1.26)	
Adjuvant CHT		0.024		0.131		< 0.001		0.575		0.014		0.481
No	Reference		Reference		Reference		Reference		Reference		Reference	
Yes	1.39 (1.05–1.82)		0.78 (0.57–1.08)		2.36 (1.64–3.40)		0.89 (0.58–1.35)		1.39 (1.08–1.80)		0.90 (0.67–1.21)	

AJCC American Joint Committee on Cancer, *ASA* American Society of Anesthesiologists score, *BMI* body mass index, *CEA* carcinoembryonic antigen, *CHT* chemotherapy, *CI* confidence interval, *HR* hazard ratio of death or recurrence, *PDA* peridural analgesia, *OR* odds ratio

[A] Univariate Cox regression analysis

[B] Multivariate Cox regression analysis full model

[C] Likelihood ratio tests

risk of cancer-specific mortality was significantly increased by 75% (HR 1.75, 95% CI 1.16–2.64, $p = 0.008$) in patients treated with PDA. After risk adjusting for the confounding factors and stratification for the year of diagnosis also in multivariable Cox regression analysis, PDA did not have an impact on overall (HR 0.89, 95% CI 0.65–1.21, $p = 0.415$), cancer-specific (HR 1.08, 95% CI 0.69–1.68, $p = 0.730$), and disease-free survival (HR 0.97, 95% CI 0.73–1.29, $p = 0.818$). Also when incorporating the AJCC stage as a second strata in addition to the strata term for the year of diagnosis in univariate and multivariable Cox regression analysis, the use of PDA had no impact on overall (HR 0.93, 95% CI 0.70–1.23, $p = 0.612$ and HR 0.90, 95% CI 0.66–1.24, $p = 0.515$), cancer-specific (HR 1.04, 95% CI 0.68–1.59, $p = 0.857$ and HR 1.12, 95% CI 0.71–1.75, $p = 0.635$), and disease-free survival (HR 1.00, 95% CI 0.77–1.29, $p = 0.972$ and HR 0.94, 95% CI 0.71–1.24, $p = 0.662$). In none of these analyses, PDA was selected as a significant predictor in the backward variable selection procedure. In this sensitivity analysis, the propensity score for patients treated with PDA was 0.404 ± 0.159 compared with 0.290 ± 0.145 in those without PDA ($p < 0.001$). During the propensity score matching procedure, 4 patients treated with PDA and 33 patients treated without PDA had to be excluded because their characteristics could not be matched with patients from the other group. After the matching procedure, the propensity score was virtually the same in the two patient groups (0.396 ± 0.152 for patients treated with PDA compared with 0.396 ± 0.151 for patients treated without PDA [$p = 0.983$]). After adjusting the data according to the propensity score analysis, it was revealed that the use of PDA had no impact on overall (HR 0.90, 95% CI 0.66–1.23, $p = 0.499$), cancer-specific (HR 1.18, 95% CI 0.77–1.79, $p = 0.454$), and disease-specific survival (HR 0.84, 95% CI 0.62–1.13, $p = 0.239$).

To further corroborate the data and to investigate if treatment with PDA had a different impact on survival in the subgroups of patients with ASA III/IV and ASA I/II, the complete analysis was repeated for these two different subgroups also excluding patients operated between 2011 and 2014 when PDA was not used.

First, a total of 271 patients classified as ASA III and IV were included. Of these, 101 patients received a PDA, and 170 patients received conventional intraoperative general and postoperative intravenous and oral analgesia. In unadjusted Cox proportional hazards regression analysis, a PDA had no significant impact on overall (HR 1.08, 95% CI 0.77–1.51, $p = 0.665$), cancer-specific (HR 1.54, 95% CI 0.91–2.62, $p = 0.112$), and on disease-free survival (HR 1.08, 95% CI 0.78–1.49, $p = 0.636$). Also after risk adjustment in multivariable Cox regression analysis, PDA did not have an impact on overall (HR 0.89, 95% CI 0.61–1.30, $p = 0.540$), cancer-specific (HR 0.87, 95% CI 0.46–1.64, $p = 0.667$), and disease-free survival (HR 0.85, 95% CI 0.59–1.23, $p = 0.390$). In this sensitivity

