ISSN 1948-5204 (online)

World Journal of *Gastrointestinal Oncology*

World J Gastrointest Oncol 2018 October 15; 10(10): 282-366

Published by Baishideng Publishing Group Inc

WJGO World Journal of Gastrointestinal Oncology

Contents

Monthly Volume 10 Number 10 October 15, 2018

ABOUT COVER

Editorial Board Member of *World Journal of Gastrointestinal Oncology*, Jong Park, PhD, Associate Professor, Division of Cancer Prevention and Control, H. Lee Moffitt Cancer Center, College of Medicine, University of South Florida, Tampa, FL 33612, United States

AIM AND SCOPE

World Journal of Gastrointestinal Oncology (*World J Gastrointest Oncol*, *WJGO*, online ISSN 1948-5204, DOI: 10.4251) is a peer-reviewed open access academic journal that aims to guide clinical practice and improve diagnostic and therapeutic skills of clinicians.

WJGO covers topics concerning carcinogenesis, tumorigenesis, metastasis, diagnosis, prevention, prognosis, clinical manifestations, nutritional support, molecular mechanisms, and therapy of benign and malignant tumors of the digestive tract. The current columns of *WJGO* include editorial, frontier, diagnostic advances, therapeutics advances, field of vision, mini-reviews, review, topic highlight, medical ethics, original articles, case report, clinical case conference (Clinicopathological conference), and autobiography. Priority publication will be given to articles concerning diagnosis and treatment of gastrointestinal oncology diseases. The following aspects are covered: Clinical diagnosis, laboratory diagnosis, differential diagnosis, imaging tests, pathological diagnosis, molecular biological diagnosis, immunological diagnosis, genetic diagnosis, functional diagnostics, and physical diagnosis; and comprehensive therapy, drug therapy, surgical therapy, interventional treatment, minimally invasive therapy, and robot-assisted therapy.

We encourage authors to submit their manuscripts to *WJGO*. We will give priority to manuscripts that are supported by major national and international foundations and those that are of great clinical significance.

INDEXING/ABSTRACTING

World Journal of Gastrointestinal Oncology (*WJGO*) is now indexed in Science Citation Index Expanded (also known as SciSearch®), PubMed, and PubMed Central. The 2018 edition of Journal Citation Reports® cites the 2017 impact factor for WJGO as 3.140 (5-year impact factor: 3.228), ranking WJGO as 39 among 80 journals in gastroenterology and hepatology (quartile in category Q2), and 114 among 222 journals in oncology (quartile in category Q3).

NAME OF JOURNAL
World Journal of Gastrointestinal Oncology

ISSN
ISSN 1948-5204 (online)

LAUNCH DATE
February 15, 2009

FREQUENCY
Monthly

EDITORIAL BOARD MEMBERS
All editorial board members resources online at http://www.wjgnet.com/1948-5204/editorialboard.htm

EDITORIAL OFFICE
Jin-Lei Wang, Director
World Journal of Gastrointestinal Oncology
Baishideng Publishing Group Inc

7901 Stoneridge Drive, Suite 501, Pleasanton, CA 94588, USA
Telephone: +1-925-2238242
Fax: +1-925-2238243
E-mail: editorialoffice@wjgnet.com
Help Desk: http://www.f6publishing.com/helpdesk
http://www.wjgnet.com

PUBLISHER
Baishideng Publishing Group Inc
7901 Stoneridge Drive,
Suite 501, Pleasanton, CA 94588, USA
Telephone: +1-925-2238242
Fax: +1-925-2238243
E-mail: bpgoffice@wjgnet.com
Help Desk: http://www.f6publishing.com/helpdesk
http://www.wjgnet.com

PUBLICATION DATE
October 15, 2018

INSTRUCTIONS TO AUTHORS
http://www.wjgnet.com/bpg/gerinfo/204

ONLINE SUBMISSION
http://www.f6publishing.com

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 282-289

DOI: 10.4251/wjgo.v10.i10.282

ISSN 1948-5204 (online)

EDITORIAL

Optimizing outcomes for patients with gastric cancer peritoneal carcinomatosis

Jennifer L Leiting, Travis E Grotz

Jennifer L Leiting, Travis E Grotz, Division of Hepatobiliary and Pancreas Surgery, Mayo Clinic, Rochester, MN 55905, United States

ORCID number: Jennifer L Leiting (0000-0002-5784-7937); Travis E Grotz (0000-0002-7753-097X).

Author contributions: Leiting JL and Grotz TE conceived and drafted the manuscript. Both authors approved the final version of the article.

Conflict-of-interest statement: The authors have no conflict of interest to declare.

Manuscript source: Invited manuscript

Correspondence to: Travis E Grotz, MD, Surgical Oncologist, Assistant Professor, Division of Hepatobiliary and Pancreas Surgery, Mayo Clinic, 200 First St Southwest, Rochester, MN 55905, United States. grotz.travis@mayo.edu
Telephone: +1-507-2841529
Fax: +1-507-2845196

Received: July 19, 2018
Peer-review started: July 19, 2018
First decision: August 2, 2018
Revised: August 7, 2018
Accepted: August 12, 2018
Article in press: August 13, 2018
Published online: October 15, 2018

Abstract

Peritoneal carcinomatosis (PC) from gastric cancer has traditionally been considered a terminal progression of the disease and is associated with poor survival outcomes. Positive peritoneal cytology similarly worsens the survival of patients with gastric cancer and treatment options for these patients have been limited. Recent advances in multimodality treatment regimens have led to innovative ways to care for and treat patients with this disease burden. One of these advances has been to use neoadjuvant therapy to try and convert patients with positive cytology or low-volume PC to negative cytology with no evidence of active peritoneal disease. These strategies include the use of neoadjuvant systemic chemotherapy alone, using neoadjuvant laparoscopic heated intraperitoneal chemotherapy (NLHIPEC) after systemic chemotherapy, or using neoadjuvant intraperitoneal and systemic chemotherapy (NIPS) in a bidirectional manner. For patients with higher volume PC, cytoreductive surgery (CRS) and hyperthermic intraperitoneal chemotherapy (HIPEC) have been mainstays of treatment. When used together, CRS and HIPEC can improve overall outcomes in properly selected patients, but overall survival outcomes remain unacceptably low. The extent of peritoneal disease, commonly measured by the peritoneal carcinomatosis index (PCI), and the completeness of cytoreduction, has been shown to greatly impact outcomes in patients undergoing CRS and HIPEC. The uses of NLHIPEC and NLHIPEC plus NIPS have both been shown to decrease the PCI and thus increase the opportunity for complete cytoreduction. Newer therapies like pressurized intraperitoneal aerosol chemotherapy and immunotherapy, such as catumaxomab, along with improved systemic chemotherapeutic regimens, are being explored with great interest. There is exciting progress being made in the management of PC from gastric cancer and its' treatment is no longer futile.

Key words: Peritoneal carcinomatosis index; Peritoneal carcinomatosis; Gastric cancer; Cytoreductive surgery; Heated intraperitoneal chemotherapy; Neoadjuvant intraperitoneal and systemic chemotherapy

Core tip: Peritoneal carcinomatosis (PC) from gastric cancer, along with positive peritoneal cytology, are associated with poor overall outcomes. The treatment of patients with this disease burden has greatly improved and new multimodality treatment regimens have been introduced. Some of these include neoadjuvant laparoscopic heated intraperitoneal chemotherapy and bidirectional therapies like neoadjuvant intraperitoneal and systemic therapy. Appropriate patient selection remains crucial for optimal outcomes but we can be optimistic about the prospects for carefully selected patients with PC from gastric cancer.

Leiting JL, Grotz TE. Optimizing outcomes for patients with gastric cancer peritoneal carcinomatosis. *World J Gastrointest Oncol* 2018; 10(10): 282-289 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/282.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.282

INTRODUCTION

Gastric cancer, more than any other malignancy, has a particular predilection for peritoneal dissemination. The incidence of peritoneal carcinomatosis (PC) at diagnosis ranges anywhere from 5%-30% depending on the staging modality used[1,2]. Furthermore, PC is the most common form of relapse after undergoing curative resection as 30% of all recurrences are in the peritoneum and up to 60% of patients have PC at their time of death[3,4]. Imaging is inadequate with computed tomography (CT) scans having a sensitivity of only 33% and specificity of 99% for detecting PC and 2-[18F]-Fluoro-2-Deoxy-D-Glucose ([18F]FDG) and positron emission tomography (PET) scans having a senstivity of 28% and specificity of 97%[5]. Therefore, diagnostic laparoscopy and peritoneal cytology is indicated for clinical stage T1b or higher gastric cancer as a vital step to detect radiologically occult PC in nearly 40% of patients[6,7]. The presence of microscopic cancer cells within the peritoneal cavity can be identified in up to 6% of patients with no other evidence of metastatic disease[8]. Patients without visible peritoneal metastases but with positive cytology are considered to have stage M1 disease according to the most recent American Joint Committee on Cancer (AJCC) staging as the outcomes are more similar to patients with gross peritoneal metastasis than those with local disease only[9-11].

PC from gastric cancer has generally been considered a terminal progression of disease and has worse outcomes than PC from other malignancies such as ovarian cancer or appendiceal cancer[9,10,12]. Survival for patients with PC is limited but varies based on the burden of disease. A recent series from MD Anderson of patients treated with modern systemic chemotherapy reported 1 year survivals of 24%, 57% and 84% for patients with radiographic PC, PC identified on diagnostic laparoscopy only and positive cytology only, respectively[13]. A similar report from Memorial Sloan-Kettering confirmed a poor overall survival (OS) for patients with gastric cancer and peritoneal cytology with a median OS of 1.3 years compared to 0.8 years for patients with radiographic evidence of peritoneal disease[7].

PERITONEAL CYTOLOGY

The management of patients with positive peritoneal cytology is an evolving field. The role for gastrectomy in patients with limited primary disease and positive cytology without any other peritoneal disease has been debated. Some small studies have shown a survival benefit with a gastrectomy in this subset of patients[14,15]. However, gastrectomy in the setting of untreated positive peritoneal cytology invariably leads to recurrence. National Comprehensive Cancer Network (NCCN) guidelines recommend peritoneal cytology be managed similar to other patients with metastatic gastric cancer with systemic chemotherapy and no surgery[16].

The need to overcome this seemingly small volume and yet unfavorable disease burden has led investigators to seek ways to convert patients with positive cytology to negative cytology so they can proceed to a curative intent gastrectomy (Table 1). The use of neoadjuvant chemotherapy is one of these methods. Aizawa *et al*[17] found that 23 of 47 patients (48.9%) with positive cytology converted to negative cytology after neoadjuvant systemic chemotherapy. R0 resections were able to be performed on all patients. The patients who had a conversion to negative cytology and underwent salvage gastrectomy had a survival benefit of 30.4 mo *vs* 15.0 mo (P = 0.03) when compared to those who had persistently positive cytology treated with gastrectomy[17]. Similarly, a study from Memorial Sloan-Kettering demonstrated that 21 of 48 (44%) patients with initially positive peritoneal cytology treated with systemic chemotherapy achieved negative cytology on repeat laparoscopy[7]. Unfortunately, the Aizawa *et al*[17] study reported that 19% of patients progressed on systemic chemotherapy and the MSKCC study reported that 56% had disease progression while receiving systemic chemotherapy. Therefore, better induction treatments are needed[7,17].

One potential induction treatment is neoadjuvant laparoscopic hyperthermic intraperitoneal chemotherapy (NLHIPEC). In a small phase 2 study, Badgwell *et al*[18] found that 7 of 19 patients (36.8%) with positive peritoneal cytology or low volume PC had resolution in their peritoneal disease and 5 were able to proceed to gastrectomy. Of note, all patients had undergone systemic chemotherapy before being enrolled in the study. Median OS from the time of diagnosis for the entire cohort was 30.2 mo and median OS for the patients who

Table 1 Studies with positive cytology or low volume peritoneal carcinomatosis

Ref.	Patient No.	Treatment group(s)	Intraperitoneal regimen	Systemic regimen	Outcomes
Aizawa *et al*[17], 2015	47	NA systemic chemo	--	Variable	48.9% converted to negative cytology Negative cytology Positive cytology Median OS: 30.4 mo Median OS: 15.0 mo
Badgwell *et al*[18], 2017	19	NA systemic chemo, then NLHIPEC, then gastrectomy if peritoneal disease cleared	MMC and cisplatin	Variable	36.8% converted to negative cytology or had clearance of PC Entire cohort median OS: 30.2 mo
Fujiwara *et al*[19], 2011	25	NA systemic and IP chemo → gastrectomy if peritoneal disease cleared	MMC and cisplatin	IV docetaxel, 5-fu, cisplatin	56% converted to negative cytology or had clearance of PC Negative Positive Median OS: 27.1 mo Median OS: 9.6 mo
Ishigami *et al*[20], 2009	40	NA systemic and IP chemo	Paclitaxel	IV paclitaxel and oral S-1	Median OS: 22.5 mo

NA: Neoadjuvant; chemo: Chemotherapy; OS: Overall survival; NLHIPEC: Neoadjuvant laparoscopic hyperthermic intraperitoneal chemotherapy; MMC: Mitomycin C; PC: Peritoneal carcinomatosis; IP: Intraperitoneal; IV: Intravenous; 5-FU: 5-fluorouracil.

proceeded to gastrectomy was 29 mo from the time of their resection[18]. This approach utilized systemic chemotherapy first, followed by direct intraperitoneal therapy, with encouraging results. Unfortunately, 63.2% of patients had persistently positive cytology or residual PC and did not go onto salvage gastrectomy.

Neoadjuvant intraperitoneal and systemic chemotherapy (NIPS) is another method that utilizes systemic chemotherapy and intraperitoneal chemotherapy, but performs this at the same time in a bidirectional design. Fujiwara *et al*[19] reported 14 of 25 patients (56%) had resolution of their peritoneal disease with either negative cytology or complete regression of PC. Median OS rate for the group with resolution of peritoneal disease was 27.1 mo *vs* 9.6 mo ($P < 0.0001$) in patients with persistently positive cytology or residual PC[19]. Ishigami *et al*[20] looked at the safety and efficacy of bidirectional treatment for patients with positive cytology or PC. They showed a median OS of 22.5 mo and 1-year survival rates of 78%.

PC

The role of gastrectomy in patients with peritoneal disease was addressed in the REGATTA trial[21]. This phase 3 trial enrolled 175 patients with a single incurable factor and randomized them to systemic chemotherapy alone or gastrectomy plus systemic chemotherapy. PC was the incurable factor in three-quarters of the patients enrolled. The authors reported no survival benefit to patients undergoing gastrectomy in addition to systemic chemotherapy[21]. This confirmed that removing the primary tumor without addressing the metastases is not beneficial to the patient.

Cytoreductive surgery (CRS) and hyperthermic intraperitoneal chemotherapy (HIPEC) attempt to address both the primary and the peritoneal metastases simultaneously (Table 2). This aggressive approach has been investigated for gastric cancer since the late 1980's[22-24]. It includes resection of all visible tumor from the peritoneal cavity, followed by the instillation of HIPEC[22]. For the past 30 years, CRS combined with HIPEC has remained the only potentially curative treatment for this advanced stage of gastric cancer[25,26]. A recent meta-analysis that included 11 randomized controlled trials and 21 high quality prospective studies demonstrated an increased median survival of 4 mo in patients with gastric cancer PC treated with HIPEC[27], however, the HIPEC group did experience a higher risk of severe complications. Similarly, CRS and HIPEC have shown a significant improvement in survival for patients with PC from other primaries like appendiceal and ovarian cancer[28,29].

Furthermore, the recent CYTO-CHIP study investigated whether CRS alone was beneficial compared to CRS with HIPEC[30]. They found a significantly improved OS in the CRS with HIPEC group (18.8 mo *vs* 12.1 mo), suggesting that it is the combination of CRS and HIPEC that improves survival[30]. Yang *et al*[31] reported similar results with improved survival for CRS and HIPEC when compared to CRS alone. Median OS for patients undergoing CRS and HIPEC was 11.0 mo compared to 6.5 mo ($P = 0.046$) for CRS alone. Lastly, in a large retrospective study, Glehen *et al*[32] reported a 9.2 mo median OS for 159 patients undergoing CRS with HIPEC or EPIC, with improvement to 15 mo if the cytoreduction was complete.

The benefit of CRS and HIPEC over systemic chemotherapy alone was shown by Rudloff *et al*[33]. In a small cohort of 16 patients, those that underwent CRS, HIPEC, and systemic chemotherapy had an overall median survival rate of 11.3 mo compared to 4.3 mo in the systemic chemotherapy alone group[33].

Unfortunately, although these studies all demonstrated a modest benefit to CRS and HIPEC, OSs remain unacceptably low. It appears that not all patients benefit from CRS and HIPEC and that appropriate patient selection is vital in to order to optimize outcomes. The two most commonly found prognostic factors for survival are consistently the extent of disease, most commonly

Table 2 Studies for peritoneal carcinomatosis with cytoreductive surgery

Ref.	Patient No.	Treatment group(s)	Intraperitoneal regimen	Systemic regimen	Outcomes	
Bonnot *et al*[30], 2018	277	CRS alone *vs* CRS + HIPEC	[1]	[1]	CRS Alone Median OS: 12.1 mo	CRS + HIPEC Median OS: 12.1 mo
Yang *et al*[31], 2011	68	CRS alone *vs* CRS + HIPEC	Cisplatin and MMC	-	CRS Alone Median OS: 6.5 mo	CRS + HIPEC Median OS: 11.0 mo
Glehen *et al*[32], 2010	159	CRS with PIC (HIPEC or EPIC)	Variable	-	Median OS: 9.2 mo	
Rudloff *et al*[33], 2014	16	CRS/HIPEC/SC *vs* SC alone	Oxaliplatin	FOLFOXIRI	SC Alone 4.3 mo	CRS/HIPEC/SC Median OS: 11.3 mo
Canbay *et al*[34], 2014	194	NA systemic and IP chemo, then CRS and HIPEC if responsive	Docetaxel and cisplatin	Oral S-1	78.3% had negative cytology and underwent CRS and HIPEC No response (no CRS or HIPEC) Median OS: 7.5 mo	Response (CRS with HIPEC) Median OS: 15.8 mo
Yonemura *et al*[38], 2017	105	NLHIPEC → CRS or NLHIPEC → NIPS → CRS	Docetaxel and cisplatin	Oral S-1, IV docetaxel and cisplatin	NLHIPEC + CRS Median OS: 14.1 mo PCI: 14.2 → 11.8	NLHIPEC + NIPS + CRS Median OS: 19.2 mo PCI: 14.8 → 9.9

[1]Abstract only, agents used not included. CRS: Cytoreductive surgery; HIPEC: Hyperthermic intraperitoneal chemotherapy; OS: Overall survival; MMC: Mitomycin C; PIC: Perioperative chemotherapy; EPIC: Early postoperative intraperitoneal chemotherapy; SC: Systemic chemotherapy; NA: Neoadjuvant; IP: Intraperitoneal; NLHIPEC: Neoadjuvant laparoscopic HIPEC; NIPS: Neoadjuvant intraperitoneal and systemic chemotherapy; PCI: Peritoneal carcinomatosis index.

measured by the peritoneal carcinomatosis index (PCI), and the completeness of cytoreduction. Glehen *et al*[32] showed that the PCI was the only independent prognostic factor in patients with a complete cytoreduction. No patient survived more than 3 years if their PCI was > 12[32]. A meta-analysis confirmed this with no patients being alive after 3 years if their PCI was > 12[4]. A lower threshold of PCI $\leqslant$ 6 was an independent prognostic factor for patients undergoing CRS and HIPEC after bidirectional chemotherapy (HR 2.16, 95%CI: 1.17-3.98, P = 0.013) in a recent Japanese study[34]. Similarly, Chia *et al*[35] found that a PCI of <7 was a significant predictor of survival. Those with PCI < 7 had a median OS of 26.4 mo compared to 10.9 mo in those who had a PCI $\geqslant$ 7 (HR 2.67, 95%CI: 1.54-4.64, P < 0.001). All the patients who were considered cured as defined by being disease-free at 5 years had a PCI < 7. This same PCI cut-off was seen in a study by Yonemura *et al*[36] who found that a PCI < 7 was associated with improved survival (median survival 2.8 years *vs* 1.1 years, P = 0.0001).

With a lower volume of disease, there is a higher probabilty of being able to completely remove all the metastatic disease. This is the only population that can be expected to have a chance at long-term survival. A meta-analysis showed that cytoreductive scores of 0 or 1 significantly improved survivals in patients with gastric PC[4]. Glehen *et al*[37] showed that patients undergoing a complete cytoreduction with a CC score of 0 or 1 achieved a median OS of 21.3 mo compared to only 6 mo for those with an incomplete cytoreduction. The 5-year OS was 29.4% for those who attained a complete cytoreduction with no survivors in the incomplete cytoreduction group[37]. Canbay *et al*[34] used bidirectional therapy (neoadjuvant systemic and intraperitoneal therapy) to reduce the volume of disease before CRS and HIPEC for patients that responded to treatment. They found better OS in patients who responded to the neoadjuvant treatment and were able to undergo CRS and HIPEC (15.8 mo *vs* 7.5 mo)[34].

