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Current Evidence of Single-Port Laparoscopic versus Single Port-Robotic Techniques in Colorectal Surgery: A Meta-Analysis

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Rezumat

Actualități privind tehnicile sigle-port laparoscopice și robotice în chirurgia colorectală: o meta-analiză

Introducere: Obiectivul principal al acestei meta-analize a fost compararea rezultatelor chirurgicale ale utilizării sistemelor de tip single port laparoscopice şi robotice în chirurgia colorectală.
Materiale şi Metode: Au fost identificate în literatura de specialitate studii care compară rezultatele operatorii şi datele de urmărire pe termen scurt pentru colectomia prin port unic efectuată laparoscopic versus robotic. Datele din studiile eligibile au fost extrase, evaluate calitativ şi incluse într-o meta-analiză. Au fost calculate odds ratios (OR) şi diferențele medii cu intervale de încredere de 95%.
Rezultate: Au fost incluse trei studii cu un total de 346 de pacienți (robotic: 112 cazuri versus laparoscopic: 234 cazuri). Nu s-au observat diferențe statistice semnificative în ceea ce priveşte morbiditatea generală, durata spitalizării şi complicațiile intra- şi postoperatorii între cele două grupuri. Totuşi, abordarea robotică a permis excizia unui număr mai mare de ganglioni limfatici în cazurile oncologice (SMD -0,25, IC 95% -0,50 la -0,01, p = 0,04, I^2 = 0%).
Concluzie: Atât tehnicile laparoscopice, cât şi cele robotice cu port unic par a fi opțiuni sigure şi fezabile în chirurgia colorectală, dovedind rezultate perioperatorii comparabile. Sunt necesare studii clinice randomizate de mai mare amploare pentru a justifica aplicarea lor, în special în ceea ce priveşte costurile procedurale.

Cuvinte cheie: chirurgie colorectală, chirurgie minim invazivă, laparoscopic, robotic, rezultat

Abstract

Introduction: The primary aim of this meta-analysis was to compare the operative outcomes of single-port laparoscopic versus single-port robotic platforms in colorectal surgery.

Materials and Methods: A comprehensive literature search was conducted for studies comparing operative outcomes and short-term follow-up data of single-port laparoscopic versus single-port robotic colectomy. Data from eligible studies were extracted, qualitatively assessed, and included in a meta-analysis. Odds ratios (ORs) and mean differences with 95 per cent confidence intervals were calculated.

Results: Three studies with a total of 346 patients (Robotic: 112 cases versus Laparoscopic: 234 cases) were included. There was no statistical difference noted with regard to overall morbidity, length of hospital stay and intra- and postoperative complications between the two groups. However, the robotic approach resulted in higher lymph nodes yield in oncologic cases (SMD -0.25, 95% CI -0.50 to -0.01, p = 0.04, I^2=0%).

Conclusion: Both single-port laparoscopic and robotic techniques appear to be safe and feasible options in colorectal surgery displaying comparable perioperative outcomes. Larger randomized controlled trials are needed to justify their application, particularly with regard to procedure-related costs.

Key words: colorectal surgery, minimally-invasive surgery, laparoscopic, robotic, outcome

Introduction

With an incidence of 10.2% and a mortality rate of 9.2%, colorectal cancer is the third most common cancer and the second leading cause of cancer-related mortality worldwide (1,2). Surgery is the gold standard for the treatment of resectable colorectal cancer (3). Minimally-invasive procedures have become a benchmark in colorectal surgery as they have been shown to offer several advantages compared to open surgery, such as less intraoperative blood loss, better postoperative recovery including less pain sensation, faster resumption of gastrointestinal motility and consecutively shorter hospital stay with equivalent oncologic outcomes (4,5). Conventional laparoscopic surgery is usually multiport and requires abdominal incisions for trocars and a mini-laparotomy incision for specimen removal. Each incision can be associated with pain and complications such as infection and abdominal wall hernias (6). The concept of single-incision laparoscopic surgery (SILS) was introduced in 2008 in an attempt to improve cosmetic results, optimise postoperative recovery and reduce surgical trauma and port-related complications (7-9). Despite its advantages and similar long-term oncological outcomes, SILS has not managed to replace multiport laparoscopy and is only performed by a few surgeons because of its technical difficulties such as loss of triangulation, poor exposure and retraction due to the limited number of instruments in situ and a more difficult learning curve (9-11). In recent years, robotic surgery has gained ground in the field of minimally-invasive colorectal surgery with its technical advantages including 3D vision, tremor stabilisation and better ergometry (3,12). As expected, trying to combine robotic surgery with single port minimally- invasive surgery, the first single-incision robotic colectomy was reported in 2009 (13). Following this trend, a single-arm, single-port robotic system has been developed, and Marks et al. (4) were first to report their experience with this new platform in colorectal surgery (4). The introduction of single-port robotic surgery aimed to further enhance the benefits of SILS by using the technical advantages of robotic surgery to overcome the technical difficulties of SILS (12). Although several publications suggest that SILS and single-port robotic surgery have similar short-term postoperative outcomes and similar oncological survival, few studies have compared the two surgical techniques (8,12,14). Therefore, the aim of this study was to perform a meta-analysis of studies comparing the complications, safety and postoperative recovery of SILS and single-port robotic surgery for colorectal procedures.