analysis, the propensity score for patients treated with PDA was 0.468 ± 0.196 compared with 0.316 ± 0.158 in those without PDA ($p < 0.001$). During the propensity score matching procedure, 4 patients treated with PDA and 7 patients treated without PDA had to be excluded because their characteristics could not be matched with patients from the other groups. After the matching procedure, the propensity score was virtually the same in the two patient groups (0.453 ± 0.184 for patients treated with PDA compared with 0.452 ± 0.185 for patients treated without PDA [$p = 0.986$]). After adjusting the data according to the propensity score analysis, the use of PDA had no impact on overall (HR 1.00, 95% CI 0.66–1.52, $p = 0.983$), cancer-specific (HR 1.38, 95% CI 0.75–2.54, $p = 0.303$), and disease-specific survival (HR 0.90, 95% CI 0.60–1.34, $p = 0.603$) in patients with ASA III and IV.

Second, a total of 365 patients classified as ASA I and II were included. Of these, 107 patients received a PDA, and 258 patients received conventional intraoperative general and postoperative intravenous and oral analgesia. In unadjusted Cox proportional hazards regression analysis, a PDA had no significant impact on overall (HR 1.27, 95% CI: 0.84–1.91, $p = 0.263$), cancer-specific (HR 1.63, 95% CI 0.95–2.78, $p = 0.082$), and on disease-free survival (HR 1.39, 95% CI 0.98–1.98, $p = 0.072$). Also after risk adjustment in multivariable Cox regression analysis, PDA did not have an impact on overall (HR 0.90, 95% CI 0.59–1.39, $p = 0.647$), cancer-specific (HR 1.16, 95% CI 0.65–2.08, $p = 0.609$), and disease-free survival (HR 1.00, 95% CI 0.69–1.45, $p = 0.987$). In this sensitivity analysis, the propensity score for patients treated with PDA was 0.370 ± 0.148 compared with 0.261 ± 0.142 in those without PDA ($p < 0.001$). During the propensity score matching procedure, 1 patient treated with PDA and 22 patients treated without PDA had to be excluded because their characteristics could not be matched with patients from the other group. After the matching procedure, the propensity score was virtually the same in the two patient groups (0.367 ± 0.144 for patients treated with PDA compared with 0.367 ± 0.144 for patients treated without PDA [$p = 0.988$]). After adjusting the data according to the propensity score analysis, the use of PDA had no impact on overall (HR 0.86, 95% CI 0.54–1.37, $p = 0.520$), cancer-specific (HR 0.98, 95% CI 0.56–1.71, $p = 0.932$), and disease-specific survival (HR 0.95, 95% CI 0.63–1.45, $p = 0.826$) in patients with ASA I and II.

Discussion

The current study represents the first to apply a propensity scoring method in order to investigate the putative impact of PDA on overall, cancer-specific, and disease-free survival in a large homogenous cohort of patients, with exclusively colon cancer. The present study revealed a strong bias with regard to patients' characteristics. After adjusting for these differences in propensity score-matched analysis, the initially found negative impact of PDA on cancer- and disease-specific survival in univariate analysis disappeared.

In contrast to Holler and colleagues' study [7] which reported a survival benefit in patients treated with PDA, the univariate analysis in this investigation revealed a poorer oncological outcome in those with PDA. These contradicting results can be contributed to significant imbalances in patients' characteristics in both studies. Whereas in the present analysis, patients treated with PDA tended to present with higher ASA scores and more advanced tumor stages, patients treated with PDA in the Holler et al. study were significantly younger and had lower ASA scores. However, also in contrast to the present study, in subgroup analysis, the overall survival benefit in Hollers' study was more pronounced in patients with ASA classification 3 to 4. When analyzing overall survival, concurring events of death have to be considered especially in old patients with co-morbidities. As shown in a study analyzing relative survival in patients with rectal cancer, only half of the observed deaths in patients with stages I to IV and only one third of the observed death in patients with stages I to III undergoing resection were related to cancer. Because the incidence of colorectal cancer increases with age and approximately 60% of all patients survive for 5 years, colorectal cancer patients have a relevant risk of dying from causes other than colorectal cancer itself [17]. Therefore, a perceived better overall survival does not necessarily mean that the oncological outcome is improved. In summary, to obtain valid results, every effort should be made to optimally adjust for all known confounders. As seen in this study, when using multivariable Cox regression and propensity score-matched analysis, it could be shown that the use of PDA did not significantly impact survival. The poor oncological outcome reported in the present analysis is, as such, a result of the imbalances between the two groups rather than the use of PDA itself.