There is substantial interest in novel and innovative ways to reduce the PCI prior to cytoreduction. This is crucial because PCI is a determinant in achieving a complete cytoreduction and only patients with a low volume of disease who undergo a complete cytoreduction have a long-term survival benefit from the procedure. Yonemura *et al*[38] used NLHIPEC and NLHIPEC plus NIPS to try and reduce PCI levels before CRS. They found that while NLHIPEC alone reduced PCI levels (14.2 ± 10.7 to 11.8 ± 11.0, P = 0.023), NLHIPC plus NIPS doubled the PCI reduction (14.8 ± 11.4 to 9.9 ± 11.3, P < 0.0001). This may provide more patients with the opportunity for a complete cytoreduction when this would have otherwise not been possible due to a high PCI.

UNRESECTABLE PC

Even with all the advances in therapy for patients with PC from gastric cancer, there are still a large number of patients who are not eligible for these therapies given their high tumor burden or conditional status. Palliative treatment for these patients includes chemotherapy, chemoradiation, or supportive care. None of these regimens treat the peritoneal disease burden and patients generally have very limited survivals.

A new experimental therapy that has emerged to treat these patients is pressurized intraperitoneal aerosol chemotherapy, or PIPAC[39]. This method delivers aerosolized chemotherapy to the peritoneum. The benefit of this method is that the pressure allows

Table 3 Immunotherapy studies

Ref.	Patient No.	Treatment group(s)	Intraperitoneal regimen	Systemic regimen	Outcomes	
Heiss *et al*[45], 2010	66	Paracentesis + catumaxomab *vs* Paracentesis alone	Catumaxomab	-	Paracentesis Alone Median OS: 44 d	Paracentesis + Catumaxomab Median OS: 71 d
Bokemeyer *et al*[46], 2015	54	NA chemotherapy, surgery, intra- and post-op catumaxomab	Catumaxomab	Variable	4 yr DFS: 38% 4 yr OS: 50%	

OS: Overall survival; NA: Neoadjuvant; DFS: Disease free survival.

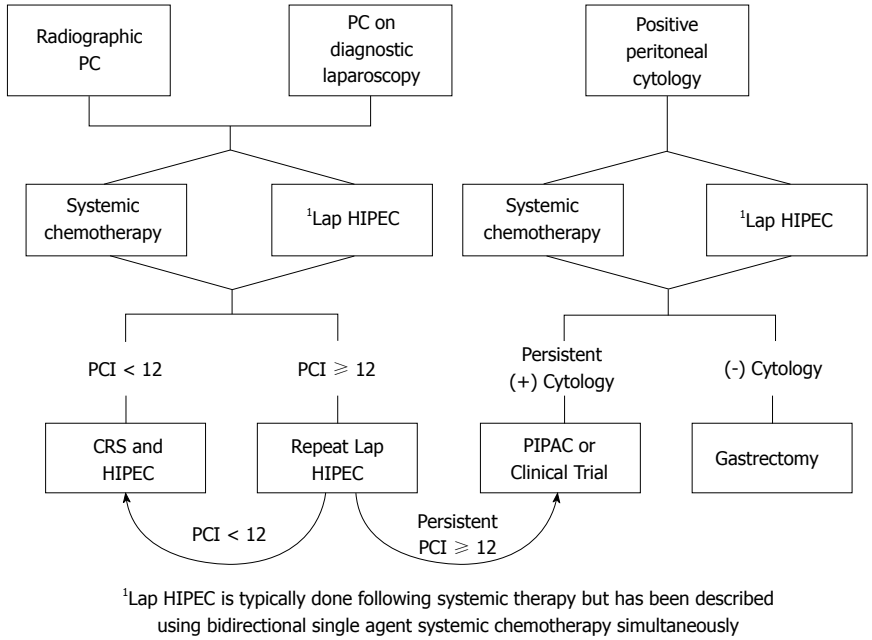


Figure 1 Treatment algorithm for gastric cancer peritoneal carcinomatosis. PC: Peritoneal carcinomatosis; CRS: Cytoreductive surgery; HIPEC: Hyperthermic intraperitoneal chemotherapy; PCI: Peritoneal carcinomatosis index.

for greater lesion penetration as well as allowing for diffuse and even coverage throughout the abdomen[40]. This deeper penetration is likely more critical in these patients with advanced bulky peritoneal metastases. Nadiradze *et al*[39] recently published data on 24 patients with end stage gastric cancer with PC. These patients underwent 1 or more rounds of PIPAC with doxorubicin and cisplatin. The median OS for these patients was 15.4 mo with 52% alive at one year[39]. A multi-center study of PIPAC for advanced PC from a variety of histologies including gastric cancer demonstrated that 63.5% of patients achieved resolution of symptoms[41]. This therapy may prove to be beneficial for more than just end stage gastric cancer patients but additional research is needed.

FUTURE EFFORTS

Innovative discoveries and continued efforts to optimize treatment for patients with PC from gastric cancer are needed. This includes improved systemic chemotherapy options such as FLOT, which has been demonstrated to be effective in patients with limited metastatic disease[42]. The AIO-FLOT3 trial reported a median OS of 31.3 mo and a 60% radiographic response rate for patients who were treated with perioperative FLOT systemic chemotherapy and surgical resection of all metastatic disease[42].

Another innovative approach is the use of immunotherapy, like catumaxomab, as an intraperitoneal treatment (Table 3). Catumaxomab is an antibody that binds to both epithelial cells through epithelial cell adhesion molecule (EpCAM) and T-cells through CD3[43]. Gastric cancer expresses high levels of EpCAM so the intraperitoneal administration of EpCAM provides targeted therapy to peritoneal implants[44]. In patients with malignant ascites from PC of gastric origin, it was found to significantly prolong OS from 44 to 71 d[45]. Bokemeyer *et al*[46] conducted a phase 2 study where patients underwent intra- and post-operative intraperitoneal catumaxomab administration after undergoing neoadjuvant chemotherapy and resection. These patients had four-year disease-free survival rates of 38% and four-year OS rates as high as 50%. Though catumaxomab is no lon-

ger available, the use of intraperitoneal immunotherapy remains promising and is under continued investigation[47].

There remain many areas related to the management of PC from gastric cancer that can be improved. Better detection of early occult peritoneal metastases would allow the clinician to select more appropriate patients for these multidisciplinary treatments. This may be in the form of improved imaging modalities like fluorescence and antibody-labelled imaging[48] or the use of RT-PCR with cytology to improve the sensitivity of detecting cancer cells in peritoneal washings[49]. The optimal chemotherapeutic agent, or agents, to use is unclear, both systemically and in the peritoneal cavity. Many of the studies discussed here used different treatment regimens with some varying even within the same study, so it is difficult to compare outcomes from one study to the next. Also, the ideal sequence, route, and duration of treatment for these patients that will deliver the greatest long-term benefit with manageable side-effects is unknown, though there are many promising options.

Appropriate patient selection remains crucial for optimal outcomes in patients with gastric cancer, but patients with PC or positive cytology should no longer be immediately excluded from potentially curative multimodality treatment regimens. There are treatment options that can be offered to suitable patients with PC from gastric cancer that have the possibility of extended survival (Figure 1). We are finally seeing progress in the management of a disease that has traditionally been thought of as terminal and it is time to change our approach. We are not yet at a point where we can offer these patients a cure, but the treatment of PC from gastric cancer is no longer a futile endeavor and can be approached with careful optimism.

REFERENCES

1 **Yonemura Y**, Canbay E, Li Y, Coccolini F, Glehen O, Sugarbaker PH, Morris D, Moran B, Gonzaletz-Moreno S, Deraco M, Piso P, Elias D, Batlett D, Ishibashi H, Mizumoto A, Verwaal V, Mahtem H. A comprehensive treatment for peritoneal metastases from gastric cancer with curative intent. *Eur J Surg Oncol* 2016; **42**: 1123-1131 [PMID: 27160355 DOI: 10.1016/j.ejso.2016.03.016]

2 **Goéré D**, Gras-Chaput N, Aupérin A, Flament C, Mariette C, Glehen O, Zitvogel L, Elias D. Treatment of gastric peritoneal carcinomatosis by combining complete surgical resection of lesions and intraperitoneal immunotherapy using catumaxomab. *BMC Cancer* 2014; **14**: 148 [PMID: 24589307 DOI: 10.1186/1471-2407-14-148]

3 **D'Angelica M**, Gonen M, Brennan MF, Turnbull AD, Bains M, Karpeh MS. Patterns of initial recurrence in completely resected gastric adenocarcinoma. *Ann Surg* 2004; **240**: 808-816 [PMID: 15492562 DOI: 10.1097/01.sla.0000143245.28656.15]

4 **Coccolini F**, Catena F, Glehen O, Yonemura Y, Sugarbaker PH, Piso P, Montori G, Ansaloni L. Complete versus incomplete cytoreduction in peritoneal carcinosis from gastric cancer, with consideration to PCI cut-off. Systematic review and meta-analysis. *Eur J Surg Oncol* 2015; **41**: 911-919 [PMID: 25936764 DOI: 10.1016/j.ejso.2015.03.231]

5 **Wang Z**, Chen JQ. Imaging in assessing hepatic and peritoneal metastases of gastric cancer: a systematic review. *BMC Gastroenterol* 2011; **11**: 19 [PMID: 21385469 DOI: 10.1186/1471-230X-11-19]

6 **Burbidge S**, Mahady K, Naik K. The role of CT and staging laparoscopy in the staging of gastric cancer. *Clin Radiol* 2013; **68**: 251-255 [PMID: 22985749 DOI: 10.1016/j.crad.2012.07.015]

7 **Mezhir JJ**, Shah MA, Jacks LM, Brennan MF, Coit DG, Strong VE. Positive peritoneal cytology in patients with gastric cancer: natural history and outcome of 291 patients. *Ann Surg Oncol* 2010; **17**: 3173-3180 [PMID: 20585870 DOI: 10.1245/s10434-010-1183-0]

8 **Bentrem D**, Wilton A, Mazumdar M, Brennan M, Coit D. The value of peritoneal cytology as a preoperative predictor in patients with gastric carcinoma undergoing a curative resection. *Ann Surg Oncol* 2005; **12**: 347-353 [PMID: 15915368 DOI: 10.1245/ASO.2005.03.065]

9 **National Comprehensive Cancer Network**. Gastric Cancer. Version 2. 2018. Available from: URL: https://www.nccn.org/store/login/login.aspx?ReturnURL=https://www.nccn.org/professionals/physician_gls/pdf/gastric.pdf

10 **Isobe Y**, Nashimoto A, Akazawa K, Oda I, Hayashi K, Miyashiro I, Katai H, Tsujitani S, Kodera Y, Seto Y, Kaminishi M. Gastric cancer treatment in Japan: 2008 annual report of the JGCA nationwide registry. *Gastric Cancer* 2011; **14**: 301-316 [PMID: 21894577 DOI: 10.1007/s10120-011-0085-6]

11 **Leake PA**, Cardoso R, Seevaratnam R, Lourenco L, Helyer L, Mahar A, Rowsell C, Coburn NG. A systematic review of the accuracy and utility of peritoneal cytology in patients with gastric cancer. *Gastric Cancer* 2012; **15** Suppl 1: S27-S37 [PMID: 21809111 DOI: 10.1007/s10120-011-0071-z]

12 **Glehen O**, Gilly FN, Boutitie F, Bereder JM, Quenet F, Sideris L, Mansvelt B, Lorimier G, Msika S, Elias D; French Surgical Association. Toward curative treatment of peritoneal carcinomatosis from nonovarian origin by cytoreductive surgery combined with perioperative intraperitoneal chemotherapy: a multi-institutional study of 1,290 patients. *Cancer* 2010; **116**: 5608-5618 [PMID: 20737573 DOI: 10.1002/cncr.25356]

13 **Shiozaki H**, Elimova E, Slack RS, Chen HC, Staerkel GA, Sneige N, Shimodaira Y, Sagebiel T, Lee JH, Bhutani MS, Das P, Mansfield PF, Estrella JS, Badgwell BD, Ajani JA. Prognosis of gastric adenocarcinoma patients with various burdens of peritoneal metastases. *J Surg Oncol* 2016; **113**: 29-35 [PMID: 26603684 DOI: 10.1002/jso.24087]

14 **Nakagohri T**, Yoneyama Y, Kinoshita T, Konishi M, Inoue K, Takahashi S. Prognostic significance of peritoneal washing cytology in patients with potentially resectable gastric cancer. *Hepatogastroenterology* 2008; **55**: 1913-1915 [PMID: 19102421 DOI: 10.1002/bjs.7812]

15 **Suzuki O**, Fukuchi M, Mochiki E, Ishiguro T, Sobajima J, Onozawa H, Imaizumi H, Kumagai Y, Baba H, Kumamoto K, Tsuji Y, Ishibashi K, Ishida H. Prognostic role of gastrectomy in patients with gastric cancer with positive peritoneal cytology. *Int Surg* 2014; **99**: 830-834 [PMID: 25437595 DOI: 10.9738/INTSURG-D-14-00119.1]

16 **Pak LM**, Coit DG, Eaton AA, Allen PJ, D'Angelica MI, DeMatteo RP, Jarnagin WR, Strong VE, Kingham TP. Percutaneous Peritoneal Lavage for the Rapid Staging of Gastric and Pancreatic Cancer. *Ann Surg Oncol* 2017; **24**: 1174-1179 [PMID: 28058561 DOI: 10.1245/s10434-016-5757-3]

17 **Aizawa M**, Nashimoto A, Yabusaki H, Nakagawa S, Matsuki A, Homma K, Kawasaki T. The clinical significance of potentially curative resection for gastric cancer following the clearance of free cancer cells in the peritoneal cavity by induction chemotherapy. *Surg Today* 2015; **45**: 611-617 [PMID: 25027056 DOI: 10.1007/s00595-014-0979-0]

18 **Badgwell B**, Blum M, Das P, Estrella J, Wang X, Ho L, Fournier K, Royal R, Mansfield P, Ajani J. Phase II Trial of Laparoscopic Hyperthermic Intraperitoneal Chemoperfusion for Peritoneal Carcinomatosis or Positive Peritoneal Cytology in Patients with Gastric Adenocarcinoma. *Ann Surg Oncol* 2017; **24**: 3338-3344 [PMID: 28799004 DOI: 10.1245/s10434-017-6047-4]

19 **Fujiwara Y**, Takiguchi S, Nakajima K, Miyata H, Yamasaki M, Kurokawa Y, Okada K, Mori M, Doki Y. Neoadjuvant intraperitoneal and systemic chemotherapy for gastric cancer patients with peritoneal dissemination. *Ann Surg Oncol* 2011; **18**: 3726-3731 [PMID: 21584835 DOI: 10.1245/s10434-011-1770-8]

20 **Ishigami H**, Kitayama J, Kaisaki S, Hidemura A, Kato M, Otani K, Kamei T, Soma D, Miyato H, Yamashita H, Nagawa H. Phase II study of weekly intravenous and intraperitoneal paclitaxel combined with S-1 for advanced gastric cancer with peritoneal metastasis. *Ann Oncol* 2010; **21**: 67-70 [PMID: 19605503 DOI: 10.1093/annonc/mdp260]

21 **Fujitani K**, Yang HK, Mizusawa J, Kim YW, Terashima M, Han SU, Iwasaki Y, Hyung WJ, Takagane A, Park DJ, Yoshikawa T, Hahn S, Nakamura K, Park CH, Kurokawa Y, Bang YJ, Park BJ, Sasako M, Tsujinaka T; REGATTA study investigators. Gastrectomy plus chemotherapy versus chemotherapy alone for advanced gastric cancer with a single non-curable factor (REGATTA): a phase 3, randomised controlled trial. *Lancet Oncol* 2016; **17**: 309-318 [PMID: 26822397 DOI: 10.1016/S1470-2045(15)00553-7]

22 **Fujimoto S**, Shrestha RD, Kokubun M, Ohta M, Takahashi M, Kobayashi K, Kiuchi S, Okui K, Miyoshi T, Arimizu N. Intraperitoneal hyperthermic perfusion combined with surgery effective for gastric cancer patients with peritoneal seeding. *Ann Surg* 1988; **208**: 36-41 [PMID: 3133994 DOI: 10.1097/00000658-198807000-00005]

23 **Koga S**, Hamazoe R, Maeta M, Shimizu N, Murakami A, Wakatsuki T. Prophylactic therapy for peritoneal recurrence of gastric cancer by continuous hyperthermic peritoneal perfusion with mitomycin C. *Cancer* 1988; **61**: 232-237 [PMID: 3121165 DOI: 10.1002/1097-0142(19880115)61:2<232::AID-CNCR2820610205>3.0.CO;2-U]

24 **Fujimoto S**, Shrestha RD, Kokubun M, Kobayashi K, Kiuchi S, Konno C, Ohta M, Takahashi M, Kitsukawa Y, Mizutani M. Positive results of combined therapy of surgery and intraperitoneal hyperthermic perfusion for far-advanced gastric cancer. *Ann Surg* 1990; **212**: 592-596 [PMID: 2241314 DOI: 10.1097/00000658-199011000-00005]

25 **Glehen O**, Gilly FN, Cotte E. Hyperthermic intraperitoneal chemotherapy in advanced gastric cancer: the end of skepticism? *Ann Surg Oncol* 2011; **18**: 1524-1526 [PMID: 21384246 DOI: 10.1245/s10434-011-1632-4]

26 **Yonemura Y**, Kawamura T, Bandou E, Takahashi S, Sawa T, Matsuki N. Treatment of peritoneal dissemination from gastric cancer by peritonectomy and chemohyperthermic peritoneal perfusion. *Br J Surg* 2005; **92**: 370-375 [PMID: 15739249 DOI: 10.1002/bjs.4695]

27 **Desiderio J**, Chao J, Melstrom L, Warner S, Tozzi F, Fong Y, Parisi A, Woo Y. The 30-year experience-A meta-analysis of randomised and high-quality non-randomised studies of hyperthermic intraperitoneal chemotherapy in the treatment of gastric cancer. *Eur J Cancer* 2017; **79**: 1-14 [PMID: 28456089 DOI: 10.1016/j.ejca.2017.03.030]

28 **Verwaal VJ**, van Ruth S, de Bree E, van Sloothen GW, van Tinteren H, Boot H, Zoetmulder FA. Randomized trial of cytoreduction and hyperthermic intraperitoneal chemotherapy versus systemic chemotherapy and palliative surgery in patients with peritoneal carcinomatosis of colorectal cancer. *J Clin Oncol* 2003; **21**: 3737-3743 [PMID: 14551293 DOI: 10.1200/JCO.2003.04.187]

29 **Shaib WL**, Martin LK, Choi M, Chen Z, Krishna K, Kim S, Brutcher E, Staley C 3rd, Maithel SK, Philip P, Abdel-Misih S, Bekaii-Saab TS, El-Rayes BF. Hyperthermic Intraperitoneal Chemotherapy Following Cytoreductive Surgery Improves Outcome in Patients With Primary Appendiceal Mucinous Adenocarcinoma: A Pooled Analysis From Three Tertiary Care Centers. *Oncologist* 2015; **20**: 907-914 [PMID: 26070916 DOI: 10.1634/theoncologist.2014-0294]

30 **Bonnot PE**, Piessen G, Pocard M, Meunier B, Bereder JM, Abboud K, Marchal F, Quenet F, Goere D, Msika S, Arvieux C, Pirro N, Wernert R, RAT P, Pezet D, Lefevre J, Courvoisier T, Kianmanesh R, Meeus P, Glehen O. CYTO-CHIP: Cytoreductive surgery versus cytoreductive surgery and hyperthermic intraperitoneal chemotherapy for gastric cancer with peritoneal metastasis: A propensity-score analysis from BIG RENAPE and FREGAT working groups. *J Clin Oncol* 2018; **36**: 8

31 **Yang XJ**, Huang CQ, Suo T, Mei LJ, Yang GL, Cheng FL, Zhou YF, Xiong B, Yonemura Y, Li Y. Cytoreductive surgery and hyperthermic intraperitoneal chemotherapy improves survival of patients with peritoneal carcinomatosis from gastric cancer: final results of a phase III randomized clinical trial. *Ann Surg Oncol* 2011; **18**: 1575-1581 [PMID: 21431408 DOI: 10.1245/s10434-011-1631-5]

32 **Glehen O**, Gilly FN, Arvieux C, Cotte E, Boutitie F, Mansvelt B, Bereder JM, Lorimier G, Quenet F, Elias D; Association Française de Chirurgie. Peritoneal carcinomatosis from gastric cancer: a multi-institutional study of 159 patients treated by cytoreductive surgery combined with perioperative intraperitoneal chemotherapy. *Ann Surg Oncol* 2010; **17**: 2370-2377 [PMID: 20336386 DOI: 10.1245/s10434-010-1039-7]

33 **Rudloff U**, Langan RC, Mullinax JE, Beane JD, Steinberg SM, Beresnev T, Webb CC, Walker M, Toomey MA, Schrump D, Pandalai P, Stojadinovic A, Avital I. Impact of maximal cytoreductive surgery plus regional heated intraperitoneal chemotherapy (HIPEC) on outcome of patients with peritoneal carcinomatosis of gastric origin: results of the GYMSSA trial. *J Surg Oncol* 2014; **110**: 275-284 [PMID: 25042700 DOI: 10.1002/jso.23633]