Material and Methods

This meta-analysis was conducted according to the current Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) checklist and the Cochrane Handbook for Systematic Reviews of Interventions (15,16).

Search Strategy

A systematic database search of Pubmed (Medline), Google scholar, and the Cochrane Central Trials Register was performed without time or language limitations. The following keywords were used in combination with the Boolean operators AND or OR: "Single-port", "single-incision", "sils", " laparosc*", " robotic", " colorectal", " colo*", " rect*". In addition, potentially relevant citations for the analysis were identified by screening the reference lists of the retrieved studies. Each selected abstract and study was independently assessed by two reviewers (S.V., and D.P.) for eligibility and inclusion in the meta-analysis. Disagreements were resolved either by consensus or by consulting a third author (M.C.S.) when necessary. The last literature search was conducted on 21 April 2024.

Eligibility Criteria

All original studies comparing single-port laparoscopic versus single-port robotic colorectal surgery for benign and malignant diseases were considered eligible. Studies had to report on at least one of the following procedure-related outcomes to be included in the meta-analysis: operative characteristics and the intraoperative course (including conversion rate, number of additional trocars, blood loss, and procedural time), postoperative complications, and GI-recovery parameters. Non-comparative irrelevant studies were excluded. In the case of duplication or overlap of articles from the same institution and authors, the most recent study was selected for inclusion.

Data Extraction

A self-developed electronic data extraction sheet was used independently by two authors (S.V., and M.C. S.) to enter all relevant data, if complete, from studies meeting the eligibility criteria. Disagreements were resolved by consensus or third author (D.P.) reassessment. From each included study, the following data were retrieved:

1. General study characteristics: first author, year and country of origin, study design, enrollment period, number of considered patients in each group, inclusion and exclusion criteria, follow-up period, study endpoints
2. Demographics: Age, sex, BMI (body mass index), ASA (American Society of Anesthesiologists) classification, comorbidities, previous abdominal surgery, neoadjuvant chemotherapy, pelvic radiation, tumor pathology and location, TNM stage, resection margin, specimen length.
3. Technical Aspects and Operative Characteristics: type of access (robotic or laparoscopic), surgery duration, conversion rate to open surgery, intraoperative blood loss, number of harvested lymph nodes.

4. Postoperative Complications: intra-readmission rate, intraluminal bleeding, intraperitoneal abscess, postoperative ileus, intraoperative morbidity
5. Postoperative recovery data: time to first postoperative bowel movement or flatus, and length of hospital stay

The primary outcomes of this study were overall morbidity, and length of hospital stay. The secondary outcomes were as follows: readmission rate, surgery duration, conversion to open surgery, intraluminal bleeding, intraoperative blood loss, intraperitoneal abscess, postoperative ileus, intraoperative morbidity and number of harvested lymph nodes.

Quality and Certainty Assessment

The risk of bias of the included non-randomised trials was assessed by two authors (S.V. and D.P.) independently of each other using the ROBINS-I tool, which consists of 7 different domains of bias at 3 time points: Pre-intervention (confounding and selection of participants), at intervention (classification of interventions) and post-intervention (bias due to deviations from the intended interventions, missing data, measurement of outcomes and selection of the reported outcome) (17). Each study's risk of bias was accordingly categorised as low, moderate, severe or critical. The reviewers were not blinded to the study authors. Disagreements in grading studies were discussed and resolved by consensus or by third author (M.C.S.) reassessment. The Grading of Recommendations, Assessment, Development, and Evaluation (GRADE) methodology was used to adequately document the strength of evidence for the main significant outcomes, implementing four levels of evidence (high, moderate, low, and very low) (18,19).