The results of the present analysis also contradicted those reported in patients receiving radical prostatectomy and resection of ovarian serous adenocarcinoma. In these studies, patients treated with PDA had a significantly better oncological outcome with either a reduced risk of local recurrence or a longer overall survival when compared with patients operated on under general anesthesia alone without the addition of PDA [18, 19]. Furthermore, the excision of a malignant melanoma under general anesthesia was associated with decreased survival when compared with excision under local anesthesia [20]. However, a randomized controlled trial including 503 patients undergoing abdominal cancer surgery could not identify a significant improvement of cancer-free survival in patients treated with PDA [21]. In patients with colorectal cancer, the impact of PDA on oncological outcome is even more controversial, whereas one study was able to demonstrate a survival benefit within the first 2 years after colon cancer resection; another study was unable to confirm such a benefit [21, 22].

The underlying question is why should the use of PDA influence oncological outcome? General anesthesia and surgical stress may lead to immunodeficiency. This may be the result of direct suppression of the immune system, especially natural killer cell activity, or by activation of the hypothalamic–pituitary–adrenal axis and the sympathetic nervous system [23]. Furthermore, opioids, including morphine, have been shown to have immunosuppressive effects, thereby compromising cellular and humoral immune function and, thus, host defense against malignancies [24–26]. Therefore, the benefit of PDA use may lie in the decreased immunosuppressive effects of surgery and general anesthesia and by reducing the amount of perioperative necessary opioids required to control pain. In this context, cancer immunogenicity, defined as the ability of a tumor to induce an immune response, may play a central role. The underlying mechanisms are missense, frameshift, and splice site mutations, all leading to the expression of neoantigens. Presumably, tumor immunogenicity increases with the rate of mutations [27]. Consequently, immunomodulating mechanisms may affect various tumor entities in different ways. In comparison with immunogenic tumors like melanomas with high neoantigen expression, colon and rectum cancers show a much lower level of neoantigen expression [28] and therefore possess lower immunogenic tumor potential. Therefore, this effect may play a smaller role compared with other relevant parameters in rectum and colon cancers

We would like to acknowledge the limitations of the present investigation. First, this is a single-institution retrospective cohort study with the inherent risk of selection bias. However, it is questionable that a randomized controlled trial investigating the impact of PDA on oncological outcome in colon cancer patients will ever be performed. As such, a cohort study using the Cox proportional hazard regression analysis as well as the propensity scoring methods seems adequate. Second, the application of PDA decreased over time with the last PDA applied at the end of 2010. However, the possible bias arising from this change in practice was addressed in several sensitivity analyses. All the auxiliary analysis confirmed the previous results. Furthermore, the percentage of patients treated with PDA was low in our study population. It is possible that the low sample size may lead to bias or type II error. The exclusion of patients in the propensity score-matched analysis resulted in an additional loss of statistical power. Third, although risk adjustment was performed for a variety of observed confounding factors, potential bias due to unknown or unobserved confounders cannot be excluded. Finally, the follow-up for patients treated with PDA was significantly longer than for patients not treated with PDA, therefore possibly increasing outcome assessment bias.

In summary, in the present retrospective investigation, PDA in patients after colon cancer resection was not associated with a better oncological outcome after risk adjusting in multivariable Cox regression and propensity score analyses. Further studies investigating the putative effect of PDA on oncological outcome after colorectal cancer surgery are needed. The authors of the present study doubt that a randomized controlled trial investigating the impact of PDA on oncological outcome in colorectal cancer patients will ever be successfully performed. Therefore, a meta-analysis summarizing data from optimally adjusted cohort studies will provide the highest evidence level. In this respect, the present investigation may help further elucidate the possible association between PDA and oncological outcome. At present, a possibly better oncological outcome should not serve as a reason to treat patients with colon cancer with PDA.