34 **Canbay E**, Mizumoto A, Ichinose M, Ishibashi H, Sako S, Hirano M, Takao N, Yonemura Y. Outcome data of patients with peritoneal carcinomatosis from gastric origin treated by a strategy of bidirectional chemotherapy prior to cytoreductive surgery and hyperthermic intraperitoneal chemotherapy in a single specialized center in Japan. *Ann Surg Oncol* 2014; **21**: 1147-1152 [PMID: 24356799 DOI: 10.1245/s10434-013-3443-2]

35 **Chia CS**, You B, Decullier E, Vaudoyer D, Lorimier G, Abboud K, Bereder JM, Arvieux C, Boschetti G, Glehen O; BIG RENAPE Group. Patients with Peritoneal Carcinomatosis from Gastric Cancer Treated with Cytoreductive Surgery and Hyperthermic Intraperitoneal Chemotherapy: Is Cure a Possibility? *Ann Surg Oncol* 2016; **23**: 1971-1979 [PMID: 26753751 DOI: 10.1245/s10434-015-5081-3]

36 **Yonemura Y**, Elnemr A, Endou Y, Hirano M, Mizumoto A, Takao N, Ichinose M, Miura M, Li Y. Multidisciplinary therapy for treatment of patients with peritoneal carcinomatosis from gastric cancer. *World J Gastrointest Oncol* 2010; **2**: 85-97 [PMID: 21160926 DOI: 10.4251/wjgo.v2.i2.85]

37 **Glehen O**, Schreiber V, Cotte E, Sayag-Beaujard AC, Osinsky D, Freyer G, François Y, Vignal J, Gilly FN. Cytoreductive surgery and intraperitoneal chemohyperthermia for peritoneal carcinomatosis arising from gastric cancer. *Arch Surg* 2004; **139**: 20-26 [PMID: 14718269 DOI: 10.1001/archsurg.139.1.20]

38 **Yonemura Y**, Ishibashi H, Hirano M, Mizumoto A, Takeshita K, Noguchi K, Takao N, Ichinose M, Liu Y, Li Y. Effects of Neoadjuvant Laparoscopic Hyperthermic Intraperitoneal Chemotherapy and Neoadjuvant Intraperitoneal/Systemic Chemotherapy on Peritoneal Metastases from Gastric Cancer. *Ann Surg Oncol* 2017; **24**: 478-485 [PMID: 27506661 DOI: 10.1245/s10434-016-5487-6]

39 **Nadiradze G**, Giger-Pabst U, Zieren J, Strumberg D, Solass W, Reymond MA. Pressurized Intraperitoneal Aerosol Chemotherapy (PIPAC) with Low-Dose Cisplatin and Doxorubicin in Gastric Peritoneal Metastasis. *J Gastrointest Surg* 2016; **20**: 367-373 [PMID: 26511950 DOI: 10.1007/s11605-015-2995-9]

40 **Solass W**, Kerb R, Mürdter T, Giger-Pabst U, Strumberg D, Tempfer C, Zieren J, Schwab M, Reymond MA. Intraperitoneal chemotherapy of peritoneal carcinomatosis using pressurized aerosol as an alternative to liquid solution: first evidence for efficacy. *Ann Surg Oncol* 2014; **21**: 553-559 [PMID: 24006094 DOI: 10.1245/s10434-013-3213-1]

41 **Alyami M**, Gagniere J, Sgarbura O, Cabelguenne D, Villeneuve L, Pezet D, Quenet F, Glehen O, Bakrin N, Passot G. Multicentric initial experience with the use of the pressurized intraperitoneal aerosol chemotherapy (PIPAC) in the management of unresectable peritoneal carcinomatosis. *Eur J Surg Oncol* 2017; **43**: 2178-2183 [PMID: 28964609 DOI: 10.1016/j.ejso.2017.09.010]

42 **Al-Batran SE**, Homann N, Pauligk C, Illerhaus G, Martens UM, Stoehlmacher J, Schmalenberg H, Luley KB, Prasnikar N, Egger M, Probst S, Messmann H, Moehler M, Fischbach W, Hartmann JT, Mayer F, Höffkes HG, Koenigsmann M, Arnold D, Kraus TW, Grimm K, Berkhoff S, Post S, Jäger E, Bechstein W, Ronellenfitsch U, Mönig S, Hofheinz RD. Effect of Neoadjuvant Chemotherapy Followed by Surgical Resection on Survival in Patients With Limited Metastatic Gastric or Gastroesophageal Junction Cancer: The AIO-FLOT3 Trial. *JAMA Oncol* 2017; **3**: 1237-1244 [PMID: 28448662 DOI: 10.1001/jamaoncol.2017.0515]

43 **Atanackovic D**, Reinhard H, Meyer S, Spöck S, Grob T, Luetkens T, Yousef S, Cao Y, Hildebrandt Y, Templin J, Bartels K, Lajmi N, Stoiber H, Kröger N, Atz J, Seimetz D, Izbicki JR, Bokemeyer C. The trifunctional antibody catumaxomab amplifies and shapes tumor-specific immunity when applied to gastric cancer patients in the adjuvant setting. *Hum Vaccin Immunother* 2013; **9**: 2533-2542 [PMID: 23955093 DOI: 10.4161/hv.26065]

44 **Warneke VS**, Behrens HM, Haag J, Krüger S, Simon E, Mathiak M, Ebert MP, Röcken C. Members of the EpCAM signalling pathway are expressed in gastric cancer tissue and are correlated with patient prognosis. *Br J Cancer* 2013; **109**: 2217-2227 [PMID: 24008668 DOI: 10.1038/bjc.2013.536]

45 **Heiss MM**, Murawa P, Koralewski P, Kutarska E, Kolesnik OO, Ivanchenko VV, Dudnichenko AS, Aleknaviciene B, Razbadauskas A, Gore M, Ganea-Motan E, Ciuleanu T, Wimberger P, Schmittel A, Schmalfeldt B, Burges A, Bokemeyer C, Lindhofer H, Lahr A, Parsons SL. The trifunctional antibody catumaxomab for the treatment of malignant ascites due to epithelial cancer: Results of a prospective randomized phase II/III trial. *Int J Cancer* 2010; **127**: 2209-2221 [PMID: 20473913 DOI: 10.1002/ijc.25423]

46 **Bokemeyer C**, Stein A, Ridwelski K, Atanackovic D, Arnold D, Wöll E, Ulrich A, Fischer R, Krüger C, Schuhmacher C. A phase II study of catumaxomab administered intra- and postoperatively as part of a multimodal approach in primarily resectable gastric cancer. *Gastric Cancer* 2015; **18**: 833-842 [PMID: 25214034 DOI: 10.1007/s10120-014-0423-6]

47 **Seidl C**, Zöckler C, Beck R, Quintanilla-Martinez L, Bruchertseifer F, Senekowitsch-Schmidtke R. 177Lu-immunotherapy of experimental peritoneal carcinomatosis shows comparable effectiveness to 213Bi-immunotherapy, but causes toxicity not observed with 213Bi. *Eur J Nucl Med Mol Imaging* 2011; **38**: 312-322 [PMID: 21072513 DOI: 10.1007/s00259-010-1639-2]

48 **Ito A**, Ito Y, Matsushima S, Tsuchida D, Ogasawara M, Hasegawa J, Misawa K, Kondo E, Kaneda N, Nakanishi H. New whole-body multimodality imaging of gastric cancer peritoneal metastasis combining fluorescence imaging with ICG-labeled antibody and MRI in mice. *Gastric Cancer* 2014; **17**: 497-507 [PMID: 24288123 DOI: 10.1007/s10120-013-0316-0]

49 **To EM**, Chan WY, Chow C, Ng EK, Chung SC. Gastric cancer cell detection in peritoneal washing: cytology versus RT-PCR for CEA transcripts. *Diagn Mol Pathol* 2003; **12**: 88-95 [PMID: 12766613 DOI: 10.1097/00019606-200306000-00004]

P- Reviewer: Fiorentini G, Jeong KY, Mohamed SY, Saglam S, Shu X **S- Editor**: Ji FF **L- Editor**: A **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 290-292

DOI: 10.4251/wjgo.v10.i10.290

ISSN 1948-5204 (online)

EDITORIAL

Inhibiting focal adhesion kinase: A potential target for enhancing therapeutic efficacy in colorectal cancer therapy

Keun-Yeong Jeong

Keun-Yeong Jeong, Division of Research and Development, Metimedi Pharmaceuticals, Incheon 22006, South Korea

ORCID number: Keun-Yeong Jeong (0000-0002-4933-3493).

Author contributions: Jeong KY conceived the study and drafted the manuscript; this author approved the final version of the article.

Conflict-of-interest statement: This author has no conflict of interest to declare.

Manuscript source: Invited manuscript

Correspondence to: Keun-Yeong Jeong, PhD, SVP, Head of R and D, Division of Research and Development, Metimedi Pharmaceuticals, R and D Division, Metimedi Pharmaceuticals Co., 263, Central-ro, Yeonsu-Gu, Incheon 22006, South Korea. alvirus@naver.com
Telephone: +82-32-2050541
Fax: +82-32-2050542

Received: July 17, 2018
Peer-review started: July 17, 2018
First decision: August 2, 2018
Revised: August 16, 2018
Accepted: August 27, 2018
Article in press: August 28, 2018
Published online: October 15, 2018

Abstract

Focal adhesion kinase (FAK) is a major integrin-dependent tyrosine phosphorylated protein, recently, FAK association with colorectal cancer (CRC) has gained attention. The various cancer-promoting mechanisms that associated with FAK can be implicated in the progression of CRC. The interactions between structural features of FAK and various kinases could be closely related to growth, survival, and metastasis in CRC cells. These interactions include human epithelial growth factor receptor, c-Met, platelet-derived growth factor receptor, vascular endothelial growth factor receptor, and Src. Such interactions can trigger the survival signaling of CRC cells and are also involved signaling downstream of phosphatidylinositol 3-kinase, AKT, and the extracellular regulated kinase. Based on this scientific background, many pharmaceutical companies are taking efforts to develop FAK inhibitors to treat solid cancer including CRC. Although the anti-cancer efficacies have been noted in many studies, the commercial drugs have not been developed yet. Therefore, the FAK research on CRC is expected to gain momentum and be highly appreciated as a potential field for developing the new drugs. Therefore, the studies on FAK that effect on the progression of human CRC s would be possible to suggest various approaches to CRC treatment, and FAK could be a potential target as an anticancer candidate for CRC therapies.

Key words: Colorectal cancer; Focal adhesion kinase; Focal adhesion kinase inhibitor; Anticancer effect

Core tip: Despite ongoing development in treatment for colorectal cancer (CRC), effective markers for treatment of CRC have not been elucidated. FAK association with various kinases for progression and invasion of CRC has recently gained attention. The possibility for this association is accounted that FAK is interactions with integrins, growth factor receptors, and adjacent kinase domain. Targeting FAK is possible to explain the mechanism at

the upstream level by which can mediate the expression of various survival signaling and inhibition of oncosuppressor genes as well as inducing migration and invasion of the CRC cells. Therefore, FAK could be a prognostic marker and a potential candidate target for CRC therapies.

Jeong KY. Inhibiting focal adhesion kinase: A potential target for enhancing therapeutic efficacy in colorectal cancer therapy. *World J Gastrointest Oncol* 2018; 10(10): 290-292 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/290.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.290

Focal adhesion kinase (FAK) is a major integrin-dependent tyrosine phosphorylated protein and a non-receptor tyrosine kinase that is localized to cellular focal adhesions[1]. Although there have been many studies on the role of FAK in breast cancer, its association with colorectal cancer (CRC) has recently gained attention. FAK, known as protein tyrosine kinase 2, is related to other tyrosine kinases, such as Src kinase[2]. FAK comprises a central kinase domain between an N-terminal FERM domain and a C-terminal domain that includes the focal adhesion sequence. The construction of the N-terminal FERM domain is similar to that of cytoskeletal proteins and several tyrosine phosphatases and tyrosine kinases. This domain mediates FAK interactions with integrins and growth factor receptors and interacts with the adjacent kinase domain in FAK. The C-terminal domain contains proline-rich sequences for SH3 domain-containing proteins and acts to recruit additional signaling proteins[3,4].

The interactions between structural features of FAK and various kinases could be closely related to cancer growth, survival, and metastasis. FAK is activated by the direct interaction of the Src kinase with the integrin β cytoplasmic domain[4]. Integrin can trigger the survival signaling of cancer cells at locations further downstream of phosphatidylinositol 3-kinase (PI3K), AKT, and the extracellular regulated kinase (ERK)[1,5]. The kinase complex with Src is reportedly affected in the activation of these survival pathways. In addition, FAK interacts with several receptor tyrosine kinases, including human epithelial growth factor receptor, c-Met, platelet-derived growth factor receptor, and vascular endothelial growth factor receptor (VEGFR), which also mediates the survival pathway of cancer cells[2,6]. The detailed mechanism of PI3K signaling is as follows. The PI3K/AKT pathway induces the expression of apoptosis inhibitory proteins through nuclear factor kappa (NF-κ) B and protects the cells from stress-induced apoptosis. It is also associated with expression of cancer suppressor genes[5,6]. FAK promotes cell survival via suppression of p53 activation. This is mediated by the kinase-independent FAK FERM domain, and it suppresses the transcriptional activation of target genes that is mediated by p53 activation. Therefore, FAK can enhance cell survival through both kinase-dependent and-independent mechanisms[7]. Further, the expression of an active mutant of ERK has indicated a direct role of FAK in promoting cancer growth. It is suggested that FAK signaling through the ERK pathway is needed to maintain cancer cell development[8]. Furthermore, the kinase activity of FAK is estimated to be significant for the invasive phenotype and for cancer metastasis. FAK reportedly promotes cancer cell invasion through the regulation of matrix metalloproteinases (MMPs)[1,9]. In v-Src transformed cells, the Rac1 and JNK is activated in FAK/Src complex and is induced the MMP2 and MMP9 expression. Thus, FAK promotes increased invasiveness of cancer cells[10].

Of course, the various cancer-promoting mechanisms associated with FAK described above could also be implicated in the progression of CRC. Colon cells including epithelial and fibrous cells increases the FAK expression at early stages of carcinogenesis, even before the cancer has formed[1,11]. The up-regulation of FAK promotes the adhesive properties of CRC cells and their survival[11]. FAK signaling is associated with the binding of the Rho guanine nucleotide exchange factor, and this signaling complex promotes the local invasion of colon carcinoma. The increase in FAK activation is thus related to elevated tyrosine phosphorylation and an adaptor protein, such as paxillin, involved in the growth of the CRC cells[1,2,12]. Further, FAK signaling contributes to epithelial-mesenchymal transition (EMT) profile change in CRC cells. FAK scaffolding increases, thus leading to alterations in EMT markers, including MMP-induced motility of CRC cells. Therefore, FAK acts to affect the dynamic internalization of E-cadherin in CRC cells[2,13]. Furthermore, FAK FERM overexpression can reduce steady-state p53 levels in CRC cells, particularly HCT-116 cells. As increased FAK expression is often found in early-stage CRCs, the FAK FERM-mediated cell survival pathway is expected to have an important function in the survival of CRC cells[7,14]. During cancer progression and metastasis, an anchorage-independent pathway can facilitate the spread of cells from the primary cancer site. Under these conditions, the cancer cells that show higher levels of FAK may be more resistant to apoptosis by non-integrin-associated FAK to translocate to the nucleus and prevent excessive p53 activation[2,7,15]. It is associated with that alternative-spliced transcripts encompassing the N-terminal FERM domain without the FAK kinase or C-terminal regions would be related to the progression of CRC[2,7].

Based on this scientific background, many pharmaceutical companies are taking efforts to develop FAK inhibitors. TAE-226 by Novartis exhibits nanomolar inhibitory activity toward FAK and protein tyrosine kinases and has anti-cancer activity. It particularly blocks cell proliferation and invasion and showed increased apoptosis in many xenograft animal models[7]. Further, TAE-226 in combination with docetaxel, a microtubule stabili-

zer, significantly decreases angiogenesis and cancer cell invasion[15]. Pfizer has developed PF-228 that shows more specific FAK inhibitory activity. It inhibits cancer cell migration *in vitro*. Pfizer has also developed PF-573, 228 compound, and the results indicated cancer growth inhibition in the colon xenograft cancer model[16]. In addition, several other FAK inhibitors have been developed, including GSK2256098 by GlaxoSmithKline as a formulation for oral intake and VS-4718 by Poniard as an improved version of the previous product, PND-1186[17,18]. Although efficacy has been noted in non-clinical and early-stage clinical trials, the drugs have not been commercialized yet. Therefore, the FAK research on CRC is expected to gain momentum and be highly appreciated as a potential field for developing the new drugs.

The kinase-dependent function and kinase-independent ability of FAK are essential for cancer development[19]. Multifunctional characteristics of FAK have been highlighted has modulators of numerous signal transductions in CRC cells. The established role of FAK in cancer progression and metastasis has obviously proposed that increase in FAK expression contributes a very important part in CRC development. Various inhibitors by small-molecules for targeting inhibition of FAK kinase and autophosphorylation have been produced by many pharmaceutical companies. Although some clinical trials have already been undergoing and potential efficacy has been noted, further studies must be needed to confirm if FAK expression has important role in a progression of human CRC and elaborates on the clear mechanisms and downstream effectors in the context of carcinogenicity. Taken together, based on the clinical observations, the over-expression of FAK at both transcriptional and translational levels in human CRCs would imply that targeting FAK could be a prognostic marker and a potential anticancer candidate for CRC therapy.

REFERENCES

1 **Yoon H**, Dehart JP, Murphy JM, Lim ST. Understanding the roles of FAK in cancer: inhibitors, genetic models, and new insights. *J Histochem Cytochem* 2015; **63**: 114-128 [PMID: 25380750 DOI: 10.1369/0022155414561498]

2 **Sulzmaier FJ**, Jean C, Schlaepfer DD. FAK in cancer: mechanistic findings and clinical applications. *Nat Rev Cancer* 2014; **14**: 598-610 [PMID: 25098269 DOI: 10.1038/nrc3792]

3 **Lietha D**, Cai X, Ceccarelli DF, Li Y, Schaller MD, Eck MJ. Structural basis for the autoinhibition of focal adhesion kinase. *Cell* 2007; **129**: 1177-1187 [PMID: 17574028 DOI: 10.1016/j.cell.2007.05.041]

4 **Dunty JM**, Gabarra-Niecko V, King ML, Ceccarelli DF, Eck MJ, Schaller MD. FERM domain interaction promotes FAK signaling. *Mol Cell Biol* 2004; **24**: 5353-5368 [PMID: 15169899 DOI: 10.1128/MCB.24.12.5353-5368.2004]

5 **Bianconi D**, Unseld M, Prager GW. Integrins in the Spotlight of Cancer. *Int J Mol Sci* 2016; **17**: pii: E2037 [PMID: 27929432 DOI: 10.3390/ijms17122037]

6 **Mitra SK**, Schlaepfer DD. Integrin-regulated FAK-Src signaling in normal and cancer cells. *Curr Opin Cell Biol* 2006; **18**: 516-523 [PMID: 16919435 DOI: 10.1016/j.ceb.2006.08.011]

7 **Lim ST**, Mikolon D, Stupack DG, Schlaepfer DD. FERM control of FAK function: implications for cancer therapy. *Cell Cycle* 2008; **7**: 2306-2314 [PMID: 18677107 DOI: 10.4161/cc.6367]

8 **Zheng Y**, Xia Y, Hawke D, Halle M, Tremblay ML, Gao X, Zhou XZ, Aldape K, Cobb MH, Xie K, He J, Lu Z. FAK phosphorylation by ERK primes ras-induced tyrosine dephosphorylation of FAK mediated by PIN1 and PTP-PEST. *Mol Cell* 2009; **35**: 11-25 [PMID: 19595712 DOI: 10.1016/j.molcel.2009.06.013]

9 **Prifti S**, Zourab Y, Koumouridis A, Bohlmann M, Strowitzki T, Rabe T. Role of integrins in invasion of endometrial cancer cell lines. *Gynecol Oncol* 2002; **84**: 12-20 [PMID: 11748970 DOI: 10.1006/gyno.2001.6410]

10 **Van Slambrouck S**, Grijelmo C, De Wever O, Bruyneel E, Emami S, Gespach C, Steelant WF. Activation of the FAK-src molecular scaffolds and p130Cas-JNK signaling cascades by alpha1-integrins during colon cancer cell invasion. *Int J Oncol* 2007; **31**: 1501-1508 [PMID: 17982677 DOI: 10.3892/ijo.31.6.1501]

11 **Owen KA**, Abshire MY, Tilghman RW, Casanova JE, Bouton AH. FAK regulates intestinal epithelial cell survival and proliferation during mucosal wound healing. *PLoS One* 2011; **6**: e23123 [PMID: 21887232 DOI: 10.1371/journal.pone.0023123]

12 **Deakin NO**, Pignatelli J, Turner CE. Diverse roles for the paxillin family of proteins in cancer. *Genes Cancer* 2012; **3**: 362-370 [PMID: 23226574 DOI: 10.1177/1947601912458582]

13 **Bolós V**, Gasent JM, López-Tarruella S, Grande E. The dual kinase complex FAK-Src as a promising therapeutic target in cancer. *Onco Targets Ther* 2010; **3**: 83-97 [PMID: 20616959 DOI: 10.2147/OTT.S6909]