Statistical Analysis

Statistical analysis was performed using RevMan software (version 5.3. Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2014) according to the recommendations of the Cochrane Collaboration guidelines. Pairwise meta-analyses were conducted. For each outcome of interest, summary estimates of the treatment effect were calculated with 95% confidence intervals (CIs). The odds ratios (ORs) were chosen for outcomes that were dichotomous. Standardised mean differences (SMDs) were calculated for the analysis of continuous outcomes. For continuous variables, the available data on medians and IQRs have been converted into means and standard deviations using the method proposed by Luo et al. (20). Of note, continuous values were expressed in days (time to first bowel movement/flatus and length of hospital stay) and minutes (operative time). The conversion rate was defined as any conversion to open or multiport surgery requiring additional ports during the operative course. Using the Cochrane Q test (chi-squared test; chi^2) and the measurement of inconsistency (I^2), the degree of heterogeneity among the included studies was interpreted as follows: 0%-40% low heterogeneity and may not be important, 30%-60% moderate heterogeneity, 50%-90% substantial heterogeneity, > 75% high heterogeneity (21). If heterogeneity was low or moderate ($I^2 < 50\%$), summary estimates were calculated using a fixed-effects method. When $I^2 > 50\%$, the randomised model was used. Summary estimates were calculated with a fixed-effects method in case of low or moderate heterogeneity ($I^2 < 50\%$). Tests for publication bias or funnel plots were not performed because of the small number of studies included. P values < 0.05 of pooled data were considered significant.

Results

The study selection flowchart is shown in *Fig.1.* The initial electronic database search identified 5816 articles, of which three studies (8,12,14) met the inclusion criteria and were eligible for the final meta-analysis.

Study and Patient Characteristics

A total of 346 patients (male/female: 175/171)