Funding The study did not receive any funding.

Compliance with ethical standards

An independent ethics committee at the University Heidelberg approved the present study (S-649/2012).

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Wu CL, Cohen SR, Richman JM, Rowlingson AJ, Courpas GE, Cheung K, Lin EE, Liu SS (2005) Efficacy of postoperative patient-controlled and continuous infusion epidural analgesia versus intravenous patient-controlled analgesia with opioids: a meta-analysis. Anesthesiology 103(5):1079–1088 quiz 1109-1010
2. Guay J, Nishimori M, Kopp SL (2016) Epidural local anesthetics versus opioid-based analgesic regimens for postoperative gastrointestinal paralysis, vomiting, and pain after abdominal surgery: a Cochrane review. Anesth Analg 123(6):1591–1602. https://doi.org/10.1213/ANE.0000000000001628
3. Popping DM, Elia N, Van Aken HK, Marret E, Schug SA, Kranke P, Wenk M, Tramer MR (2014) Impact of epidural analgesia on mortality and morbidity after surgery: systematic review and meta-analysis of randomized controlled trials. Ann Surg 259(6):1056–1067. https://doi.org/10.1097/SLA.0000000000000237
4. Warschkow R, Steffen T, Luthi A, Filipovic M, Beutner U, Schmied BM, Muller SA, Tarantino I (2011) Epidural analgesia in open resection of colorectal cancer: is there a clinical benefit? A retrospective study on 1,470 patients. J Gastrointest Surg 15(8):1386–1393. https://doi.org/10.1007/s11605-011-1582-y
5. Grant MC, Yang D, Wu CL, Makary MA, Wick EC (2017) Impact of enhanced recovery after surgery and fast track surgery pathways on healthcare-associated infections: results from a systematic review and meta-analysis. Ann Surg 265(1):68–79. https://doi.org/10.1097/SLA.0000000000001703
6. Elsharydah A, Zuo LW, Minhajuddin A, Joshi GP (2017) Effects of epidural analgesia on recovery after open colorectal surgery. Proc (Bayl Univ Med Cent) 30(3):255–258
7. Holler JP, Ahlbrandt J, Burkhardt E, Gruss M, Rohrig R, Knapheide J, Hecker A, Padberg W, Weigand MA (2013) Peridural analgesia may affect long-term survival in patients with colorectal cancer after surgery (PACO-RAS-study): an analysis of a cancer registry.