14 **Golubovskaya VM**, Ho B, Zheng M, Magis A, Ostrov D, Morrison C, Cance WG. Disruption of focal adhesion kinase and p53 interaction with small molecule compound R2 reactivated p53 and blocked tumor growth. *BMC Cancer* 2013; **13**: 342 [PMID: 23841915 DOI: 10.1186/1471-2407-13-342]

15 **Paoli P**, Giannoni E, Chiarugi P. Anoikis molecular pathways and its role in cancer progression. *Biochim Biophys Acta* 2013; **1833**: 3481-3498 [PMID: 23830918 DOI: 10.1016/j.bbamcr.2013.06.026]

16 **Golubovskaya VM**, Figel S, Ho BT, Johnson CP, Yemma M, Huang G, Zheng M, Nyberg C, Magis A, Ostrov DA, Gelman IH, Cance WG. A small molecule focal adhesion kinase (FAK) inhibitor, targeting Y397 site: 1-(2-hydroxyethyl)-3, 5, 7-triaza-1-azoniatricyclo [3.3.1.1(3,7)]decane; bromide effectively inhibits FAK autophosphorylation activity and decreases cancer cell viability, clonogenicity and tumor growth in vivo. *Carcinogenesis* 2012; **33**: 1004-1013 [PMID: 22402131 DOI: 10.1093/carcin/bgs120]

17 **Zhang J**, He DH, Zajac-Kaye M, Hochwald SN. A small molecule FAK kinase inhibitor, GSK2256098, inhibits growth and survival of pancreatic ductal adenocarcinoma cells. *Cell Cycle* 2014; **13**: 3143-3149 [PMID: 25486573 DOI: 10.4161/15384101.2014.949550]

18 **Kolev VN**, Tam WF, Wright QG, McDermott SP, Vidal CM, Shapiro IM, Xu Q, Wicha MS, Pachter JA, Weaver DT. Inhibition of FAK kinase activity preferentially targets cancer stem cells. *Oncotarget* 2017; **8**: 51733-51747 [PMID: 28881682 DOI: 10.18632/oncotarget.18517]

19 **Tai YL**, Chen LC, Shen TL. Emerging roles of focal adhesion kinase in cancer. *Biomed Res Int* 2015; **2015**: 690690 [PMID: 25918719 DOI: 10.1155/2015/690690]

P- Reviewer: Gazouli M, Lin Q, Nishiyama M, Sekar D, Tanabe S
S- Editor: Ji FF **L- Editor**: A **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 293-316

DOI: 10.4251/wjgo.v10.i10.293

ISSN 1948-5204 (online)

REVIEW

Simultaneous curative resection of double colorectal carcinoma with synchronous bilobar liver metastases

Emilio De Raffele, Mariateresa Mirarchi, Dajana Cuicchi, Ferdinando Lecce, Claudio Ricci, Riccardo Casadei, Bruno Cola, Francesco Minni

Emilio De Raffele, Dajana Cuicchi, Ferdinando Lecce, Claudio Ricci, Riccardo Casadei, Francesco Minni, Unità Operativa di Chirurgia Generale, Dipartimento dell'Apparato Digerente, Azienda Ospedaliero-Universitaria di Bologna, Policlinico S.Orsola-Malpighi, Bologna 40138, Italy

Mariateresa Mirarchi, U.O. di Chirurgia Generale, Dipartimento Strutturale Chirurgico, Ospedale "Antonio e Margherita," Tortona (AL) 15057, Italy

Bruno Cola, Dipartimento di Scienze Mediche e Chirurgiche (DIMEC), Alma Mater Studiorum, Policlinico S.Orsola-Malpighi, University of Bologna, Bologna 40138, Italy

ORCID number: Emilio De Raffele (0000-0003-1743-7471); Mariateresa Mirarchi (0000-0003-1896-2438); Dajana Cuicchi (0000-0002-1504-4888); Ferdinando Lecce (0000-0003-2042-0339); Claudio Ricci (0000-0001-5566-8444); Riccardo Casadei (0000-0002-4044-3557); Bruno Cola (0000-0002-3568-9835); Francesco Minni (0000-0002-9679-8971).

Author contributions: De Raffele E and Mirarchi M contributed to the conception and design of the study, acquisition, analysis, and interpretation of data, and wrote the manuscript; Cuicchi D, Lecce F, Ricci C and Casadei R contributed to acquisition, analysis, and interpretation of data; Cola B and Minni F made critical revisions and final approval of the paper.

Conflict-of-interest statement: The authors have no conflict of interest related to this publication.

Manuscript source: Invited manuscript

Correspondence to: Emilio De Raffele, MD, PhD, Surgeon, Surgical Oncologist, Unità Operativa di Chirurgia Generale, Dipartimento dell'Apparato Digerente, Azienda Ospedaliero-Universitaria di Bologna, Policlinico S.Orsola-Malpighi, Via Massarenti 9, Bologna 40138, Italy. e.deraffele@aosp.bo.it
Telephone: +39-51-6364235

Received: June 9, 2018
Peer-review started: June 9, 2018
First decision: July 13, 2018
Revised: July 28, 2018
Accepted: August 21, 2018
Article in press: August 21, 2018
Published online: October 15, 2018

Abstract

Synchronous colorectal carcinoma (SCRC) indicates more than one primary colorectal carcinoma (CRC) discovered at the time of initial presentation, accounts for 3.1%-3.9% of CRC, and may occur either in the same or in different colorectal segments. The accurate preoperative diagnosis of SCRC is difficult and diagnostic failures may lead to inappropriate treatment and poorer prognosis. SCRC requires colorectal resections tailored to individual patients, based on the number, location, and stage of the tumours, from conventional or extended hemicolectomies to total colectomy or proctocolectomy, when established predisposing conditions exist. The overall perioperative risks of surgery for SCRC seem to be higher than for solitary CRC. Simultaneous colorectal and liver resection represents an appealing surgical strategy in selected patients with CRC and synchronous liver metastases (CRLM), even though the cumulative risks of the two procedures need to be adequately evaluated. Simultaneous resections have the noticeable advantage of avoiding a second laparotomy, give the opportunity of an earlier initiation of adjuvant therapy, and may significantly reduce the hospital costs. Because an increasing number of recent studies have shown good

results, with morbidity, perioperative hospitalization, and mortality rates comparable to staged resections, simultaneous procedures can be selectively proposed even in case of complex colorectal resections, including those for SCRC and rectal cancer. However, in patients with multiple bilobar CRLM, major hepatectomies performed simultaneously with colorectal resection have been associated with significant perioperative risks. Conservative or parenchymal-sparing hepatectomies reduce the extent of hepatectomy while preserving oncological radicality, and may represent the best option for selected patients with multiple CRLM involving both liver lobes. Parenchymal-sparing liver resection, instead of major or two-stage hepatectomy for bilobar disease, seemingly reduces the overall operative risk of candidates to simultaneous colorectal and liver resection, and may represent the most appropriate surgical strategy whenever possible, also for patients with advanced SCRC and multiple bilobar liver metastases.

Key words: Colorectal surgery; Synchronous colorectal liver metastases; Major hepatectomy; Parenchymal-sparing hepatectomy; Intraoperative ultrasonography; Simultaneous colorectal and liver surgery; Synchronous colorectal carcinoma; Ablative therapies

Core tip: Simultaneous colorectal and liver resection represents an appealing surgical strategy in selected patients with colorectal cancer and resectable synchronous liver metastases (CRLM). Synchronous colorectal carcinoma may represent an adequate indication to simultaneous resections, even though it may require more complex colorectal resections. In patients with multiple bilobar synchronous CRLM, major hepatectomies performed simultaneously with colorectal surgery have been associated with increased perioperative risks compared to major hepatectomies alone. Conservative or parenchymal-sparing hepatectomies reduce the extent of hepatectomy while preserving oncological radicality, and may represent the best option to reduce the perioperative risks of simultaneous colorectal and liver resection.

De Raffele E, Mirarchi M, Cuicchi D, Lecce F, Ricci C, Casadei R, Cola B, Minni F. Simultaneous curative resection of double colorectal carcinoma with synchronous bilobar liver metastases. *World J Gastrointest Oncol* 2018; 10(10): 293-316 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/293.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.293

INTRODUCTION

Colorectal cancer (CRC) is one of the most frequent causes of cancer-related death in Western countries[1,2]. The development of at least two different neoplasms is defined as multiple primary CRC (MPCRC), which represents 5% to 10% of all CRCs[3,4]. Synchronous colorectal carcinoma (SCRC) indicates more than one primary CRC discovered in a single patient at the time of initial presentation, while neoplasms diagnosed some time after the resection and/or diagnosis of the first lesion are called metachronous CRC[3,4]. Compared with solitary CRC, SCRC possess distinctive features that need to be extensively investigated in preoperative evaluation to ensure adequate diagnosis and treatment[5]. SCRC account for 3.1% to 3.9% of CRCs[3,6], and may occur either in the same segment of the large intestine or separately in different colon segments[3,5]. Multiple factors, including inflammatory bowel diseases, hereditary nonpolyposis colorectal cancer (HNPCC) or Lynch syndrome, and familial adenomatous polyposis (FAP)[3,7], predispose to CRC and have also been associated with a higher risk of SCRC, though predisposing factors only account for a minority of cases[8]. Patients with SCRC have in most cases an overall oncological prognosis similar to those with solitary CRC, at least when the pathological stages of tumours are comparable and the resections are curative[4-6,8-13]. Nonetheless, the accurate preoperative diagnosis of SCRC remains difficult and diagnostic failures may lead to inappropriate treatment and poorer prognosis[5]. The presence of SCRC or multiple neoplasms requires operative techniques tailored to individual patients, based on the number, location, and stage of the tumours. Patients with SCRC and established predisposing conditions such as HNPCC, FAP, and ulcerative colitis require extensive surgery, usually total colectomy or proctocolectomy. In the other cases the optimal surgical strategy is still debated. Conventional hemicolectomies or extended hemicolectomies can be indicated if multiple tumours are located in adjacent segments[12]. When SCRC are located in distant colonic segments, some authors suggest total or subtotal colectomy[14,15], while others suggest more conservative surgical strategies with resection of two intestinal segments, either open or laparoscopic-assisted[13,16-18], seemingly resulting in a higher risk of anastomotic dehiscence[6]. However, overall perioperative results of colorectal resections for SCRC seem to be worse than those of solitary CRC with more postoperative complications and reinterventions and longer hospital stays[6]. As a consequence, an accurate preoperative workup and adequate surgical strategies are required for SCRC especially when adjunctive simultaneous surgical procedures are needed to obtain potential cure.

Synchronous liver metastases (CRLM) are evident in nearly 15% to 25% of patients with CRC at the time of diagnosis[1]. Radical liver resection (LR) is presently considered the only curative therapy capable of achieving long-term survival with more recent series describing 5-year overall survival (OS) rates of 37% to 58% after hepatectomy[19,20]. Nonetheless, the management of patients who present with CRC and synchronous metastases is more complex because they are considered to have less favourable cancer biology

and expected long-term results than those with metachronous liver disease[2,21]. The optimal timing for surgical resection in case of synchronous presentation of CRC and liver metastases is still controversial. Most surgeons usually prefer a staged approach with initial resection of the colorectal primary followed by hepatectomy[19], presuming that this strategy avoids increased perioperative complications associated with simultaneous procedures[20,22], and avoids also inappropriate hepatic surgery in patients with progression of the liver disease after colectomy especially if occurred during interval chemotherapy (CHT)[22]. More recently an increasing number of studies have shown satisfactory perioperative outcomes for simultaneous procedures comparable to those of staged strategies[19,23-30]. Simultaneous colorectal and liver procedures have the obvious advantage of avoiding a second surgical procedure, along with the chance of an earlier initiation of adjuvant CHT. However, an adequate evaluation of the cumulative risks of the two procedures is mandatory. In the last decade, the paradigm of surgical strategies for synchronous presentation of primary CRC and liver metastases is progressively changing, even though a consensus is far from being reached. Simultaneous colorectal resection and minor hepatectomy have perioperative results similar to minor hepatectomy alone, and are at present considered the treatment of choice in most patients with limited liver disease[19,23-30]. In patients requiring simultaneous colorectal and major LR the perioperative results are much more conflicting. Most investigators have reported worse perioperative outcomes than for major LR alone[20], while others remark that simultaneous colorectal and major hepatic resection can be performed safely in selected cases, with perioperative risks comparable to major LR alone[31-33]. Also simultaneous resection of rectal primaries and major hepatic resections have been considered reasonable in carefully selected patients[33,34].

Major hepatectomies have been traditionally preferred in the past to obtain radical resection of CRLM, especially in the case of large and/or multiple nodules. However, extensive hepatectomies have been associated with significant morbidity and mortality rates, usually related to posthepatectomy liver failure[35,36]. Several strategies have been developed to improve the feasibility of LR without increasing the risk of postoperative liver failure. Different systemic and locoregional chemotherapy protocols may significantly reduce the neoplastic burden in the liver with the aim of converting initially unresectable to resectable CRLM[37], but also of limiting the extension of LR[38]. Some technical innovations have permitted an increase in the amount of the future remnant liver (FRL) in candidates for major hepatectomy at increased risk of posthepatectomy liver failure based on the preoperative hyperplasia of the estimated remnant liver parenchyma, including preoperative portal vein embolization (PVE) and two-stage hepatectomy (TSH)[39]. An alternative strategy is to remove liver tumours with the minimum sufficient oncological margin to preserve as much non-tumourous liver parenchyma as possible, to limit the risk of liver failure in the perioperative period even for patients with advanced neoplastic liver disease[35], but also to preserve the major intrahepatic vessels whenever possible in order to increase the chance of reresection in case of hepatic recurrence (salvageability)[40,41]. In fact, reresection of relapsed CRLM has been widely demonstrated to have the potential for cure in selected patients with recurrent disease[20,42], with comparable morbidity and mortality rates than those of initial resection[43,44]. An accurate preoperative planning and an expert use of intraoperative ultrasonography (IOUS) are of paramount importance to achieve adequate oncological and surgical results. This strategy has been termed "conservative" or "parenchymal-sparing" liver resection (PSLR)[40,41]. A progressive shift toward more conservative hepatectomies has been observed in the last decade also for multiple and/or bilobar CRLM, and has been correlated with decreased morbidity and mortality rates and similar oncological results compared to major hepatectomies[45-47].

There is growing evidence, at least in numerous experimental studies, that surgical procedures for primary and metastatic CRC can activate multiple local and systemic events, such as hypoxia, inflammation, immune depression, release of multiple factors after the resection of the primary tumour and/or the CRLM, and release of tumour cells during surgical manipulation[48]. These events can exert local tumour-promoting effects, such as favouring the implantation and the proliferation of the residual neoplastic cells (predisposing the patient to local recurrences), activating dormant tumour cells in distant organs, and/or establishing a pre-metastatic niche (predisposing the patient to the occurrence of distant metastases)[48]. The real impact of these events in the clinical setting is still uncertain. On the other hand, LR activates within few hours multiple molecular changes (upregulation of several cytokines and growth factors) with subsequent activation and proliferation of mature hepatocytes, hepatic progenitor cells, and non-parenchymal liver cells to restore the optimal liver volume. These specific regenerative factors determine a complex microenvironment, which has been demonstrated to promote either the proliferation of residual cancer cells or tumour propagation in the remnant liver and also at distant sites, at least in various experimental models[48-52]. In patients with multiple bilobar CRLM, extended hepatectomies are traditionally considered to achieve potentially curative LR. In selected patients, PVE with or without TSH is proposed to induce preoperative hyperplasia of the FRL and increase the resectability rate. As for liver regeneration, several experimental and clinical studies have demonstrated that also PVE promotes tumour progression, either through an upregulation of cytokines and growth factors or by haemodynamic changes in the blood supply to the liver, which may adversely influence the subsequent management of the neoplastic disease[49,53-55]. Taken together, these experimental and clinical observations

support the theoretical advantages of simultaneous colorectal and liver resection, to prevent the drawbacks of multiple surgical procedures, and of conservative hepatectomies, to limit the impact of liver regeneration on tumour growth and metastatization.

The aim of the present review is to critically analyse the available data to determine whether complex colorectal resections for synchronous CRC are compatible with the simultaneous resection of CRLM, even in the case of multiple and/or bilobar CRLM.

SYNCHRONOUS COLORECTAL CARCINOMA

Epidemiology and predisposing conditions

The overall prevalence of SCRC ranges from 1% to 8% in different studies[3,6]. In four large multicentric studies including a study population between 13000 and 25000 patients with CRC, the prevalence ranged from 3.1% to 3.9%[6,10,11,56], while a recent systematic review pooling data from 39 series reported an overall prevalence of 3.5%[3]. In these series, SCRC had a higher male to female ratio when compared to solitary carcinoma, ranging between 1.5 and 2.2[3,6,10,11,56]. The mean age at presentation was 63 years in a systematic review pooling data from 32 series[3], usually higher than in patients with solitary CRC[3,6], even though this point is somewhat controversial[5,11]. Preferred locations of SCRC are still debated. Some authors have reported that many SCRC occur in the same segment of the large intestine, while others believe that most SCRC occur separately in different colon segments[3,5]. Moreover, SCRC are located in the ascending colon probably more often than described for solitary CRC[3,5,6], but also this point is controversial[5]. A minority of patients develop more than two SCRC[7,16], with a maximum of seven simultaneous colorectal lesions described in a single patient[56].

Possible predisposing factors, including inflammatory bowel diseases, HNPCC or Lynch syndrome, and FAP, to CRC have also been associated with a higher risk of SCRC[3,7]. SCRC has been diagnosed in up to 20% of patients with CRC associated with inflammatory bowel disease[57,58] (more frequently ulcerative colitis than Crohn's disease[59]), and in 21% of patients with CRC associated with FAP[57]. Patients with known predisposing factors might account for about 12% of SCRC[8]. Dysplasia induced by chronic inflammation and adenomas are involved in the development of SCRC in these patients[8,60]. Colorectal serrated polyps have more than a two-fold increase risk of detection of advanced CRC, with proximal and large serrated polyps having the highest risk[61]. Also the serrated neoplastic pathway may predispose to MPCRC[62]. Higher incidence rates of associated benign neoplasms have been described for SCRC than for single cancers[5,11,13]. The higher incidence of mucinous carcinoma in SCRC is still controversial[3,7]. Metachronous CRC can also occur after resection of SCRC, especially in patients with inflammatory bowel disease[59].

Mechanisms of carcinogenesis and molecular biology

MPCRC usually develop on a common etiologic substrate, either hereditary or environmental. Multiple recent studies on molecular carcinogenesis have demonstrated that chromosomal instability, microsatellite instability, and gene methylation are all mechanisms implicated in multiple lesions or events predisposing to SCRC. This may be due either to familial predisposition or more frequently to individual factors (mainly environmental exposure). Factors involved in the development of MPCRC have been recently reviewed[3-5,62]. CRC has a substantial heritable component[63]. Based on multicentric data derived from almost 45000 pairs of twins, the estimated effect of heritability on CRC is up to 35%[64], even though involved genetic factors are still incompletely understood. Well-known hereditary CRC syndromes, including HNPCC and FAP (which account for 3% to 5% of all CRC[65]), present germline mutations and promote the development of several neoplasms over time[8]. Other diseases and conditions, such as inflammatory bowel diseases, may extensively involve colorectal mucosa, thus promoting the formation of multiple foci of dysplasia and cancer[57]. In most cases however, the origin of SCRC is unknown, likely due to the coexistence of genetic predisposition and environmental factors[4]. As for other neoplasms, also for SCRC the concept of a field defect has been proposed to explain tumour multiplicity through a generalized cellular or molecular disorder in the entire colorectal mucosa[66]. Because only a minority of all SCRC are related to hereditary diseases, an important proportion of SCRC lack a clear basis of inheritance[4,9], being possibly related to individual predisposition to MPCRC. As for sporadic CRC, the prevalence of SCRC increases with age[9,10], indicating the possible role of cumulative environmental damage, even though this point has not been confirmed in other studies[8,11]. Alcohol intake and tobacco smoke, which consists of different genotoxic substances, have been related to an augmented risk of MPCRC[4,5].

Molecular biology and mechanisms of development of SCRC are heterogeneous. The majority of CRC follows the classical adenoma-carcinoma sequence of tumour progression, and dysplastic adenomas are the most common form of premalignant precursor lesions[63,67]. However, more than 15% of sporadic CRC develop through alternative pathways of molecular events, including cancers originating from serrated precursor lesions[63,68]. Molecular pathways of development of SCRC have been recently reviewed[3,5,9,66,68-70] and are out of scope for this review. Nonetheless, the complex mechanism of carcinogenesis involved in the development of SCRC is still largely unknown and only partially related to known genetic mutations commonly found in CRC.

Prognosis

The prognosis of patients with SCRC compared to solitary CRC is still debated. Even though the first prospective study on the outcome of SCRC reported worse long-term results than solitary CRC[15,69], most recent studies could not demonstrate different survival rates between SCRCs and CRCs when the pathological stages of tumours were matched and the resections were curative. However, some authors have reported marginal survival benefits of patients with SCRC[3,5,6,8-13].