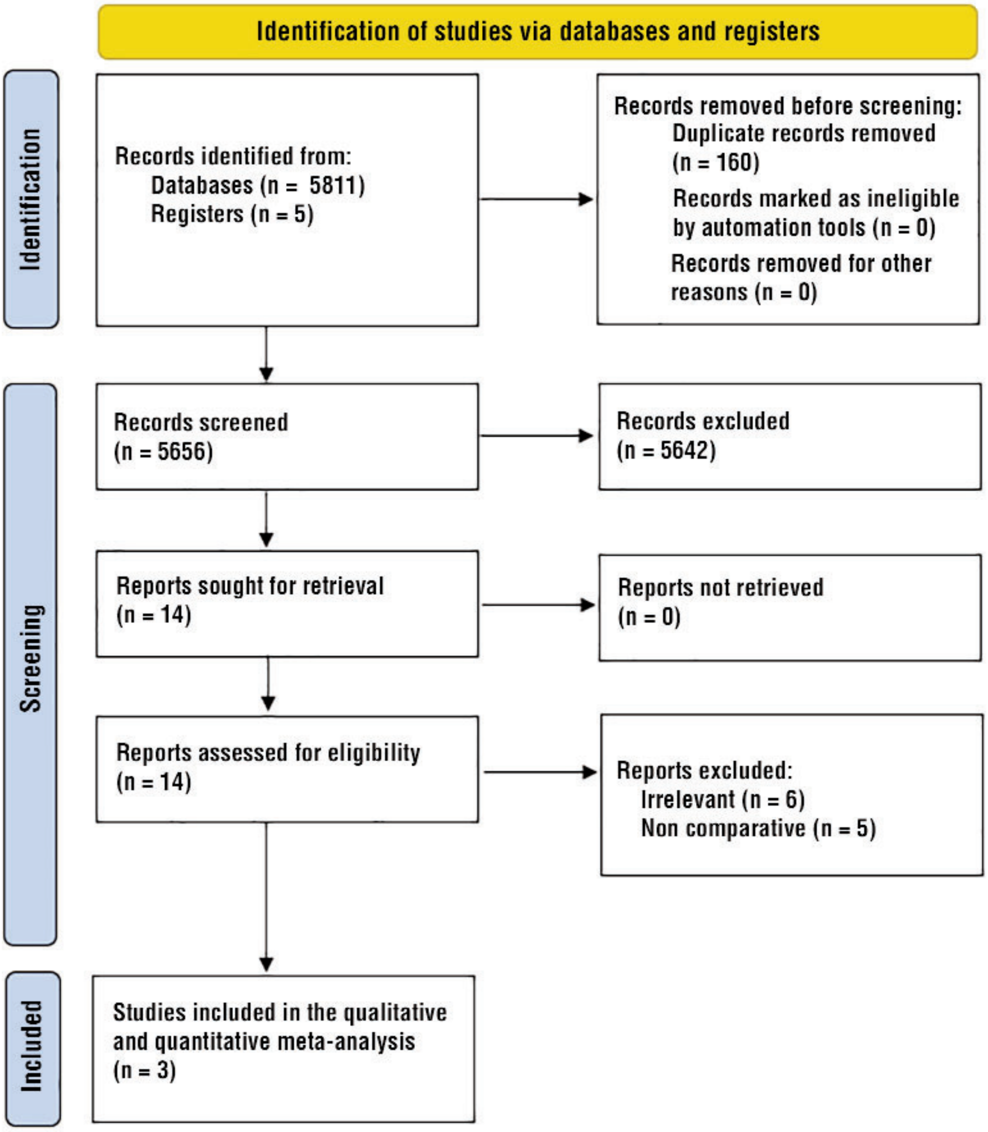

Figure 1. PRISMA diagram of study identification and selection for review analysis

originating from three different countries (South Korea, Taiwan, and USA) that underwent single-incision laparoscopic colectomy or single-port robotic colectomy (Robotic: 112 cases versus Laparoscopic: 234 cases) were included in three studies (8,12,14). The study enrolment period was from January 2010 to January 2022. Except for one study with a multi-center design (8), the other two studies were single-center studies (12,14). The operative indication was malignant tumors in 3 studies while Chang et al. (8) also included seven cases with benign diseases. One study (14) only included cases of right and extended right hemicolectomy, while the other two (8,12) included a variety of right and left-sided colorectal operations (Chang et al (8): Right-sided hemicolectomy: 35, Left Hemicolectomy: 5, Anterior Resection: 62; Keller et al (12): Right colectomy: 12, Left colectomy: 10, Transanal Transabdominal radical proctosigmoidectomy: 52, Low anterior resection: 24,

Table 1. Study characteristics and protocols

Author	Year	Origin	Study period	Study design	da Vinci system	Total sample size	Follow up period
Lim et al. (2023)(14)	2023	Korea	January 2019 - December 2020	single-center, prospective	da Vinci Sp	151	30 days after discharge
Chang et al.(2021)(8)	2021	Taiwan	Robotic: January 2017 - December 2019 Laparoscopic: January 2013 - December 2018	multi-center, retrospective (matched case control study)	da Vinci Si	105	30 days after discharge
Keller et al. (2024)(12)	2024	USA	January 2010 - December 2022	single-center, retrospective (matched cohort study)	NA	100	30 days after discharge

Abdominoperineal resection: 2). In all of the studies the follow up period was 30 days after discharge. The complete study and patient characteristics are summarized in *Table 1* and *Table 2*.

Study Quality and Risk of Bias

The main quality-limiting factor of the included studies was the non-randomized patient allocation process. While Chang et al. (8) and Keller et al. (12) conducted a retrospective study, Lim et al. (14) presented their results in a prospectively designed work. Propensity matching was performed in two (8,12) of the three studies. According to the ROBINS-I tool, the overall risk of bias was low to moderate (*Fig. 2*).

Primary Outcome Analysis

The primary outcomes of this meta-analysis were overall morbidity and length of hospital stay. Both outcomes were reported in all three studies (8,12,14). The meta-analysis of the pooled data for overall morbidity and length of stay showed no significant differences between