Ann Surg 258(6):989–993. https://doi.org/10.1097/SLA.0b013e3182915f61
8. van der Sijp MP, Bastiaannet E, Mesker WE, van der Geest LG, Breugom AJ, Steup WH, Marinelli AW, Tseng LN, Tollenaar RA, van de Velde CJ, Dekker JW (2016) Differences between colon and rectal cancer in complications, short-term survival and recurrences. Int J Color Dis 31(10):1683–1691. https://doi.org/10.1007/s00384-016-2633-3
9. Hohenberger W, Weber K, Matzel K, Papadopoulos T, Merkel S (2009) Standardized surgery for colonic cancer: complete mesocolic excision and central ligation—technical notes and outcome. Color Dis 11(4):354–364; discussion 364-355. https://doi.org/10.1111/j.1463-1318.2008.01735.x
10. Pox C, Aretz S, Bischoff SC, Graeven U, Hass M, Heussner P, Hohenberger W, Holstege A, Hubner J, Kolligs F, Kreis M, Lux P, Ockenga J, Porschen R, Post S, Rahner N, Reinacher-Schick A, Riemann JF, Sauer R, Sieg A, Scheppach W, Schmitt W, Schmoll HJ, Schulmann K, Tannapfel A, Schmiegel W (2013) Leitlinienprogramm Onkologie der A, Deutschen Krebsgesellschaft e V, Deutschen Krebshilfe e V (2013) [S3-guideline colorectal cancer version 1.0]. Z Gastroenterol 51(8):753–854. https://doi.org/10.1055/s-0033-1350264
11. Ishwaran H, Kogalur UB, Blackstone EH, Lauer MS (2008) Random survival forests. Ann Appl Stat 2(3):841–860
12. Joffe MM, Rosenbaum PR (1999) Invited commentary: propensity scores. Am J Epidemiol 150(4):327–333
13. Rosenbaum PR (1987) Model-based direct adjustment. J Am Stat Assoc 82(98):387–394
14. Rosenbaum PR (1989) Optimal matching for observational studies. J Am Stat Assoc 84(408):1024–1032
15. Hansen BB, Klopfer SO (2006) Optimal full matching and related designs via network flows. J Comput Graph Stat 15(3):609–627
16. Sekhon JS (2011) Multivariate and propensity score matching software with automated balance optimization: the matching package for R. J Stat Softw 42(7). doi:https://doi.org/10.18637/jss.v042.i07
17. Tarantino I, Muller SA, Warschkow R, Kulu Y, Schmied BM, Buchler MW, Ulrich A (2014) Baseline mortality-adjusted survival in resected rectal cancer patients. J Gastrointest Surg 18(10):1837–1844. https://doi.org/10.1007/s11605-014-2618-x
18. Lee BM, Singh Ghotra V, Karam JA, Hernandez M, Pratt G, Cata JP (2015) Regional anesthesia/analgesia and the risk of cancer recurrence and mortality after prostatectomy: a meta-analysis. Pain Manag 5(5):387–395. https://doi.org/10.2217/pmt.15.30
19. Lin L, Liu C, Tan H, Ouyang H, Zhang Y, Zeng W (2011) Anaesthetic technique may affect prognosis for ovarian serous adenocarcinoma: a retrospective analysis. Br J Anaesth 106(6):814–822. https://doi.org/10.1093/bja/aer055
20. Schlagenhauff B, Ellwanger U, Breuninger H, Stroebel W, Rassner G, Garbe C (2000) Prognostic impact of the type of anaesthesia used during the excision of primary cutaneous melanoma. Melanoma Res 10(2):165–169
21. Myles PS, Peyton P, Silbert B, Hunt J, Rigg JR, Sessler DI, Investigators ATG (2011) Perioperative epidural analgesia for major abdominal surgery for cancer and recurrence-free survival: randomised trial. BMJ 342:d1491. https://doi.org/10.1136/bmj.d1491
22. Christopherson R, James KE, Tableman M, Marshall P, Johnson FE (2008) Long-term survival after colon cancer surgery: a variation associated with choice of anesthesia. Anesth Analg 107(1):325–332. https://doi.org/10.1213/ane.0b013e3181770f55
23. Kurosawa S, Kato M (2008) Anesthetics, immune cells, and immune responses. J Anesth 22(3):263–277. https://doi.org/10.1007/s00540-008-0626-2
24. Franchi S, Panerai AE, Sacerdote P (2007) Buprenorphine ameliorates the effect of surgery on hypothalamus-pituitary-adrenal axis, natural killer cell activity and metastatic colonization in rats in comparison with morphine or fentanyl treatment. Brain Behav Immun 21(6):767–774. https://doi.org/10.1016/j.bbi.2007.01.001
25. Ishikawa M, Tanno K, Kamo A, Takayanagi Y, Sasaki K (1993) Enhancement of tumor growth by morphine and its possible mechanism in mice. Biol Pharm Bull 16(8):762–766
26. Sacerdote P, Bianchi M, Gaspani L, Manfredi B, Maucione A, Terno G, Ammatuna M, Panerai AE (2000) The effects of tramadol and morphine on immune responses and pain after surgery in cancer patients. Anesth Analg 90(6):1411–1414
27. Tian T, Olson S, Whitacre JM, Harding A (2011) The origins of cancer robustness and evolvability. Integr Biol (Camb) 3(1):17–30. https://doi.org/10.1039/c0ib00046a
28. Rajasagi M, Shukla SA, Fritsch EF, Keskin DB, DeLuca D, Carmona E, Zhang W, Sougnez C, Cibulskis K, Sidney J, Stevenson K, Ritz J, Neuberg D, Brusic V, Gabriel S, Lander ES, Getz G, Hacohen N, Wu CJ (2014) Systematic identification of personal tumor-specific neoantigens in chronic lymphocytic leukemia. Blood 124(3):453–462. https://doi.org/10.1182/blood-2014-04-567933