Diagnosis

The preoperative diagnosis of multiple SCRC remains difficult (Table 1). Additional tumours may be ignored or missed at the time of diagnosis of the first cancer, with diagnostic failure leading to inappropriate treatment and poorer prognosis[5]. Routine preoperative colonoscopy is mandatory to identify synchronous neoplasms[71]. Because preoperative evaluation of the colon during colonoscopy is often incomplete due to bowel obstruction, poor bowel preparation, or technical reasons, double-contrast barium enema and computed tomographic (CT) colonography, magnetic resonance (MR) colonography, and/or positron emission tomography/computer tomography (PET/CT) colonography are advisable[5,63,72-74]. Also the use of intraoperative colonoscopy has been recommended in selected cases[5,16,75]. At the time of operation, it is also important to palpate the entire colon and check pathological specimens thoroughly[5,16]. An adequate combination of these imaging techniques with the traditional colonoscopy usually permits an accurate definition of number and location of synchronous colorectal neoplasms and an appropriate plan of the optimal surgical procedures[6]. Patients with mid and low rectal adenocarcinoma should routinely receive endorectal ultrasound and pelvic magnetic resonance imaging because the quality of preoperative imaging for local staging is essential to pursue an appropriate therapeutic strategy[76-78], which includes perioperative chemoradiotherapy and surgical resection for locally advanced extraperitoneal tumours[77,78].

Surgical treatment strategies

The standard surgical procedure for the treatment of rectal cancer is total mesorectal excision consisting of the removal of the rectum together with the mesorectum, which contains most of the involved lymph nodes and tumour deposits, and the mesorectal fascia[76] along with clear circumferential margins[77]. The appropriate removal of the rectal cancer reduces the risk of local recurrence and the development of distant metastases[77,78]. Surgical procedures for colon cancer entail resection of the tumour with the corresponding lymph nodes. The extent of colonic resection is determined by the tumour location and the supplying blood vessels. The presence of SCRC or multiple neoplasms requires operative techniques tailored to individual patients based on the number, location, and stage of the tumours. Patients with SCRC and established predisposing conditions such as HNPCC, FAP, and ulcerative colitis require extensive surgery, usually total colectomy or proctocolectomy. In the other cases, the optimal surgical strategy is still debated. Early-stage lesions can be removed during colonoscopy with endoscopic mucosal or submucosal resection. Hemicolectomy or extended hemicolectomy can be indicated if multiple tumours are located in adjacent segments[12]. When SCRC are located in distant colonic segments, some authors suggest total or subtotal colectomy to remove synchronous tumours or polyps eventually undetected at preoperative imaging and to prevent the development of metachronous neoplasms[14,15]. In the same circumstances, other authors suggest more conservative surgical strategies, with resection of two intestinal segments (either open or laparoscopic-assisted)[13,16-18] and two anastomoses, seemingly resulting in a higher risk of anastomotic dehiscence[6].

Perioperative results of colorectal resections for SCRC are also debated. In a multicentric study of 884 patients who were operated for SCRC between January 2009 and December 2011 and were registered in the Dutch Surgical Colorectal Audit[6], extended surgery (*e.g.*, subtotal colectomy, proctocolectomy, or combined resection) was performed in more than 35% of cases. The application of neoadjuvant chemoradiation for rectal tumours was lower for synchronous than for solitary CRC (20% *vs* 38%), laparoscopic resections were less frequent, and more (permanent and deviating) stomas were constructed during surgery than for solitary tumours. Overall, the perioperative outcomes of SCRC were worse than for solitary CRC: postoperative complications, reinterventions, 30-day mortality, and time of hospital stay were significantly increased in patients with SCRC. After adjustment for patient- and tumour-related factors, having SCRC was still associated with a higher risk of severe postoperative complications and reinterventions, but not with higher 30-d mortality. The authors concluded that the higher risk of unfavourable perioperative outcomes could be explained by the more extended surgical resection often required for SCRC. Holubar *et al*[17] reported 69 patients who underwent multiple colonic anastomoses, laparoscopic-assisted in ten (17%) cases, with a 44% conversion rate. Length of stay was seven (5-10) days, overall 30-day morbidity was 36% without anastomotic leaks or fistulas, and 30-day mortality was 3%. Li *et al*[18] examined a personal series of 11 patients and 52 adjunctive patients collected from six previous reports of the literature who underwent laparoscopic-assisted combined bowel anastomoses for SCRC, and concluded that combined bowel anastomoses are potentially feasible and safe procedures for SCRC when performed by experienced surgeons.

Table 1 Diagnostic evaluation of synchronous colorectal cancer

Local tumour staging
Preoperative colonoscopy with histological assessment of all colorectal lesions
CT of the abdomen and pelvis
In case of rectal cancer include
Endorectal ultrasound
Pelvic magnetic resonance imaging
If preoperative evaluation during colonoscopy is incomplete (bowel obstruction, poor bowel preparation, technical reasons, *etc.*)
Double-contrast barium enema
CT colonography, if available
MRI colonography, if available
PET–CT colonography, if available
Intraoperative assessment
Intraoperative colonoscopy
Palpation of the entire colon
Thorough examination of pathological specimens
Evaluation of metastatic disease
CT of the chest, abdomen, and pelvis
MRI of the chest, abdomen, and pelvis, in selected cases
18FDG-PET–CT, in selected cases
Patient performance status
Thorough evaluation of coexisting morbidities
Pulmonary function tests, in selected cases
Echocardiography, in selected cases

CT: Computer tomography; MRI: Magnetic resonance imaging; PET-CT: Positron emission tomography/computer tomography.

SURGICAL STRATEGIES FOR SYNCHRONOUS CRLM

Surgical strategies in patients with resectable CRC and upfront resectable synchronous metastases limited to the liver have been widely debated in the last decades. The traditional "staged" or "classic" approach with resection of the colorectal tumour followed by hepatectomy is probably still favoured in most cases because the risks of the colorectal and the liver surgery are not cumulated[20,22,79,80], and CHT can be selectively administered between the two procedures[22]. In the case of large synchronous CRLM and uncomplicated primary tumour, a reversed therapeutic strategy with LR followed by colorectal resection has been proposed, to minimize the risk of progression of the metastatic liver disease to unresectability. This strategy is termed "reverse" or "liver-first" approach[22,81,82] and has become more widely used, either in patients with borderline resectable liver involvment and uncomplicated primary tumour or in patients with resectable CRLM and locally advanced rectal cancer that can be treated with neoadjuvant chemoradiotherapy and subsequent rectal surgery[22,81,83-85]. Moreover, in a small proportion of patients, a complete clinical, endoscopic, and radiological response of the primary tumour to chemoradiotherapy subsequent to initial radical LR has been reported, thus delaying or even avoiding bowel surgery[85]. However, simultaneous colorectal and liver resection remains the most appealing approach and is obtaining a growing consensus due to the advances in oncological concepts and continued development of anaesthesia, critical care, radiological imaging, and techniques of hepatobiliary surgery favouring the expansion of resectability criteria[40]. Simultaneous resections have clear advantages because the patient experience is improved and psychological stress is limited by decreasing the time to removal of the disease, the total number of surgical procedures, the duration of perioperative CHT[19,29]. Also the cumulative costs of hospitalization are substantially decreased in selected cases[86]. Nonetheless, the real impact on the oncological outcome and on the perioperative results are still debated[2,20].

Preoperative assessment

The accurate preoperative staging of advanced CRC is of paramount importance (Table 2) and can be obtained with cross-sectional imaging by CT or MRI[1,2,87,88]. The current guidelines of the North American National Comprehensive Cancer Network (NCCN) suggest the use of CT or MRI of the chest, abdomen, and pelvis. 18FDG-PET–CT imaging is reserved for patients who may undergo potentially curative surgical resection[2]. Preoperative liver imaging should be accurately evaluated to define the number and the site of CRLM, the tumour-vessels relationship, the pattern of intrahepatic vasculature, the presence of anatomical variations, and the FRL volumes[35,89-91]. Recent studies underline the favourable impact of preoperative MRI on the overall oncological outcome of patients with multiple CRLM[92]. The accurate assessment of patient performance status is mandatory to determine suitability for more complex therapies, especially those including liver surgery. Coexisting morbidities and liver steatosis should be

Table 2 Diagnostic evaluation of synchronous colorectal liver metastases

Local tumour staging
CT and/or MRI of the liver, to evaluate
Number and location of CRLM
Tumour-vessels relationship
Pattern of the hepatic vasculature
Presence of anatomical variations
Future remnant liver volumes
Intraoperative assessment
Intraoperative ultrasonography
Evaluation of metastatic disease
CT of the chest, abdomen, and pelvis
MRI of the chest, abdomen, and pelvis, in selected cases
18FDG-PET–CT
Patient performance status
Thorough evaluation of coexisting morbidities
Pulmonary function tests
Echocardiography
In the case of suspected liver disease/steatosis include (elderly patients, metabolic syndrome, previous systemic CHT, *etc.*)
Liver function tests
Evaluation of the grade of steatosis, in selected cases

CT: Computer tomography; CRLM: Colorectal cancer and synchronous liver metastases; CHT: Chemotherapy; MRI: Magnetic resonance imaging; PET-CT: Positron emission tomography/computer tomography.

adequately assessed. Accurate stratification of the perioperative risks should include liver function tests with evaluation of the grade of steatosis in selected cases, and pneumological and cardiological evaluation with pulmonary function tests and echocardiography[88]. Even though up to 70% of the normal adult human liver can be removed, previous systemic CHT may seriously alter liver function and the consequent ability to tolerate extended resections[93-96]. Oxaliplatin-based regimens are associated with augmented risks of vascular lesions, including the sinusoidal obstruction syndrome (SOS), which has been reported to increase morbidity after major LR, especially after administration of more than six cycles[97]. Irinotecan-based regimens are associated with the occurrence of various degrees of steatosis up to the chemotherapy-associated steatohepatitis (CASH), which may worsen perioperative morbidity and mortality rates after LR[97]. The impact of adding targeted molecular therapies, including cetuximab or bevacizumab, to conventional systemic chemotherapy on perioperative morbidity or mortality rates after hepatectomy is still controversial[97].

Simultaneous vs staged colorectal and liver resection

Many recent systematic reviews and meta-analyses have compared the perioperative and long-term outcomes of simultaneous versus delayed hepatectomy for synchronous CRLM. In a systematic review of the literature including 16 controlled trials comparing simultaneous resection of synchronous CRLM and of the primary cancer with a staged approach, where the metastases were resected at a later stage, there was a tendency towards shorter hospital stays and lower perioperative morbidity after simultaneous resection[23]. Perioperative mortality seemed to be lower with the staged approach, and five-year survival rates seemed to be similar in the two groups. The authors underlined that all studies were retrospective and had a general bias because staged procedures were significantly preferred in patients with left-sided primary CRC and larger, more numerous and bilobar metastases. They concluded that simultaneous resections might be selectively undertaken. In a meta-analysis evaluating 14 comparative studies comprising 2204 patients[24], those undergoing simultaneous resection had similar operative time and intraoperative blood loss, shorter hospital stay, and lower morbidity rate. One-, three- and five-year survival rates were similar between groups. The authors concluded that simultaneous resection is a safe and effective treatment for patients with synchronous CRLM and might be considered as the preferred treatment in appropriately selected patients. Another systematic review and meta-analysis of 19 non-randomized controlled trials including 2724 patients came to similar conclusions[25]. Yin *et al*[26] conducted a systematic review and meta-analysis of 17 retrospective studies including 2880 patients, of whom 1015 with simultaneous resection and 1865 with delayed resection. The simultaneous group had lower postoperative complications, whereas postoperative mortality within 60 d and overall and recurrence-free survival (RFS) were similar between groups. Moreover, the authors proposed precise selection criteria for patients suitable for a simultaneous resection, including LR of no more than three segments, colon resection (especially the right-sided colectomy), age < 70 years, and exclusion of severe comorbidities.

Somewhat different conclusions were drawn in a wider meta-analysis including 24 studies published between 1991 and 2010, which comprised 3159 patients, of whom 1381 had simultaneous resections and 1778

had delayed resections[27]. Significantly fewer patients received neoadjuvant CHT in the simultaneous resection group. The bilobar distribution (P = 0.01), the size of CRLM (P < 0.001), and the proportion of major LR (P < 0.001) were found to be higher in the delayed resection group. Operative blood loss and length of surgery were similar between groups, and length of hospital stay was significantly reduced in simultaneous resections (P = 0.007). Post-operative complications, OS, and disease-free survival (DFS) were similar between groups. The authors concluded that delayed resections may result in better outcomes because patients undergoing delayed resection had intraoperative parameters, postoperative complications, and survival rates comparable to those of patients undergoing simultaneous resection, despite more extensive metastatic liver disease. A subsequent meta-analysis evaluating 4494 patients from 22 studies published between January 2000 and April 2013[28] questioned the reliability of some previously published meta-analyses because important biases of the examined retrospective studies, mainly the fact that significantly more patients with mild conditions underwent simultaneous procedures, were not corrected. Summarized baseline analyses to find imbalanced factors between simultaneous and staged groups showed that patients were more likely to undergo simultaneous resection when they had less CRLM (single nodule, P = 0.002; ⩽ 3 nodules, P < 0.0001), of smaller size (diameter ⩽ 5 cm, P = 0.04; smaller mean diameter, P < 0.00001), with unilobar distribution (P = 0.0002), requiring minor LR rather than major LR for curative resection (P < 0.00001), and a right-sided CRC rather than left-sided (P = 0.0006). After correction of baseline imbalance, simultaneous and staged resections had comparable safety and efficacy, with similar postoperative morbidity and mortality, and overall and disease-free survivals. Similar results were found in another recent systematic review and meta-analysis of 30 studies including 5300 patients, of whom 2235 patients received simultaneous resections and 3065 patients received staged resections[26]. Patients undergoing delayed surgery were more likely to have received neoadjuvant treatment, have bilobar disease, or undergo major LR. Parameters relating to safety and efficacy were similar between the two groups. The average length of hospital stay was six days shorter with the simultaneous approach (P < 0.001). Long-term survival was similar for the two approaches.

The discordant results of the numerous meta-analyses published in recent years is due to the limitations intrinsic to meta-analysis of retrospective studies, mainly due to the fact that compared to RCTs retrospective studies are not randomized. As a consequence, experimental and control groups are often poorly comparable, and the baseline imbalances may significantly compromise the accuracy of the results. Without adequate correction of baseline imbalances before pooled analyses, ideally using methods based on the individual patient data analysis (which however is not always available), meta-analyses can only improve the precision, not the accuracy, of the pooled results, which should be interpreted and applied with great prudence[28]. The copious studies comparing simultaneous and classical staged resections, where the colorectal resection is followed by hepatectomy usually with interval CHT, must be interpreted cautiously because at least two major confounding factors are usually present. Candidates to simultaneous resection were usually younger, in better clinical conditions, with right-sided primary cancer, and more limited liver involvement usually necessitating minor hepatectomies[23,26-30]. On the other hand, patients enrolled in the staged groups included significantly more patients who received pre-operative CHT[27-29], and only those who had received successful staged resections, while patients who developed progressive liver disease during the interval were excluded. For these reasons, the overall survival of patients selected for staged approaches could be overestimated by including only patients with more favourable cancer biology or responsive to perioperative (neoadjuvant and/or interval) CHT. Future studies should avoid this selection bias by including patients with progressive metastatic disease after colorectal resection that missed the subsequent hepatectomy[28].

More recent studies have compared all the available surgical strategies, the staged primary-first *vs* the staged liver-first *vs* the simultaneous resection. In a small series of 57 patients with rectal cancer and synchronous CRLM, the authors compared the traditional staged resections with the simultaneous resections and the liver-first approach[98]. The overall morbidity rate was 24.6%, without in-hospital mortality. The median in-hospital stay was significantly shorter for the simultaneous approach. The five-year OS rate was 38%, with an estimated median survival of 47 mo. The authors concluded that long-term survival can be achieved using an individualized approach in patients with rectal cancer and synchronous CRLM and that simultaneous procedures as well as the liver-first approach are attractive alternatives to traditional staged procedures. In another series of 156 consecutive patients with synchronous CRLM, Brouquet *et al*[81] compared the results of the three different surgical strategies, and found comparable three- and five-year OS rates. The only factors independently associated with the OS were a liver tumour size > 3 cm and the cumulative perioperative morbidity. Similar conclusions have been drawn in a multi-institutional study including over 1000 patients from four major hepatobiliary centres[82]. The median OS was 50.9 mo and the cumulative one-, three- and five-year survivals were 89%, 60%, and 44%, respectively, without significant differences between simultaneous and staged surgical procedures. The cumulative recurrence rate was 57%, and was similar between patients undergoing simultaneous and staged procedures. Independent factors of worse long-term prognosis were being male, a rectal primary, and combined LR plus ablation. The authors concluded that tumour biology rather than surgical strategy was the main effector of the oncological

outcome. A systematic literature review of 18 studies comparing the different surgical approaches in patients with synchronous CRLM concluded that none of the three surgical strategies appeared inferior to the others[99]. Similarly, a network meta-analysis review of 3605 patients comparing classic staged, simultaneous, and liver-first surgical strategies could not demonstrate significant differences of 30-day mortality, postoperative complications, and five-year OS rates[100]. In a systematic review of three cohort studies comprising a pooled population of 1203 patients who underwent surgical treatment of CRC with synchronous CRLM between 1982 and 2011 and where the different treatment modalities were reported separately[101], 62.2% of patients received bowel-first surgery, 6.2% of patients received liver-first surgery, and 31.6% of patients received simultaneous surgery. Perioperative outcomes were similar between the three methods with low overall treatment-related mortality and similar survival rates.

Neoadjuvant CHT in resectable liver disease

Strategies including different CHT protocols to augment resectability in the case of initially unresectable synchronous CRLM are out of scope for this review. The role of neoadjuvant CHT in patients with resectable CRLM is still controversial. The EORTC Intergroup trial 4098386 was a randomized comparison of perioperative oxaliplatin-based CHT administered either before or after LR *vs* LR alone in patients with limited CRLM ($\leqslant$ 4) classified as resectable at baseline assessment[102]. Thirty-five percent of patients had synchronous disease. The overall results revealed an absolute increase in the rate of progression-free survival at three years in the patients randomized to receive perioperative CHT, but significantly more frequent reversible postoperative complications in the same group. However, the absolute differences in outcomes observed between groups were small and the study received much criticism[28,30]. Moreover, a long-term follow-up report of this trial could not find any difference in survival between the groups[103]. A systematic review of 23 trials evaluating the clinical response and outcomes of neoadjuvant systemic CHT for resectable CRLM suggested that preoperative CHT may achieve objective response with improvement in DFS[104]. However, also this study was considered to have enough limitations to affect the final conclusions[30]. Another systematic literature review concluded that, while combination regimens resulted in enhanced tumour response and resectability rates in up to 30% for unresectable CRLM, studies on neoadjuvant CHT failed to convincingly demonstrate a survival benefit for resectable lesions, with most reports describing increased postoperative complications in a subset of patients due to parenchymal alterations associated with CHT[97]. A recent analysis of a multi-centric cohort from the LiverMetSurvey International Registry, which included patients who had received curative LR for synchronous CRLM, compared 693 patients who received neoadjuvant CHT prior to liver surgery with 608 patients treated by surgery alone, and could not find any survival advantage between the groups[105]. Discouraging results were also obtained associating the targeted molecular agent cetuximab with conventional neoadjuvant CHT protocols[106].

CONSERVATIVE OR PARENCHYMAL-SPARING LIVER SURGERY

Resectability of CRLM has significantly improved over the last decades. The traditional criteria related to the features of liver tumours to evaluate resectability have been replaced by an accurate preoperative estimation of what remains after LR. Tumours should be considered resectable if complete liver tumour excision can be obtained with curative intent (macroscopically uninvolved surgical margins), in the absence of unresectable extrahepatic disease, and the estimated FRL parenchyma is sufficient to prevent liver failure[107]. Major liver resections, including conventional major hepatectomies and more recently described two-stage procedures, with or without PVE, are traditionally preferred by most surgeons to obtain radical resection of CRLM, especially in the case of large and/or multiple nodules. However, extensive hepatectomies have been associated with significant morbidity and mortality rates, usually related to posthepatectomy liver failure[35,36]. "Conservative" or "parenchymal-sparing" hepatectomies are based on the expert use of IOUS, which permits removal of liver tumours with the minimum sufficient oncological margin to preserve as much non-tumourous liver parenchyma as possible, to limit the risk of perioperative liver failure[35], but also to preserve the major intrahepatic vessels whenever possible with the aim of increasing salvageability in case of hepatic recurrence[40,41]. The progressive diffusion of conservative strategies of LR is related to at least three factors: The increasing evidence that CRLM have different intrahepatic diffusion patterns than hepatocellular carcinoma, so that anatomical resections per se have no impact on the oncological outcome; the evolution of the concept of adequate surgical resection margin (RM), where the "1-cm rule" proposed by Ekberg *et al*[108] has been progressively abandoned in favour of the concept of "negative margin" without considering margin width; and the increasing evidence that also patients with large numbers of CRLM are potential candidates for curative liver surgery in the context of multimodal treatment strategies of advanced CRC.