Table 2. Demographic data and characteristics of the included patients

Author	Groups	No. of patients	Age (years) mean/SD	Gender (M/F)	BMI (kg/m^2) mean/SD	ASA score	Previous abdominal surgery	Operative indication		Number of surgeons	Operation performed	T-Stage
								Malignant	Benign			
Lim et al. (2023)(14)	Robotic	41	60.80±11.17	20/21	23.80±3.16	I: 9 II: 30 III: 2	10	41	0	Single	Right hemicolectomy: 26 Extended right hemicolectomy: 15	Tis-2: 13 T3-4: 28
	Laparoscopic	100	65.74±11.48	46/54	23.79±3.13	I: 19 II:64 III:17	22	100	0	Single	Right hemicolectomy: 62 Extended right hemicolectomy: 38	Tis-2: 31 T3-4:69
Chang et al. (2021) (8)	Robotic	21	62.1±11.9	9/12	24.7±3.9	I-II: 20 III-IV: 76	NA	19	2	2	Right hemicolectomy: 7 Left hemicolectomy: 1 AR: 13	NA
	Laparoscopic	84	65.6±13.6	43/41	24.6±3.1	I-II: 1 III-IV: 26	NA	79	5	2	Right hemicolectomy: 28 Left hemicolectomy: 4 AR: 52	NA
Keller et al. (2024) (12)	Robotic	50	60.708± 10.686	29/21	26.135± 6.030	II:27 III:21 IV:2	10	50	0	NA	Right colectomy: 6 Left colectomy: 5 TATA: 20 LAR: 18 APR: 1	T0:7 T1:4 T2:16 T3: 23
	Laparoscopic	50	58.584± 12.213	28/22	27.834± 6.87	II:28 III:21 IV:1	12	50	0	NA	Right colectomy: 6 Left colectomy: 5 TATA: 32 LAR: 6 APR: 1	T0:9 T1:4 T2:24 T3:13

ASA score: American Society of Anesthesiologists; BMI: Body max index; SD: Standard deviation; AR: Anterior Resection; LAR: Low anterior resection; TATA: Transanal Transabdominal radical proctosigmoidectomy; APR: Abdominoperianeal resection

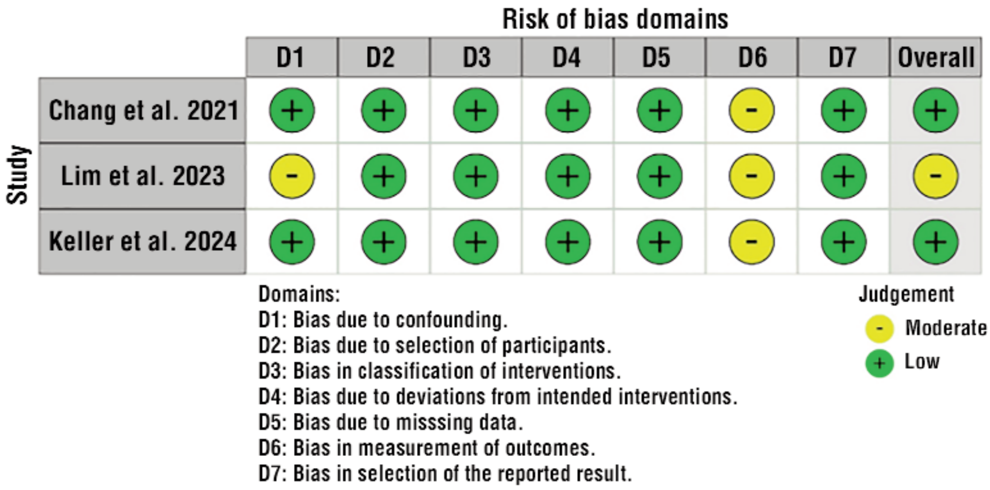

Figure 2. Risk of bias summary and graphical visualization of the included studies based on ROBINS-I-tool

SILS and single-port robotic colo-rectal surgery (*Fig. 3 A, B*).

Secondary Outcome Analysis

Number of harvest lymph nodes

The number of harvest lymph nodes was reported in all three studies (8,12,14). Fewer lymph nodes were harvested in the laparoscopic group compared to the robotic group (SMD -0.25, 95% CI -0.50 to -0.01, p=0.04) (*Fig. 4*). A low level of heterogeneity was observed (I^2=0%, Chi^2 test: p=0.39). Of note, it is important to point out that Keller et al. (12) only documented the number of lymph nodes removed in patients with rectal cancer. The quality of evidence for the number of harvest lymph nodes was low respect to the GRADE criteria (*Table S1*).