Anatomic vs non-anatomic resection

Adequate resection of liver tumours should involve resection of the tumour with enough margin to prevent recurrence and to achieve potentially curative treatment. Hepatocellular carcinoma has a high propensity for vascular invasion and metastatic spread through the portal venous system. As a consequence, anatomic

resection (AR) is considered the optimal surgical strategy because it eradicates portal tributaries close to the tumour, possibly reduces the risk of local tumour spread, and may ultimately determine a survival benefit compared to non-anatomic resection (NAR)[35,109]. Multiple surgical strategies which limit the extension of LR while respecting the segmental or subsegmental distribution of intrahepatic vessels have been described over the last 30 years and successfully performed due to the expert use of IOUS, either for primary or for metastatic liver tumours[40,110-114]. Metastatic tumours can spread within the liver by different pathways. Neoplastic cells might disseminate within and outside the liver through portal and hepatic veins, lymphatic vessels, bile ducts, and perineural spaces[115]. Sasaki *et al*[116] defined portal vein, hepatic vein, and bile duct invasion as the growth of cancer cells into blood vessels or bile duct branches in the liver parenchyma, and defined intrahepatic lymphatic invasion as the growth of cancer cells in luminal structures located in the portal spaces and lined by endothelial cells. Korita *et al*[117] described intrahepatic lymphatic invasion as the presence of isolated cancer cells or cell clusters within vessels with immunoreactivity for D2-40 antibody[117,118]. Other studies about the prognostic role of different patterns of intrahepatic diffusion of CRLM did not describe the method used to define vascular invasion, so that differentiation between invasion of blood vessels and of lymphatic vessels was uncertain[115]. With these limitations, the prognostic role of the portal vein and the hepatic vein invasion is still uncertain[115,118], while migration of tumour cells from CRLM through intrahepatic lymphatic vessels has a documented adverse impact on survival[116-119]. For these reasons, AR including portal tributaries close to the tumour and the corresponding liver parenchyma should not be theoretically justified for CRLM, and NAR with adequate surgical margin is presently considered an appropriate surgical strategy[35,90,120-125]. A recent meta-analysis of seven non-randomized controlled studies including 1662 patients with CRLM, compared 989 patients who underwent AR and 673 who underwent NAR[121]. NAR reduced the operation time and blood transfusion requirements whereas postoperative morbidity and mortality were similar between groups. Also oncological outcomes, including surgical margins, OS, and DFS survival were similar between the groups. Another systematic review of 12 studies included 2005 patients, who underwent either PSLR (1087 patients) or AR (1418 patients) for CRLM[122]. Most studies included a large subset of patients with solitary tumours and a reported median tumour number of one to two regardless of surgical strategy. While there was considerable inter-study variability regarding RM status, there was no difference in the incidence of R0 resection between groups. Median postoperative length-of-stay was similar; also OS was similar after PSLR (five-year OS: mean 44.7%, range 29%-62%) and AR (five-year OS: mean 44.6%, range 27%-64%). The authors concluded that PSLR had comparable safety and efficacy profiles compared with AR without compromising oncological outcomes.

Since the early 2000s, the systematic use of conservative procedures of LR, either for primary or for metastatic liver tumours, has been considered of paramount importance in some Japanese studies to achieve zero mortality and low morbidity rates. Meticulous attention to the balance between the hepatic functional reserve and the hepatic volume to be removed, the routine use of NAR with adequate surgical margin for resection of liver metastases whenever possible, and the attitude to perform simultaneous colorectal and liver resections for synchronous CRLM were among the most important criteria to perform safe hepatectomies without perioperative mortality[35]. Kokudo *et al*[123] retrospectively evaluated 115 patients with unilobar single or double tumours undergoing major AR (64 patients) or limited NAR (51 patients) and found that survival rates were similar between the groups. Anatomical major hepatectomy was unnecessary in 80.4% of the cases if the tumours were resectable by limited NAR, and 90% of the ipsilateral recurrence, which could have been avoided if the first operation was anatomical hemihepatectomy, could undergo a second hepatectomy with a five-year survival rate of 58.3%. The authors concluded that limited NAR should be a basic surgical procedure for CRLM to minimize surgical stress and operative risks. Mise *et al*[124] have recently evaluated a series of 300 patients with a solitary CRLM $\leqslant$ 30 mm undergoing PSLR (156 patients) or more extended hepatectomy (144 patient), including right hepatectomy, left hepatectomy, or left lateral sectionectomy. The rate of PSLR increased during the 20-year study period. PSLR did not negatively impact OS, RFS, and liver-only recurrence-free survival compared to non-PSLR. Repeat LR was more frequently performed in the PSLR group (68% *vs* 24%, $P < 0.01$). Subanalysis of patients with recurrence limited to the liver revealed better five-year OS from initial LR (72.4% *vs* 47.2%; $P = 0.047$) and from hepatic recurrence (73.6% *vs* 30.1%; $P = 0.018$) in the PSLR group. Upon multivariate analysis, non-PSLR was an independent significant risk of non-candidacy for repeat hepatectomy. The authors concluded that conservative resections did not increase recurrence in the liver remnant while increasing the opportunity of salvage reresection and the five-year survival rate in case of recurrence. These results have been subsequently confirmed in a multicentric cohort of 1720 patients from the LiverMetSurvey registry, with a single CRLM $\leqslant$ 30 mm located in the right hemiliver[125]. Eight-six percent of patients underwent PSLR and fourteen percent underwent right hepatectomy. PSLR was associated with lower major complication rates (3% *vs* 10%; $P < 0.001$) and 90-day mortality rates (1% *vs* 3%; $P = 0.008$). Hepatic recurrence was similar between groups (20% *vs* 22%; $P = 0.39$), as well as the five-year OS and RFS rates. However, in patients with liver-only recurrence, repeat LR was more frequently performed after PSLR than after right hepatectomy (67% *vs* 31%;

$P < 0.001$), and the five-year OS rate was significantly higher after PSLR than after right hepatectomy (55% *vs* 23%; $P < 0.001$). Taken together, these results indicate that a combination of conservative NAR followed by liver reresection in the case of recurrence limited to the liver offers superior oncological benefits than major LR in most patients with limited hepatic disease, and should be considered at present the most appropriate surgical strategy[123-125].

Similar results have been recently reported in patients with two or more CRLM. Karanjia *et al*[126] evaluated 283 consecutive patients who underwent successful LR for CRLM over ten years and compared 128 patients who had right and extended right hepatectomy with 155 patients who had other types of LR. Operative mortality was 3.9% and 0.7% after right hepatectomy and after other types of LR, respectively ($P = 0.04$). Morbidity was 31.3% and 18% after right hepatectomy and after other types of LR, respectively. The one-, three- and five-year OS rates were 84.1%, 54.3%, and 38.9% after right hepatectomy and 95.4%, 65.9%, and 53.3% after other types of LR, respectively ($P = 0.03$). The one-, three- and five-year DFS rates were 69.5%, 34.4%, and 25.5% after right hepatectomy and 68.4%, 34.91%, and 34.91% after other types of LR, respectively ($P = 0.46$). The authors concluded that in patients with CRLM, right and extended right hepatectomy have greater operative morbidity and mortality and significantly worse OS compared to all other types of LR. In a more recent series of 917 consecutive patients who received LR for CRLM from 2000 to 2010, Lordan *et al*[127] compared 238 patients who underwent PSLR case-matched with 238 patients who had major hepatectomy using a propensity scoring system. Fewer PSLR patients received perioperative blood transfusions ($P < 0.0001$). PSLR patients had a lower incidence of complications ($P = 0.04$), grade Ⅲ/Ⅳ complications ($P = 0.01$), 90-day mortality ($P = 0.03$), and a shorter hospital stay ($P = 0.04$). OS and DFS rates were similar. The authors concluded that patients with resectable CRLM should be offered PSLR if technically feasible because PSLR is safer than major hepatectomy without compromising long-term survival. Parenchymal-sparing hepatectomies are effective also for CRLM deeply placed where major hepatectomies have been traditionally preferred. Matsuki *et al*[128] evaluated 63 patients who received first curative LR for deeply placed CRLM whose centre was located > 30 mm from the liver surface. PSLR and major hepatectomy were performed in 63% and 37% of patients, respectively. Resected volume was smaller after PSLR than after major hepatectomy (251 g *vs* 560 g) ($P < 0.01$). Total operation time, amount of blood loss, rate of major complications, and positive operative margins were similar. OS, RFS, and liver recurrence-free survivals did not differ between the two groups. The authors underlined that direct major hepatectomy without PVE was unfeasible in 40% of the PSLR group because of the small FLR and concluded that PSLR for deeply placed CRLM can be performed safely without compromising oncologic radicality and can also increase the number of patients eligible for a direct surgical treatment by limiting the resection volume.

Resection margin

There has long been controversy over the impact of the width of the resection margin on the oncological outcome of LR candidates for CRLM. Since the 1980s surgeons have advocated for R0 resection margins of 10 mm or greater, the so-called "1-cm rule", in order to prevent local recurrence and optimize overall survival[38,108,129]. The presence of residual microscopic deposits of neoplastic cells after removal of metastatic nodules is considered an important source of remetastasis and a significant factor of adverse prognosis[115,129]. As for the primary tumour, micrometastases may occur in CRLM. Intrahepatic micrometastases are defined as detectable microscopic tumour nests within the liver parenchyma or portal tracts surrounding the dominant tumour, but separated by a rim of non-tumourous parenchyma, are predominant within 4 mm to 10 mm of the tumour margin, and are considered the morphological expression of remetastasis from existing liver metastases[40,119,130,131]. Their role as a prognostic factor in the oncological outcome of patients with CRLM is however still controversial. One study reported that patients with intrahepatic micrometastases had higher incidence of intrahepatic recurrence and worse survival, with ten-year survival rates of 21.9% compared to 64.3% for patients without micrometastases[132]. In another study, intrahepatic micrometastases were less frequently detected in patients treated with neoadjuvant CHT than in those untreated[133]. A 2 mm RM is however considered acceptable to significantly reduce the incidence of local recurrence in the series where the role of intrahepatic micrometastases has been evaluated[119,130]. In a small series based on the detection of tumour-specific mutant DNA in liver tissue surrounding metastases, mutant DNA was detectable in surrounding liver tissue within 4 mm of the tumour border, while biopsies at 8 mm, 12 mm, and 16 mm from the macroscopically visible margin were free from microscopically visible tumour cells and detectable mutant DNA, even in patients whose tumours were larger before CHT[131]. Also the presence of fibrotic tissue between the tumour and the surrounding hepatic parenchyma has been recognized as a favourable prognostic factor in CRLM and may be relevant in the evaluation of the RM. Yamamoto *et al*[134] reported that the five-year survival rate was 71% in patients with a thick pseudocapsule, 63% in those with a thin pseudocapsule, and only 19% in the absence of a pseudocapsule. Similar results were reported in the study by Okano *et al*[135], where five-year survival rates were 88% in patients with a thick pseudocapsule, 64% in patients with a thin pseudocapsule, and 31% in those without a pseudocapsule. Taken together, these data show that CRLM are usually well circumscribed, with very low incidences of satellite nodules or micrometasta-

ses, so that limited negative resection margins may have a limited impact on recurrence and survival rates, even though RM width of 10 mm should be achieved whenever possible[38].

R1 resection

The presence of residual macroscopic or microscopic tumour on RM after surgery for CRLM is traditionally considered a significant factor of adverse prognosis[108] due to increased local and intrahepatic recurrence as well as decreased OS and DFS. As a consequence, the adequate evaluation of the RM is of paramount importance to define the postoperative oncological prognosis. However, the accurate assessment of margin status depends on multiple factors. Different techniques of liver transection create different extensions of tissue loss[38]. The thermal effects of energy devices and of the argon-beam coagulation on the cut surface of the liver causes extensive cell killing within 2-5 mm of the RM[130,136]. Also pathologic assessment of the exact distance between the excised tumour and the end of the liver parenchyma has multiple limitations[137]. With these limitations, there is strong evidence that microscopically positive RM (R1) negatively impacts overall oncological results. R1 resection has been associated with an increased risk of recurrence at the surgical margin[119,131,138-140] and of intrahepatic recurrence[139,141]. Tranchart *et al*[142] found that R1 resection was an independent adverse predictor of OS and DFS, and the use of postoperative CHT was the only independent predictor of improved DFS in patients with R1 resection. The adverse effect of R1 LR on survival has been confirmed by other studies[138,143,144]. However also the protective effect of postoperative CHT after R1 resection has been recently confirmed[141,145].

The role of neoadjuvant CHT on the oncological outcome of R1 resection is controversial. Ayez *et al*[146] found that R1 resection remained an adverse prognostic factor in OS and DFS in patients receiving LR for CRLM not treated with neoadjuvant CHT, but not in those who had undergone neoadjuvant CHT. Different results were obtained in a study of 378 patients treated with neoadjuvant CHT and subsequent LR, where the effect of positive margins on OS was analysed in relation to response to CHT[147]. Fourteen percent of resections were R1 (tumour-free RM < 1 mm). The five-year overall survival rates were 55% for patients with R0 resection (tumour-free RM ⩾ 1 mm) and 26% for those with R1 resection (P = 0.017). R1 resection and a minor pathologic response to CHT at histology were independently correlated with worsened survival upon multivariate analysis. The survival advantage correlated with negative resection margins (R0 *vs* R1 LR) was higher in patients with suboptimal morphologic response at CT scans after CHT (five-year OS: 62% *vs* 11%; P = 0.007) than in those with optimal response (three-year OS: 92% *vs* 88%; P = 0.917), and higher in patients with a minor pathologic response at histologic evaluation (five-year OS: 46% *vs* 0%; P = 0.002) than in those with a major response (five-year OS: 63% *vs* 67%; P = 0.587). The authors concluded that with the current neoadjuvant CHT protocols, negative resection margins still represent a crucial prognostic factor and should remain the principal purpose of LR, and that the adverse influence of positive RM is most evident in the presence of suboptimal response to neoadjuvant CHT. In a similar study of 227 patients who received neoadjuvant oxaliplatin and/or irinotecan and 5-FU and subsequent curative LR[148], positive margins (tumour-free RM < 1 mm) significantly increased the risk of death without postoperative CHT (P = 0.0077), but not with postoperative CHT. Negative RM sizes of ⩾ 1– < 5, ⩾ 5– < 10, and ⩾ 10 mm were not significant predictors of OS. The authors concluded that patients undergoing LR for CRLM should receive postoperative CHT if negative margins cannot be achieved, and that negative margins wider than 1 mm do not improve OS for patients receiving neoadjuvant CHT. It should be noted however that when neoadjuvant CHT is interrupted, regardless of previous response, regrowth may occur at the periphery rather than in the centre of the metastasis, with clustering of viable cancer cells infiltrating the liver tissue for several millimetres at the periphery of the metastasis, irrespective of any signs of response in its centre, a phenomenon called "dangerous halo"[136]. Similarly, it has been found that neoadjuvant CHT may determine irregular borders of CRLM, particularly evident in lesions with significant contraction, and sometimes discrete islands of viable tumour cells outside of the main tumour, but all close to the peripheral margin of the tumour mass[149]. The possible progression of the dangerous halo is particularly worrying, and the planned surgical margin should be wide enough to limit the risk of local recurrence, especially if CHT has been interrupted for a relatively long time. It has been suggested that the argon-beam coagulation of the cut surface of the liver might reduce the risk of recurrence by providing a layer of necrosis of 2 mm to 5 mm[136].

Also submillimetric clear margins have been considered adequate for resection of CRLM. A total of 2368 patients undergoing LR for CRLM at Memorial Sloan Kettering Cancer Center between 1992 and 2012 were examined to evaluate the impact of margin width on OS[144]. The median OS of the R1, 0.1-0.9 mm, 1-9 mm, and ⩾ 10 mm groups was 32 mo, 40 mo, 53 mo, and 56 mo, respectively (P < 0.001). Compared with R1 LR, all RM widths, together with submillimetric margins, were associated with increased OS (P < 0.05). The significant association of RM width and OS remained when adjusted for all the other pathological and clinical factors of prognosis. The authors concluded that RM width is independently predictive of better survival rates, so that adequate margins should be obtained whenever possible. However, LR should be performed also in patients where narrow RM are anticipated because submillimetric margin clearance may improve

survival. The authors also suggested that the favourable outcome observed with submillimetric margins could be the expression of the biological behaviour of the tumour rather than the result of the surgical technique. Detachment of CRLM from intrahepatic vessels has been proposed as part of IOUS-guided PSLR[113,150]. Even though this kind of resection implies formally R1 resection margins, oncological outcomes seem to be similar to those described for R0 resections. In a recent series of 627 resection areas in 226 consecutive patients with CRLM, Viganò *et al*[151] compared the outcomes of R1 surgery (RM < 1 mm), distinguishing standard R1 resection and R1 resection with detachment of CLM from major intrahepatic vessels (R1 vascular). Five percent of recurrences at surgical RM occurred in 12.4% of patients. Local recurrence risk was similar between the R0 and R1 vascular groups but increased in the standard R1 resection group (P < 0.05 for both). Standard R1 resection had a higher rate of hepatic-only recurrences (P = 0.042) and was an independent negative prognostic factor of OS on multivariate analysis (P = 0.034). Conversely, R1 vascular resections had oncological outcomes similar to those of R0 resections suggesting that CRLM detachment from intrahepatic vessels can be safely pursued to increase resectability. Similar strategies of conservative IOUS-guided LR sparing intrahepatic vessels have been used in simultaneous colorectal and liver resection of advanced CRC with synchronous CRLM to limit the extension of LR with the aim of reducing the overall risk of the simultaneous procedures[91].

The data on whether R1 margin status is an independent predictor of survival have been conflicting because some authors have found that R1 margin status was not associated with survival after controlling for competing risk factors on multivariate analyses[138,139,141]. Tumour biology might play a determinant role in the impact of RM status on oncological outcome, where R1 resections could not have a prognostic value per se but reflect a more severe disease[38,40,129,138,141,145]. Recent changes in the prognostic value of R1 resections could reflect in part the beneficial effect of perioperative CHT[142,145-148]. In a recent series of 1784 hepatectomies analysed from a multicentric retrospective cohort of hepatectomies performed for CRLM in 32 French centres from January 2006 to December 2013[152], positive primary tumour lymph nodes at colorectal resection (P = 0.02), operative time > 240 minutes (P = 0.05), synchronous CRLM (P = 0.02), clamping of the hepatic pedicle > 40 min (P = 0.001), tumour size > 50 mm (P = 0.001), recurrent hepatectomy (P = 0.001), > 3 nodules (P=0.0001), and bilateral nodules (P = 0.0001) were recognized as risk factors for R1 resection upon multivariate analysis. After a propensity score matching according to Fong criteria, however, R1 resection still maintained an adverse impact on OS and DFS, with one-, three-, and five-year OS of 94%, 81%, and 70% in R0 LR *vs* 92%, 75%, and 58% in R1 LR, respectively (P = 0.008), and with one-, three-, and five-year DFS of 64%, 41%, and 28%in R0 LR versus 51%, 28%, and 18% in R1 LR, respectively (P = 0.0002).

R0 resection: the optimal free resection margin

Determining the optimal free RM in surgery of CRLM is much more controversial, since the traditional 1-cm rule to consider oncologically adequate the RM has been widely debated in the last decades. Pawlik *et al*[138] in 2005 demonstrated that OS, DFS, recurrence risk, and site of recurrence were not significantly different among patients undergoing resection of CRLM with RM of 1-4 mm, 5-9 mm, and ⩾ 10 mm, and suggested that predicted margin of < 1 cm after LR should not contraindicate LR. A similar study including 1019 patients from the Memorial Sloan Kettering Cancer Center showed that patients undergoing LR with RM > 10 mm had better survival than those with RM < 10 mm. However, within the latter group there was no significant difference in survival when stratified according to RM width, and patients with subcentimetric RM had an overall survival of 42 months (significantly better than similar patients treated with systemic CHT or ablative therapies)[153]. In another multicentric study of 2715 patients who received primary resection of CRLM, a 1-mm tumour-free RM was sufficient to obtain 33% five-year overall DFS, while extra RM width did not further increase DFS. After the propensity case-match analysis, the authors did not find a statistical difference in DFS between patients with negative narrow RM and wider RM clearance[143]. Recent meta-analyses however support the need of achieving adequate resection margins whenever possible. Dhir *et al*[154] examined 4821 patients with negative RM from 18 studies and found that the five-year OS for the ⩾ 1 cm negative RM subgroup was 46% when compared with 38% for < 1 cm negative RM subgroup (P = 0.009). In another meta-analysis based on 18 studies including 6790 patients[155], R1 resection had a negative impact on OS and DFS rates and was associated with more frequent recurrences. The use of current protocols of CHT did not alter the adverse oncological outcome of R1 resection. Notably, ⩾ 1 cm negative RM obtained the best overall survival rates. Margonis *et al*[156] evaluated 34 studies including a cohort of 11147 LR. Wider RM (> 1 cm *vs* < 1 cm) was significantly associated with improved OS and DFS at three years, five years, and ten years. Also > 1 mm *vs* < 1 mm RM was significantly associated with improved OS. Meta-regression analyses did not reveal any significant impact of perioperative CHT. The authors concluded that even though a > 1 mm RM determines better prognosis than a submillimetric RM, obtaining a RM > 1 cm may determine even better oncologic results and should be attempted whenever possible. Taken together, these data suggest that the 1-cm rule still has prognostic importance in the oncological outcome of resection of CRLM and should be pursued whenever possible. However, the likelihood of local and intrahepatic recurrences seem to be frequently independent

of margin width, where tumour biology seems to be a more decisive predictive factor of both intrahepatic recurrence and poorer long-term survival. Even though R1 resections should be avoided, the actual margin width of R0 resections seems to have a limited impact on the postoperative oncological outcome. For all these reasons, failure to comply with the 1-cm rule should no longer contraindicate liver resection of colorectal metastases.

Surgical strategies for multiple bilobar metastases

In 1984 Adson *et al*[157] reported a study of 141 patients who had resection of CRLM between 1948 and 1982 and found similar five-year survival rates between patients with single metastases and those with multiple lesions. They concluded that removal of multiple hepatic metastases was advisable in selected cases. This study was contradicted by Ekberg *et al*[108] in a series of 72 LR for CRLM between 1971 and 1984, where poor prognostic factors contraindicating surgical resection of CRLM included more than four lesions, impossibility to achieve a RM $\geqslant$ 1 cm, and evidence of extrahepatic disease. These data were confirmed by Hughes *et al*[158] in a series of 100 patients who survived for more than five years after resection, where patients with $\geqslant$ 4 metastases were considered to be contraindicated for LR. The considerable improvements achieved in the 1990s in the knowledge and treatment of colorectal metastases led to substantial changes in the surgical strategies for multiple CRLM[89]. In 1995 Scheele *et al*[159] reported their experience with 32 patients undergoing LR of $\geqslant$ 4 CRLM. According to their study, five or more independent metastases had an adverse effect on resectability. However, if a radical excision of all detectable disease could be obtained, the number of metastases (1-3 *vs* $\geqslant$ 4) was not significantly predictive of either OS or DFS. Subsequently Weber *et al*[160] reported a study of 155 patients who received LR for $\geqslant$ 4 CRLM with a five-year OS of 23%. As the number of tumours increased, the five-year survival rate diminished from 33% to 14%. However, in this study there were twelve five-year survivors, including two patients with nine or more nodules. Also the potential benefits of neoadjuvant CHT were delineated. Tanaka *et al*[161] reported 71 patients who had received LR for $\geqslant$ 5 bilobar CRLM and compared the outcome of 48 patients who received neoadjuvant CHT followed by LR with that of 23 patients treated by LR alone. Patients with neoadjuvant CHT experienced better three- and five-year survival rates from the time of diagnosis than those without CHT (67.0% and 38.9% *vs* 51.8% and 20.7% respectively; P = 0.039), and required fewer extended LR (four segments or more) (81.3% *vs* 100.0%; P = 0.027) Multivariate analysis demonstrated that neoadjuvant CHT independently predicted survival. The authors concluded that in patients with bilateral multiple CRLM, neoadjuvant CHT before LR was associated with improved survival.

For patients with extensive bilobar disease, multiple strategies combining TSH and neoadjuvant CHT were described by the surgeons from the Paul Brousse Hospital[162-164]. In selected patients with multiple CRLM not eligible for a curative one-stage resection, even when downstaged by CHT, after PVE, or combined with local ablation techniques, Adam *et al*[162] proposed a TSH strategy, where the highest possible number of nodules was resected in a first non-curative procedure, and the remaining tumours were resected after an adequate period of hepatic regeneration. The three-year survival rate of the 16 patients who completed the procedure was 35%, with four patients (31%) disease-free at 7 mo, 22 mo, 36 mo, and 54 mo. The same group subsequently examined a series of 33 patients with bilobar CRLM where a right or extended right LR was planned. The first-stage hepatectomy consisted of a clearance of tumours of the left FRL by resection or radiofrequency ablation (RFTA) to prevent the growth of metastatic nodules in the estimated FRL after PVE, followed by a right PVE to induce atrophy of the right hemiliver and hyperplasia of the left hemiliver. The second-stage hepatectomy, a right or extended right hepatectomy, was performed in patients with adequate left FRL hyperplasia and without disease progression. The one- and three-year survival rates were 70.0% and 54.4%, respectively, in the 25 patients in whom the procedure was completed[164].

In all these Western studies, patients with multiple CRLM were candidates for major or extended hepatectomies in most cases. In the same period the surgeons from the Cancer Institute and the University of Tokyo were following a different approach to multiple CRLM[35,89,130]. Kokudo *et al*[89] reported a series of 183 patients who received LR with curative intent for CRLM from 1980 to 2000 with five-year OS of 41.9%. The overall outcome of 21 patients who had $\geqslant$ 4 tumours in the liver was not significantly different from that of patients with $\leqslant$ 3 tumours. In the same study the authors delineated the principles of conservative LR strategy for multiple CRLM: Accurate preoperative evaluation of the tumour number and their proximity to the major intrahepatic vasculature, careful intraoperative inspection and palpation of the liver and use of IOUS, multiple partial resections whenever possible instead of extended hepatectomies, with resection of large intrahepatic vessels only if tumour invasion was present, non-anatomical resection even with a minimum surgical margin, and preoperative PVE when the estimated volume of the remnant liver was under 40% in case of major hepatectomy. In the overall series the remnant liver was the most common site of recurrence, and repeated liver resection was carried out in approximately half of the patients after recurrence, with a five-year survival rate of 44.7% starting from the first hepatectomy. With these diagnostic and therapeutic strategies the same group performed over 1000 hepatectomies without mortality[35]. A similar approach to multiple bilobar CRLM was reported by Torzilli *et al*[165] in a series of 29 patients

with multiple (≥ 4) bilobar CRLM where the surgical strategy was based on tumour-vessel relationships at IOUS and on findings at colour-Doppler IOUS. Tumour removal was feasible in 89.7% of patients. There was no in-hospital mortality and the overall morbidity rate was 23%. After a median follow-up of 14 mo (range 6-54), three patients had died from systemic recurrence, twelve were alive without disease, and eleven were alive with recurrence. However, no local relapses were observed at the surgical RM. The authors concluded that IOUS-guided resection based on strict criteria allows one-stage LR in selected patients with multiple bilobar CRLM, and thus decreasing the need for a TSH.

In the past decade, ablative techniques, including RFTA and microwave ablation (MWA), have emerged as an appealing option for the local treatment of primary and metastatic liver tumours, including CRLM, alone or in combination with LR. The role of ablation in patients with CRLM is unclear since ablative techniques have usually shown significantly lower rates of complications, but also lower survival rates and higher rates of recurrence as compared to LR[166-168], even though RFTA might have a role equivalent to liver surgery in the treatment of small (≤ 2 cm) CRLM[167]. Recent studies have shown that LR combined with intraoperative ablation techniques is effective in the treatment of multiple bilateral CRLM, with adequate perioperative outcomes and without compromising overall oncological results compared with bilateral resection or with TSH. It may represent an excellent option to pursue effective parenchymal-sparing treatments for extensive CRLM[169-172].

A progressive shift toward more conservative hepatectomies for bilobar CRLM has been reported also by surgeons traditionally inclined to more extensive LR. In a series of 443 LR in 440 patients who received resection of bilateral CRLM at the Memorial Sloan-Kettering Cancer Center[45], a major hepatectomy including three segments, hemihepatectomy or more extended resection in most cases, was performed as part of 380 operations. Major complications were 29% and 90-day mortality was 5.4%. Estimated five-year disease-specific and recurrence-free survivals were 30% and 18%, respectively. However, the surgical technique changed over time toward parenchymal-sparing techniques based on the wider use of multiple simultaneous liver resections, wedge resections, and local ablations, which correlated with decreased mortality rates without changes in disease-specific survival or liver recurrence. The authors concluded that resection of bilateral CRLM can be achieved with reasonable morbidity, mortality, and oncologic results, and that increased use of parenchymal-sparing approaches is associated with decreased mortality without compromising oncological outcomes. The favourable results of PSLR have been recently confirmed in a multicentric retrospective series of patients who had received LR for multiple (> 3) bilobar CRLM, comparing 331 patients who had received PSLR with 360 who had received non-PSLR, defined as the resection of three or more consecutive liver segments, excluding TSH[46]. PSLR was associated with lower complications (25% *vs* 34%; *P* = 0.04) and fewer Dindo-Clavien grade Ⅲ and Ⅳ complications (10% *vs* 16%; *P* = 0.03). Liver failure was less frequent after PSLR (2% *vs* 7%; *P* = 0.006), with a shorter ICU stay (0 days *vs* 1 day, *P* = 0.004). OS and DFS were similar between the two groups. The authors concluded that PSLR for multiple bilobar CRLM represents an appropriate alternative to non-PSLR in selected patients, with lower morbidity and comparable oncological outcomes. Recent studies have further demonstrated the positive impact of PSLR in the treatment of multiple bilobar CRLM, bringing into question also the consolidated role of the TSH in these cases[47]. A bi-institutional study compared the outcome of patients with multiple bilobar CRLM who had received TSH or PSLR. The inclusion criteria were ≥ 6 CRLM, ≥ 3 CRLM in the left liver, and ≥ 1 lesion with vascular contact. A total of 74 TSH and 35 PSLR were compared. Drop-out rate of TSH was 40.5%. PSLR had significantly lower blood loss, overall morbidity, severe morbidity, and liver-specific morbidity than TSH. R0 resection rate was similar between groups. PSLR and completed TSH had similar five-year OS (38.2% *vs* 31.8%), three-year RFS (17.6% *vs* 17.7%), and recurrence sites. The authors concluded that parenchymal-sparing hepatectomies are a safe alternative to TSH for multiple, bilobar, deeply located CRLM, and that PSLR should be preferred whenever achievable because of better safety and oncological results comparable to completed TSH without the drop-out risk.

Recent reports have demonstrated that also patients with large numbers of CRLM are potential candidates for liver surgery. In a bi-institutional Japanese study of 736 patients who underwent LR for CRLM over a 16-year period[173], the authors compared 493 patients with 1-3 tumours, 141 with 4-7 tumours, and 102 with ≥ 8 tumours. Major hepatectomies had been performed in a minority of patients (21.6%). The five-year OS and DFS rates were 51% and 21%, respectively, for the entire patient cohort, 56% and 29% for patients with 1-3 tumours, 41% and 12% for those with 4-7 tumours, and 33% and 1.7% for those with ≥ 8 tumours. Positive lymph node metastasis of the primary CRC, the presence of extrahepatic metastases, a maximum tumour size > 5 cm, and tumour exposure during LR were associated with decreased survival upon multivariate analysis. The authors concluded that in patients with multiple CRLM, the number of CRLM has less prognostic impact than other factors, and that complete LR may offer a chance of cure even in patients with numerous CRLM, including those with eight or more nodules. In another bi-institutional study of 849 patients undergoing LR for CRLM[174], 743 patients with 1-7 metastases were compared to 106 with ≥ 8 metastases. The overall perioperative mortality rate was 0.4%. Patients with 1-7 metastases had higher five-year OS (44.2% *vs* 20.1%; *P* < 0.001) and DFS (28.7% *vs* 13.6%; *P* < 0.001) rates. In patients with ≥ 8 metastases, OS and

DFS were similar for patients with 8-10, 11–15, or > 15 metastases. In this group, multivariate analysis identified three preoperative factors of adverse prognosis, including extrahepatic disease (P = 0.010), no response to preoperative CHT (P = 0.023), and primary rectal cancer (P = 0.039). Patients with two or more risk factors had very poor outcomes, while those with no risk factors had survival rates similar to patients with 1-7 metastases (five-year OS rate 44.0% *vs* 44.2%). The authors concluded that LR is safe in selected patients with ⩾ 8 metastases, and offers reasonable five-year survival independent of the number of metastases. A recent French multicentric study examined the outcome of 529 patients undergoing liver surgery for ⩾ 10 CRLM from 2005 to 2013, prospectively collected in the LiverMetSurvey registry[92]. The five-year OS was 30%. A macroscopically complete (R0/R1) resection was achieved in 72.8% of patients and was associated with a three- and five-year OS of 61% and 39%, compared to 29% and 5% for R2/no resection patients (P < 0.0001). Upon multivariate analysis, R0/R1 resection resulted as the strongest favourable factor of OS (P < 0.0001). Other independent favourable factors were maximal tumour size < 40 mm (P = 0.02), age < 60 years (P = 0.005), preoperative MRI (P = 0.007), and adjuvant CHT (P = 0.04). Of the 346 patients who underwent R0/R1 resection, 74.6% had developed a recurrence at last follow-up, with three- and five-year primary DFS rates of 23% and 7%, respectively. When hepatic recurrence and extrahepatic recurrence were surgically treated, the secondary DFS rates (taking into account the impact of repeat surgery) at three years and five years were 42% and 31%, respectively. The authors concluded that, even though the oncological outcome of patients with ⩾ 10 CRLM is obviously worse compared to patients exhibiting fewer lesions, surgery remains the only hope of prolonged survival, especially if complete resection can be performed, and that the number of CRLM should not be considered per se as contraindication to surgery.

The impact of PSLR on simultaneous colorectal and liver surgery

Simultaneous colorectal resection and minor hepatectomy have perioperative results similar to minor hepatectomy alone, and are at present considered the treatment of choice in most patients with limited liver disease suitable for minor LR[1,19,24]. The results are much more conflicting in patients requiring simultaneous colorectal and major LR because most investigators have reported worse perioperative outcomes than for major LR alone also in experienced hepatobiliary centres[20,79,80,82,175], while others remark that simultaneous colorectal resection and major hepatectomy can be performed safely in selected cases with perioperative risks comparable to major LR alone[31-33]. Most studies comparing simultaneous and staged procedures are retrospective, with patients undergoing simultaneous procedures having more limited hepatic involvement, which could explain these discordant outcomes[19,22,26]. At present, most authors suggest combined resections in the case of easily accessible, uncomplicated colorectal tumours with CRLM requiring minor hepatectomies[26,27,176], while these criteria could be selectively extended in units experienced in both hepatobiliary and colorectal surgery[23]. In a recent survey reporting the opinion of colorectal and liver surgeons about simultaneous resection of CRC and liver metastases[177], most surgeons of both groups perceived that simultaneous procedures were appropriate in adequately selected patients, especially in candidates to any type of colorectal surgery with minor LR. Restorative rectal resections coupled with a major LR were considered inappropriate due to the risk of leakage of the colorectal anastomosis. Some concern did exist as well, especially among liver surgeons, about the risk of leakage also for colo-colic anastomoses if combined with major LR.

As a matter of fact, even though surgeons experienced in colorectal and hepatobiliary surgery should carefully select candidates to simultaneous resection to minimize perioperative complications, the planned extent of LR seem to represent the most important determinant of whether simultaneous procedures are individually appropriate for CRC with synchronous CRLM[19,24,178,179]. As previously discussed, IOUS-based conservative techniques of liver surgery substantially decrease the need for major hepatectomies also for multiple bilobar CRLM, with a substantial reduction of perioperative related risks and may represent an appropriate solution even for potential candidates to simultaneous colorectal and liver resection for bilobar synchronous CRLM. In a small retrospective series of 39 consecutive patients with synchronous CRLM, who underwent curative simultaneous "one-stage" hepatectomy and resection of the colorectal primary, Tanaka *et al*[178] observed that only the volume of the resected liver was a significant risk factor for postoperative complications (350 g mean resected liver volume in patients with postoperative complications *vs* 150 g in those without complications; P < 0.05). The systematic application of the criteria of conservative liver surgery have been associated with higher rates of feasibility of simultaneous colorectal and liver resections also in patients with multiple hepatic nodules. Minagawa *et al*[180] in 2006 reported 148 patients admitted with CRC and synchronous CRLM since January 1989, evaluated for simultaneous resection regardless of the location of the primary cancer and the extent of CRLM. A simultaneous resection was performed in 142 cases (feasibility rate 96%), without perioperative mortality. Fifty-one percent of patients had the primary tumour located in the rectum. With the systematic application of their principles of conservative IOUS-based liver surgery[89], only 11.3% of patients required a hemihepatectomy, while the others received limited resections (74.6%) or the resection of one or two segments (14.1%). In a more recent study of 150 patients who underwent resection of primary CRC

and synchronous CRLM between 1993 and 2011[181], the proportion of simultaneous resections was 84.7%. Among the 127 patients who had received a simultaneous colorectal and hepatic resection, there was no postoperative mortality, postoperative complications were 61.4%, major complications were 18.2%, and anastomotic failure occurred in 1.6% of patients. The three-, five- and ten-year OS was 74%, 64%, and 52%, respectively. In a small series of 45 patients who underwent elective resection of primary CRC and synchronous CRLM, a simultaneous colorectal resection with anastomosis and conservative one-stage LR was feasible in 75.6% of patients. It was possible to avoid a right hepatectomy in all the patients undergoing simultaneous restorative colorectal resection[91]. Seven patients had synchronous CRC at presentation (unpublished data), and two of them had rectal cancer within diffuse colorectal poliposis and received restorative proctocolectomy with ileoanal J-pouch and temporary diverting loop ileostomy. One patient with multiple CRLM of the right hemiliver underwent the restorative proctocolectomy after neoadjuvant CHT, with a subsequent resection of liver segments S6–S7–S8. The other had a single metastasis in segment S8 and underwent simultaneous restorative proctocolectomy and liver segmentectomy. Two patients had a simultaneous cancer proximal to a rectal cancer, with multiple bilobar CRLM. One received neoadjuvant chemoradiotherapy and subsequent resection of the sigmoid colon and of the rectum with simultaneous one-stage PSLR. In the other patient a TSH was planned to treat the hepatic disease. The patient received neoadjuvant chemoradiotherapy and a subsequent rectal resection with a first-stage LR consisting of multiple wedge resection in the left hemiliver with right portal vein ligation. At re-exploration for the second-stage LR a massive diffusion of the cancer at the hepatic hilum was found and the planned right hepatectomy was not performed. Finally, three patients had SCRC in distant colonic segments, and we opted for a restorative subtotal colectomy. One patient underwent simultaneous liver bisegmentectomy of S2-S3 with splenectomy and interaortocaval lymphadenectomy because of splenic and interaortocaval lymph node metastases. The other two underwent PSLR for multiple bilobar CRLM, associated with intraoperative RFTA in one patient. Therefore, five patients received simultaneous potentially curative colorectal and one-stage liver resection without postoperative mortality and complications requiring reoperation.

CONCLUSION

In conclusion, simultaneous procedures represent an attractive surgical option in selected patients with resectable CRC and resectable synchronous CRLM. Simultaneous resections should only be considered by surgical teams experienced in both fields. Staged procedures are still advisable in the case of complicated CRC requiring urgent colorectal resection. In all other cases, simultaneous resections should be theoretically considered whenever possible, including patients with SCRC. In these cases, if the synchronous tumours are located in distant colorectal segments, an extended restorative colectomy should be considered to prevent the risks related to multiple colorectal anastomoses, especially if prolonged hepatic pedicle clamping is planned for extensive PSLR and/or CRLM adjacent to major intrahepatic vessels. When rectal cancer is diagnosed, the indication to preoperative chemoradiotherapy and its potential benefits should be adequately considered. A systematic approach to liver resection that focuses on the need of reducing the extent of hepatectomy while preserving oncological radicality may represent the best strategy to limit the perioperative risks in candidates to simultaneous colorectal and liver resection.