Non-significant secondary outcomes

Meta-analysis of the secondary outcomes of

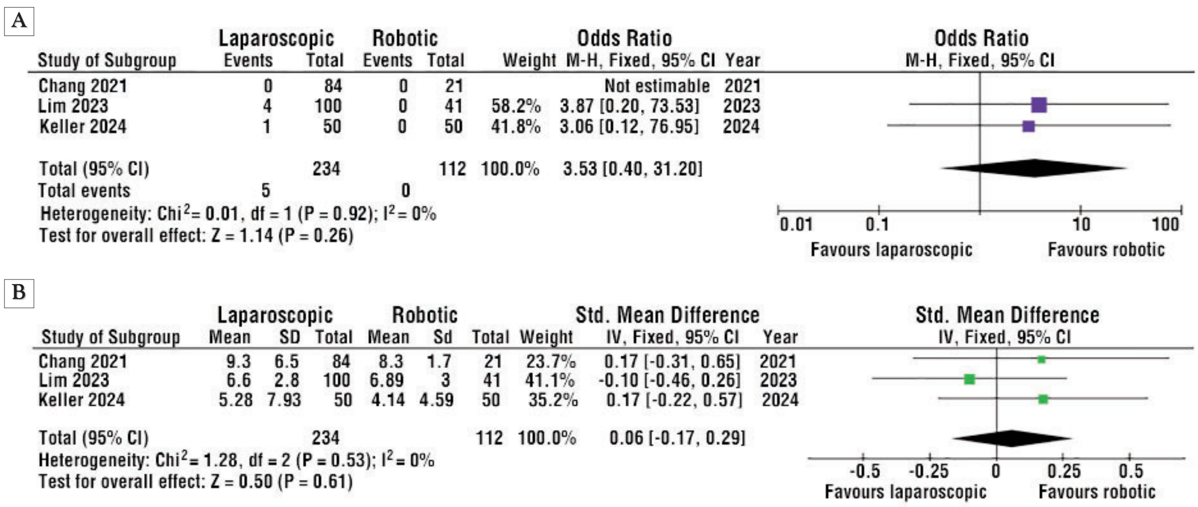

Figure 3. Forest plots of primary outcomes (Laparoscopic Versus Robotic): (A) overall morbidity; (B) length of hospital stay

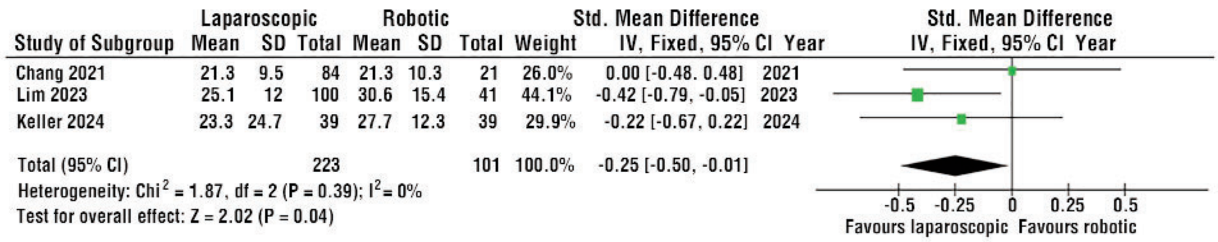

Figure 4. Forest plots of significant secondary outcome: number of harvest lymph nodes

interest revealed no statistically significant difference between the laparoscopic and robotic group in readmission rate, surgery duration, conversion to open surgery, intraluminal bleeding, intraoperative blood loss, intraperitoneal abscess, and postoperative ileus (*Table 3*).

However, substantial heterogeneity for intraoperative blood loss (*Fig. 5*) was detected. The source of heterogeneity was the study by Lim et al. (14) who only included right-and extended right hemicolectomies performed by a single surgeon. When considering the studies by Keller et al (12). and Chang et al. (8) with a higher proportion of left-sided and rectal resections, the robotic platform seems to have a lower intraoperative blood loss volume.

Discussion

The present study shows, to our knowledge, the first pooled surgical outcome analysis of single-incision laparoscopic colectomy versus single-port robotic colorectal resection. The cumulative results of three included studies (8,12,14) with 346 patients showed no significant difference between single-incision laparoscopic and single-port robotic colectomies in terms of overall morbidity and hospital stay. At the same time, important intraoperative and short-term postoperative outcomes such as operative time, conversion to open or multiport surgery, intraluminal bleeding, intraoperative blood loss, intraperitoneal abscess, postoperative ileus and readmission rate were not significantly different between the two groups. These findings are similar with some recently published studies that compare single-incision laparoscopic surgery with single-port robotic surgery in other surgical procedures such as hysterectomy, cholecystectomy and adrenalectomy (22–25).

Our study showed that the number of harvested lymph nodes as a measure of the

Table 3. Non-significant secondary outcomes

Outcomes	No. of included studies	No. of included patients		SMD/OR [95 % CI]	P-value	Heterogeneity level	
		Laparoscopic	Robotic			I² (%)	P-value
Readmission rate	3 (8,12,14)	234	112	3.53[0.40, 31.20]	0.26	0	0.92
Surgery Duration	3 (8,12,14)	234	112	-0.18[-0.85, 0.49]	0.60	87	0.0004
Conversion	3 (8,12,14)	234	112	2.33[0.40, 13.64]	0.35	0	0.70
Intraluminal Bleeding	3 (8,12,14)	234	112	0.87[0.02, 31.57]	0.94	62	0.11
Intraoperative Blood loss	3 (8,12,14)	234	112	0.01[-0.90, 0.91]	0.99	93	<0.00001
Intraperitoneal Abscess	3 (8,12,14)	234	112	1.16[0.19, 7.25]	0.87	0	0.32
Postoperative Ileus	3 (8,12,14)	234	112	1.97[0.62, 6.24]	0.25	0	0.39
Intraoperative Morbidity	3 (8,12,14)	234	112	3.14[0.37, 26.82]	0.30	0	0.64

OR Odds ratio; SMD Standardized mean difference

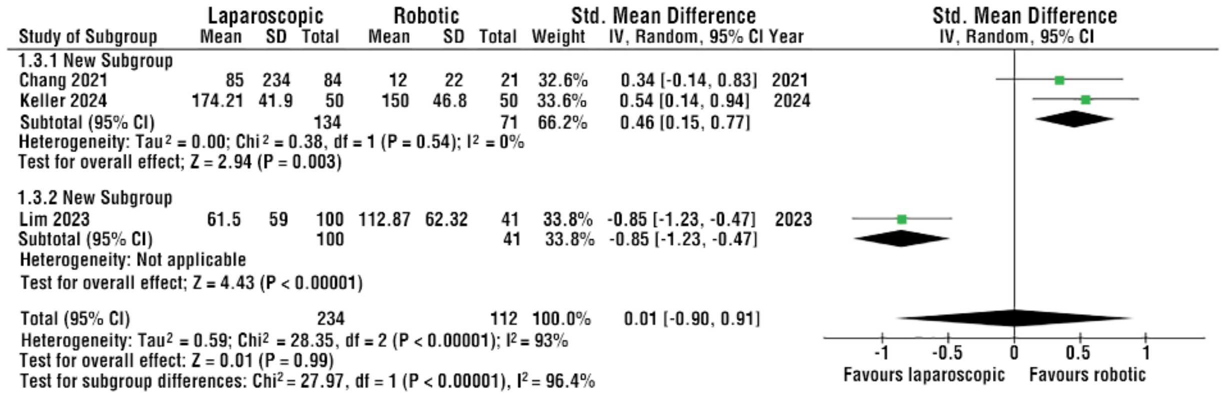

Figure 5. Forest plots of secondary outcome: intraoperative blood loss

oncological quality of a surgical procedure (25) was higher in the robotic group than in the laparoscopic group. These results are in line with recently published data, mostly referring to rectal cancer cases, comparing robotic and laparoscopic surgery (26–29). One possible explanation for this finding may rely in the fact that Keller et al. (12) analysed lymph nodes harvest exclusively in their rectal subgroup. Furthermore, in robotic colorectal surgery a higher number of lymph nodes is yielded, possibly because of the technical advantages of the approach, such as high-definition views and improved ergonomics. Conventional rigid instruments designed for laparoscopy offer five degrees of freedom whereas the robotic platform adds two independent degrees of freedom, enabling unrestricted directional steering. In light of this, it can be hypothesized that it is technically more feasible to dissect central lymph nodes and vessels more radically implementing a robotic platform, with this effect being amplified in the single-port – reduced triangulation setting, offering better technical capabilities in terms of CME, TME and lateral pelvic lymph node dissection. (26,29,30).

Colorectal surgery has evolved tremendously in recent decades from an open to a minimally- invasive approach (31). The development of minimally-invasive technology has been rapid over the last 20 years following the introduction of the robotic approach(32). A number of studies have demonstrated procedural advantages of robotic surgery for the colon and rectum, such as a stable 3D view controlled by the lead surgeon, technology that eliminates tremor, improved ergonomics and an easier learning curve than laparoscopic surgery (33). In addition, length of hospital stay and complications are comparable between robotic and laparoscopic colorectal surgery (34). Regarding oncological outcomes, there are several studies that show oncological equivalence between the two techniques (11,35,36).

As a result of the above, the number of robotic colorectal procedures performed has increased over the years, but the majority of minimally-invasive colorectal procedures is still performed laparoscopically (34). The main reason for this is also the main disadvantage of robotic surgery: the considerably higher operational cost compared to laparoscopy. This is supported by the meta-analysis by Singh et al comparing the costs of robotic and laparoscopic colorectal surgery (37). This meta-analysis, which included 13 studies, one randomised trial and 12 comparative studies, published between 2012 and 2023, showed that laparoscopic colorectal surgery appears to be more economical compared to robotic colorectal surgery in terms of both operative and total costs (37). However, it should be noted that the trials included in the meta-analysis showed increased heterogeneity (37). Besides high cost, another disadvantage of robotic colorectal surgery is the longer operation time which is mainly caused by the limited range of motion combined with the relatively slow movement of the robotic arm and the docking time (38,39). The longer operating time in robotic colorectal surgery is supported in the meta-analysis of Lorenzon et al. (40). The results of meta-analysis that included 22 studies with 2772 patients, in which a variety of colorectal procedures (right hemicolectomy, left hemicolectomy and pelvic resection) were examined, supported the point that robotic colectomies are associated with longer operation time in comparison to laparoscopic colectomies (40).

To date, single-port minimally-invasive colorectal surgery has not become widespread, mainly because of the technical disadvantages of one-port laparoscopy. Due to the technical advantages of robotic surgery as described above, the introduction of single-port robotic arms has opened new horizons for single-port minimally invasive colorectal surgery. The considerations reported between laparoscopic and robotic colorectal surgery also exist in the debate between laparoscopic versus robotic single-port colorectal surgery. The fundamental question is whether the single port technique offers the proper setting, in which the dexterity of the robotic system is fully utilized and becomes obvious when compared to the rigid single port laparoscopy, particularly in terms of operating time and procedure cost.

When interpreting the results of this meta-analysis, several limitations must be taken into account. Firstly, the study design of all included studies in this meta-analysis was retrospective, which consequently introduces relevant selection bias and limits the strength of the final conclusions. Secondly, the sample sizes of the available trials were relatively small, and in two of the trials (8,14), the number of patients in the laparoscopic group was much higher than in the robotic group. Another important concern is the heterogeneity of the studies reviewed in terms of different tumour locations and therefore different colorectal procedures. In addition, each of the included studies only reported short-term outcomes and did not assess quality of life or long-term survival. A cost-benefit analysis was not performed in any of the included studies. Finally, the differences in comorbidities between the laparoscopic and the robotic group, the learning curve, the surgeon's experience and the application of different surgical instruments may introduce some biases that are difficult to avoid and control for. However, the present meta analysis is the first one to compare the two approaches of single-port surgery in colorectal cases, performed with rigogous statistical methodology, offering valuable insights on this controversial subject.

Conclusions

According to our meta-analysis, single-port robotic and single-port laparoscopic approaches in colorectal surgery are both feasible and safe procedures demonstrating similar overall morbidity and hospital stay. Laparoscopic single port approach was generally not found to be inferior to robotic single port surgery, raising justifiable concerns in our current challenging economic landscape. However, the results of this meta-analysis are based on a very limited number of studies. Therefore, to elucidate the true value of the single-port robotic platform in colorectal surgery, there is a definite need for more high-quality randomized studies.

Author's Contributions

Study conception and design (S.V., and D.P.), Literature search and study selection (S.V., D.P., and M.C.S.), Acquisition of data (S.V., D.P., and M.C.S.), Analysis and interpretation of data (S.V., and D.P.), Statistical analysis (D.P.), Drafting of manuscript (M.C.S., and S.V.), Critical revision of manuscript (D.P., and W.T.K.). All authors have read and agreed to the published version of the manuscript.

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Conflict of Interest Statement

The authors declare that they have no conflict of interest.

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Ethics Approval

For this type of study, no ethical approval was required and obtained.

Informed Consent

For this type of study, informed consent was not required.

Data Availability Statement

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

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Supplementary Material

Table S1. The GRADE Certainty assessment for the significant outcomes

Outcomes	No. of studies	No. of included patients		SMD [95 % CI]	Quality assessment					Quality
		Laparoscopic	Robotic		Risk of bias[a]	Inconsistency	Indirectness	Imprecision	Publication bias	
Number of harvest lymph nodes	3 (8,12,14)	223	101	-0.25 [-0.50 to -0.01]	Not serious	Not serious	No indirectness	No imprecision	NA	Low

NA: not applicable, SMD: standardized mean difference
[a]Risk of bias assessed using the ROBINS-I tool.