REFERENCES

1 **Brown RE**, Bower MR, Martin RC. Hepatic resection for colorectal liver metastases. *Surg Clin North Am* 2010; **90**: 839-852 [PMID: 20637951 DOI: 10.1016/j.suc.2010.04.012]

2 **Siriwardena AK**, Mason JM, Mullamitha S, Hancock HC, Jegatheeswaran S. Management of colorectal cancer presenting with synchronous liver metastases. *Nat Rev Clin Oncol* 2014; **11**: 446-459 [PMID: 24889770 DOI: 10.1038/nrclinonc.2014.90]

3 **Lam AK**, Chan SS, Leung M. Synchronous colorectal cancer: clinical, pathological and molecular implications. *World J Gastroenterol* 2014; **20**: 6815-6820 [PMID: 24944471 DOI: 10.3748/wjg.v20.i22.6815]

4 **Pajares JA**, Perea J. Multiple primary colorectal cancer: Individual or familial predisposition? *World J Gastrointest Oncol* 2015; **7**: 434-444 [PMID: 26688706 DOI: 10.4251/wjgo.v7.i12.434]

5 **Yang J**, Peng JY, Chen W. Synchronous colorectal cancers: a review of clinical features, diagnosis, treatment, and prognosis. *Dig Surg* 2011; **28**: 379-385 [PMID: 22156665 DOI: 10.1159/000334073]

6 **van Leersum NJ**, Aalbers AG, Snijders HS, Henneman D, Wouters MW, Tollenaar RA, Eddes EH. Synchronous colorectal carcinoma: a risk factor in colorectal cancer surgery. *Dis Colon Rectum* 2014; **57**: 460-466 [PMID: 24608302 DOI: 10.1097/DCR.0000000000000068]

7 **Cheng J**, Liu X, Shuai X, Deng M, Gao J, Tao K. Synchronous triple colorectal carcinoma: a case report and review of literature. *Int J Clin Exp Pathol* 2015; **8**: 9706-9711 [PMID: 26464742]

8 **Lam AK**, Carmichael R, Gertraud Buettner P, Gopalan V, Ho YH, Siu S. Clinicopathological significance of synchronous carcinoma in colorectal cancer. *Am J Surg* 2011; **202**: 39-44 [PMID: 21600553 DOI: 10.1016/j.amjsurg.2010.05.012]

9 **Hu H**, Chang DT, Nikiforova MN, Kuan SF, Pai RK. Clinicopathologic features of synchronous colorectal carcinoma: A distinct subset arising from multiple sessile serrated adenomas and associated with high levels of microsatellite instability and favorable prognosis. *Am J Surg Pathol* 2013; **37**: 1660-1670 [PMID: 23887157 DOI: 10.1097/PAS.0b013e31829623b8]

10 **Mulder SA**, Kranse R, Damhuis RA, de Wilt JH, Ouwendijk RJ, Kuipers EJ, van Leerdam ME. Prevalence and prognosis of synchronous colorectal cancer: a Dutch population-based study. *Cancer Epidemiol* 2011; **35**: 442-447 [PMID: 21470938 DOI: 10.1016/j.canep.2010.12.007]

11 **Latournerie M**, Jooste V, Cottet V, Lepage C, Faivre J, Bouvier AM. Epidemiology and prognosis of synchronous colorectal cancers. *Br J Surg* 2008; **95**: 1528-1533 [PMID: 18991301 DOI: 10.1002/bjs.6382]

12 **Passman MA**, Pommier RF, Vetto JT. Synchronous colon primaries have the same prognosis as solitary colon cancers. *Dis Colon Rectum* 1996; **39**: 329-334 [PMID: 8603557 DOI: 10.1007/BF02049477]

13 **Adloff M**, Arnaud JP, Bergamaschi R, Schloegel M. Synchronous carcinoma of the colon and rectum: prognostic and therapeutic implications. *Am J Surg* 1989; **157**: 299-302 [PMID: 2537586 DOI: 10.1016/0002-9610(89)90555-2]

14 **Wang HZ**, Huang XF, Wang Y, Ji JF, Gu J. Clinical features, diagnosis, treatment and prognosis of multiple primary colorectal carcinoma. *World J Gastroenterol* 2004; **10**: 2136-2139 [PMID: 15237453 DOI: 10.3748/wjg.v10.i14.2136]

15 **Easson AM**, Cotterchio M, Crosby JA, Sutherland H, Dale D, Aronson M, Holowaty E, Gallinger S. A population-based study of the extent of surgical resection of potentially curable colon cancer. *Ann Surg Oncol* 2002; **9**: 380-387 [PMID: 11986190 DOI: 10.1007/BF02573873]

16 **Yeh CC**, Hsi SC, Chuu CP, Kao YH. Synchronous triple carcinoma of the colon and rectum. *World J Surg Oncol* 2013; **11**: 66 [PMID: 23497155 DOI: 10.1186/1477-7819-11-66]

17 **Holubar SD**, Wolff BG, Poola VP, Soop M. Multiple synchronous colonic anastomoses: are they safe? *Colorectal Dis* 2010; **12**: 135-140 [PMID: 19207709 DOI: 10.1111/j.1463-1318.2009.01771.x]

18 **Li Z**, Wang D, Wei Y, Liu P, Xu J. Clinical outcomes of laparoscopic-assisted synchronous bowel anastomoses for synchronous colorectal cancer: initial clinical experience. *Oncotarget* 2017; **8**: 10741-10747 [PMID: 27821798 DOI: 10.18632/oncotarget.12899]

19 **Reddy SK**, Barbas AS, Clary BM. Synchronous colorectal liver metastases: is it time to reconsider traditional paradigms of management? *Ann Surg Oncol* 2009; **16**: 2395-2410 [PMID: 19506963 DOI: 10.1245/s10434-009-0372-1]

20 **Reddy SK**, Pawlik TM, Zorzi D, Gleisner AL, Ribero D, Assumpcao L, Barbas AS, Abdalla EK, Choti MA, Vauthey JN, Ludwig KA, Mantyh CR, Morse MA, Clary BM. Simultaneous resections of colorectal cancer and synchronous liver metastases: a multi-institutional analysis. *Ann Surg Oncol* 2007; **14**: 3481-3491 [PMID: 17805933 DOI: 10.1245/s10434-007-9522-5]

21 **Schmoll HJ**, Van Cutsem E, Stein A, Valentini V, Glimelius B, Haustermans K, Nordlinger B, van de Velde CJ, Balmana J, Regula J, Nagtegaal ID, Beets-Tan RG, Arnold D, Ciardiello F, Hoff P, Kerr D, Köhne CH, Labianca R, Price T, Scheithauer W, Sobrero A, Tabernero J, Aderka D, Barroso S, Bodoky G, Douillard JY, El Ghazaly H, Gallardo J, Garin A, Glynne-Jones R, Jordan K, Meshcheryakov A, Papamichail D, Pfeiffer P, Souglakos I, Turhal S, Cervantes A. ESMO Consensus Guidelines for management of patients with colon and rectal cancer. a personalized approach to clinical decision making. *Ann Oncol* 2012; **23**: 2479-2516 [PMID: 23012255 DOI: 10.1093/annonc/mds236]

22 **Mentha G**, Majno P, Terraz S, Rubbia-Brandt L, Gervaz P, Andres A, Allal AS, Morel P, Roth AD. Treatment strategies for the management of advanced colorectal liver metastases detected synchronously with the primary tumour. *Eur J Surg Oncol* 2007; **33** Suppl 2: S76-S83 [PMID: 18006267 DOI: 10.1016/j.ejso.2007.09.016]

23 **Hillingsø JG**, Wille-Jørgensen P. Staged or simultaneous resection of synchronous liver metastases from colorectal cancer--a systematic review. *Colorectal Dis* 2009; **11**: 3-10 [PMID: 18637099 DOI: 10.1111/j.1463-1318.2008.01625.x]

24 **Chen J**, Li Q, Wang C, Zhu H, Shi Y, Zhao G. Simultaneous vs. staged resection for synchronous colorectal liver metastases: a metaanalysis. *Int J Colorectal Dis* 2011; **26**: 191-199 [PMID: 20669024 DOI: 10.1007/s00384-010-1018-2]

25 **Li ZQ**, Liu K, Duan JC, Li Z, Su CQ, Yang JH. Meta-analysis of simultaneous versus staged resection for synchronous colorectal liver metastases. *Hepatol Res* 2013; **43**: 72-83 [PMID: 22971038 DOI: 10.1111/j.1872-034X.2012.01050.x]

26 **Yin Z**, Liu C, Chen Y, Bai Y, Shang C, Yin R, Yin D, Wang J. Timing of hepatectomy in resectable synchronous colorectal liver metastases (SCRLM): Simultaneous or delayed? *Hepatology* 2013; **57**: 2346-2357 [PMID: 23359206 DOI: 10.1002/hep.26283]

27 **Slesser AA**, Simillis C, Goldin R, Brown G, Mudan S, Tekkis PP. A meta-analysis comparing simultaneous versus delayed resections in patients with synchronous colorectal liver metastases. *Surg Oncol* 2013; **22**: 36-47 [PMID: 23253399 DOI: 10.1016/j.suronc.2012.11.002]

28 **Feng Q**, Wei Y, Zhu D, Ye L, Lin Q, Li W, Qin X, Lyu M, Xu J. Timing of hepatectomy for resectable synchronous colorectal liver metastases: for whom simultaneous resection is more suitable--a meta-analysis. *PLoS One* 2014; **9**: e104348 [PMID: 25093337 DOI: 10.1371/journal.pone.0104348]

29 **Gavriilidis P**, Sutcliffe RP, Hodson J, Marudanayagam R, Isaac J, Azoulay D, Roberts KJ. Simultaneous versus delayed hepatectomy for synchronous colorectal liver metastases: a systematic review and meta-analysis. *HPB* (Oxford) 2018; **20**: 11-19 [PMID: 28888775 DOI: 10.1016/j.hpb.2017.08.008]

30 **Veereman G**, Robays J, Verleye L, Leroy R, Rolfo C, Van Cutsem E, Bielen D, Ceelen W, Danse E, De Man M, Demetter P, Flamen P, Hendlisz A, Sinapi I, Vanbeckevoort D, Ysebaert D, Peeters M. Pooled analysis of the surgical treatment for colorectal cancer liver metastases. *Crit Rev Oncol Hematol* 2015; **94**: 122-135 [PMID: 25666309 DOI: 10.1016/j.critrevonc.2014.12.004]

31 **Capussotti L**, Ferrero A, Viganò L, Ribero D, Lo Tesoriere R, Polastri R. Major liver resections synchronous with colorectal surgery. *Ann Surg Oncol* 2007; **14**: 195-201 [PMID: 17080238 DOI: 10.1245/s10434-006-9055-3]

32 **Martin RC 2nd**, Augenstein V, Reuter NP, Scoggins CR, McMasters KM. Simultaneous versus staged resection for synchronous colorectal cancer liver metastases. *J Am Coll Surg* 2009; **208**: 842-850; discussion 850-852 [PMID: 19476847 DOI: 10.1016/j.jamcollsurg.2009.01.031]

33 **Muangkaew P**, Cho JY, Han HS, Yoon YS, Choi Y, Jang JY, Choi H, Jang JS, Kwon SU. Outcomes of Simultaneous Major Liver Resection and Colorectal Surgery for Colorectal Liver Metastases. *J Gastrointest Surg* 2016; **20**: 554-563 [PMID: 26471363 DOI: 10.1007/s11605-015-2979-9]

34 **Silberhumer GR**, Paty PB, Temple LK, Araujo RL, Denton B, Gonen M, Nash GM, Allen PJ, DeMatteo RP, Guillem J, Weiser MR, D'Angelica MI, Jarnagin WR, Wong DW, Fong Y. Simultaneous resection for rectal cancer with synchronous liver metastasis is a safe procedure. *Am J Surg* 2015; **209**: 935-942 [PMID: 25601556 DOI: 10.1016/j.amjsurg.2014.09.024]

35 **Imamura H**, Seyama Y, Kokudo N, Maema A, Sugawara Y, Sano K, Takayama T, Makuuchi M. One thousand fifty-six hepatectomies without mortality in 8 years. *Arch Surg* 2003; **138**: 1198-1206; discussion 1206 [PMID: 14609867 DOI: 10.1001/archsurg.138.11.1198]

36 **Rahbari NN**, Garden OJ, Padbury R, Brooke-Smith M, Crawford M, Adam R, Koch M, Makuuchi M, Dematteo RP, Christophi C, Banting S, Usatoff V, Nagino M, Maddern G, Hugh TJ, Vauthey JN, Greig P, Rees M, Yokoyama Y, Fan ST, Nimura Y, Figueras J, Capussotti L, Büchler MW, Weitz J. Posthepatectomy liver failure: a definition and grading by the International Study Group of Liver Surgery (ISGLS). *Surgery* 2011; **149**: 713-724 [PMID: 21236455 DOI: 10.1016/j.surg.2010.10.001]

37 **Ikoma N**, Raghav K, Chang G. An Update on Randomized Clinical Trials in Metastatic Colorectal Carcinoma. *Surg Oncol Clin N Am* 2017; **26**: 667-687 [PMID: 28923224 DOI: 10.1016/j.soc.2017.05.007]

38 **Poultsides GA**, Schulick RD, Pawlik TM. Hepatic resection for colorectal metastases: the impact of surgical margin status on outcome. *HPB* (Oxford) 2010; **12**: 43-49 [PMID: 20495644 DOI: 10.1111/j.1477-2574.2009.00121.x]

39 **Yang C**, Rahbari NN, Mees ST, Schaab F, Koch M, Weitz J, Reissfelder C. Staged resection of bilobar colorectal liver metastases: surgical strategies. *Langenbecks Arch Surg* 2015; **400**: 633-640 [PMID: 26049744 DOI: 10.1007/s00423-015-1310-2]

40 **Alvarez FA**, Sanchez Claria R, Oggero S, de Santibañes E. Parenchymal-sparing liver surgery in patients with colorectal carcinoma liver metastases. *World J Gastrointest Surg* 2016; **8**: 407-423 [PMID: 27358673 DOI: 10.4240/wjgs.v8.i6.407]

41 **Moris D**, Dimitroulis D, Vernadakis S, Papalampros A, Spartalis E, Petrou A, Pawlik TM, Felekouras E. Parenchymal-sparing Hepatectomy as the New Doctrine in the Treatment of Liver-

metastatic Colorectal Disease: Beyond Oncological Outcomes. *Anticancer Res* 2017; **37**: 9-14 [PMID: 28011468 DOI: 10.21873/anticanres.11283]

42 **Oba M**, Hasegawa K, Shindoh J, Yamashita S, Sakamoto Y, Makuuchi M, Kokudo N. Survival benefit of repeat resection of successive recurrences after the initial hepatic resection for colorectal liver metastases. *Surgery* 2016; **159**: 632-640 [PMID: 26477476 DOI: 10.1016/j.surg.2015.09.003]

43 **Kulik U**, Bektas H, Klempnauer J, Lehner F. Repeat liver resection for colorectal metastases. *Br J Surg* 2013; **100**: 926-932 [PMID: 23640669 DOI: 10.1002/bjs.9132]

44 **Wicherts DA**, de Haas RJ, Salloum C, Andreani P, Pascal G, Sotirov D, Adam R, Castaing D, Azoulay D. Repeat hepatectomy for recurrent colorectal metastases. *Br J Surg* 2013; **100**: 808-818 [PMID: 23494765 DOI: 10.1002/bjs.9088]

45 **Gold JS**, Are C, Kornprat P, Jarnagin WR, Gönen M, Fong Y, DeMatteo RP, Blumgart LH, D'Angelica M. Increased use of parenchymal-sparing surgery for bilateral liver metastases from colorectal cancer is associated with improved mortality without change in oncologic outcome: trends in treatment over time in 440 patients. *Ann Surg* 2008; **247**: 109-117 [PMID: 18156930 DOI: 10.1097/SLA.0b013e3181557e47]

46 **Memeo R**, de Blasi V, Adam R, Goéré D, Azoulay D, Ayav A, Gregoire E, Kianmanesh R, Navarro F, Sa Cunha A, Pessaux P; French Colorectal Liver Metastases Working Group, Association Française de Chirurgie (AFC). Parenchymal-sparing hepatectomies (PSH) for bilobar colorectal liver metastases are associated with a lower morbidity and similar oncological results: a propensity score matching analysis. *HPB* (Oxford) 2016; **18**: 781-790 [PMID: 27593596 DOI: 10.1016/j.hpb.2016.06.004]

47 **Torzilli G**, Viganò L, Cimino M, Imai K, Vibert E, Donadon M, Mansour D, Castaing D, Adam R. Is Enhanced One-Stage Hepatectomy a Safe and Feasible Alternative to the Two-Stage Hepatectomy in the Setting of Multiple Bilobar Colorectal Liver Metastases? A Comparative Analysis between Two Pioneering Centers. *Dig Surg* 2018; **35**: 323-332 [PMID: 29439275 DOI: 10.1159/000486210]

48 **Govaert KM**, Jongen JMJ, Kranenburg O, Borel Rinkes IHM. Surgery-induced tumor growth in (metastatic) colorectal cancer. *Surg Oncol* 2017; **26**: 535-543 [PMID: 29113675 DOI: 10.1016/j.suronc.2017.10.004]

49 **Lim C**, Cauchy F, Azoulay D, Farges O, Ronot M, Pocard M. Tumour progression and liver regeneration--insights from animal models. *Nat Rev Gastroenterol Hepatol* 2013; **10**: 452-462 [PMID: 23567217 DOI: 10.1038/nrgastro.2013.55]

50 **Shi JH**, Line PD. Effect of liver regeneration on malignant hepatic tumors. *World J Gastroenterol* 2014; **20**: 16167-16177 [PMID: 25473170 DOI: 10.3748/wjg.v20.i43.16167]

51 **Rupertus K**, Kollmar O, Scheuer C, Junker B, Menger MD, Schilling MK. Major but not minor hepatectomy accelerates engraftment of extrahepatic tumor cells. *Clin Exp Metastasis* 2007; **24**: 39-48 [PMID: 17260102 DOI: 10.1007/s10585-006-9054-6]

52 **Krause P**, Flikweert H, Monin M, Seif Amir Hosseini A, Helms G, Cantanhede G, Ghadimi BM, Koenig S. Increased growth of colorectal liver metastasis following partial hepatectomy. *Clin Exp Metastasis* 2013; **30**: 681-693 [PMID: 23385555 DOI: 10.1007/s10585-013-9572-y]

53 **de Graaf W**, van den Esschert JW, van Lienden KP, van Gulik TM. Induction of tumor growth after preoperative portal vein embolization: is it a real problem? *Ann Surg Oncol* 2009; **16**: 423-430 [PMID: 19050974 DOI: 10.1245/s10434-008-0222-6]

54 **Hoekstra LT**, van Lienden KP, Doets A, Busch OR, Gouma DJ, van Gulik TM. Tumor progression after preoperative portal vein embolization. *Ann Surg* 2012; **256**: 812-817; discussion 817-818 [PMID: 23095626 DOI: 10.1097/SLA.0b013e3182733f09]

55 **Al-Sharif E**, Simoneau E, Hassanain M. Portal vein embolization effect on colorectal cancer liver metastasis progression: Lessons learned. *World J Clin Oncol* 2015; **6**: 142-146 [PMID: 26468450 DOI: 10.5306/wjco.v6.i5.142]

56 **Kaibara N**, Koga S, Jinnai D. Synchronous and metachronous malignancies of the colon and rectum in Japan with special reference to a coexisting early cancer. *Cancer* 1984; **54**: 1870-1874 [PMID: 6478423 DOI: 10.1002/1097-0142(19841101)54:9<1870::AID-CNCR2820540917>3.0.CO;2-5]

57 **Greenstein AJ**, Barth JA, Sachar DB, Aufses AH Jr. Free colonic perforation without dilatation in ulcerative colitis. *Am J Surg* 1986; **152**: 272-275 [PMID: 3752375 DOI: 10.1016/0002-9610(86)90256-4]

58 **Liu X**, Goldblum JR, Zhao Z, Landau M, Heald B, Pai R, Lin J. Distinct clinicohistologic features of inflammatory bowel disease-associated colorectal adenocarcinoma: in comparison with sporadic microsatellite-stable and Lynch syndrome-related colorectal adenocarcinoma. *Am J Surg Pathol* 2012; **36**: 1228-1233 [PMID: 22790862 DOI: 10.1097/PAS.0b013e318253645a]

59 **Kiran RP**, Khoury W, Church JM, Lavery IC, Fazio VW, Remzi FH. Colorectal cancer complicating inflammatory bowel disease: similarities and differences between Crohn's and ulcerative colitis based on three decades of experience. *Ann Surg* 2010; **252**: 330-335 [PMID: 20622662 DOI: 10.1097/SLA.0b013e3181e61e69]

60 **Mohammadi M**, Kristensen MH, Nielsen HJ, Bonde JH, Holck S. Qualities of sessile serrated adenoma/polyp/lesion and its borderline variant in the context of synchronous colorectal carcinoma. *J Clin Pathol* 2012; **65**: 924-927 [PMID: 22782936 DOI: 10.1136/jclinpath-2012-200803]

61 **Gao Q**, Tsoi KK, Hirai HW, Wong MC, Chan FK, Wu JC, Lau JY, Sung JJ, Ng SC. Serrated polyps and the risk of synchronous colorectal advanced neoplasia: a systematic review and meta-analysis. *Am J Gastroenterol* 2015; **110**: 501-509; quiz 510 [PMID: 25756237 DOI: 10.1038/ajg.2015.49]

62 **Leggett BA**, Worthley DL. Synchronous colorectal cancer: not just bad luck? *Gastroenterology* 2009; **137**: 1559-1562 [PMID: 19789087 DOI: 10.1053/j.gastro.2009.09.025]

63 **Brenner H**, Kloor M, Pox CP. Colorectal cancer. *Lancet* 2014; **383**: 1490-1502 [PMID: 24225001 DOI: 10.1016/S0140-6736(13)61649-9]

64 **Lichtenstein P**, Holm NV, Verkasalo PK, Iliadou A, Kaprio J, Koskenvuo M, Pukkala E, Skytthe A, Hemminki K. Environmental and heritable factors in the causation of cancer--analyses of cohorts of twins from Sweden, Denmark, and Finland. *N Engl J Med* 2000; **343**: 78-85 [PMID: 10891514 DOI: 10.1056/NEJM200007133430201]

65 **Lynch HT**, de la Chapelle A. Hereditary colorectal cancer. *N Engl J Med* 2003; **348**: 919-932 [PMID: 12621137 DOI: 10.1056/NEJMra012242]

66 **Gonzalo V**, Lozano JJ, Alonso-Espinaco V, Moreira L, Muñoz J, Pellisé M, Castellví-Bel S, Bessa X, Andreu M, Xicola RM, Llor X, Ruiz-Ponte C, Carracedo A, Jover R, Castells A, Balaguer F; Gastrointestinal Oncology Group of the Spanish Gastroenterological Association. Multiple sporadic colorectal cancers display a unique methylation phenotype. *PLoS One* 2014; **9**: e91033 [PMID: 24643221 DOI: 10.1371/journal.pone.0091033]

67 **Jass JR**. Classification of colorectal cancer based on correlation of clinical, morphological and molecular features. *Histopathology* 2007; **50**: 113-130 [PMID: 17204026 DOI: 10.1111/j.1365-2559.2006.02549.x]

68 **Kloor M**, Staffa L, Ahadova A, von Knebel Doeberitz M. Clinical significance of microsatellite instability in colorectal cancer. *Langenbecks Arch Surg* 2014; **399**: 23-31 [PMID: 24048684 DOI: 10.1007/s00423-013-1112-3]

69 **Nosho K**, Kure S, Irahara N, Shima K, Baba Y, Spiegelman D, Meyerhardt JA, Giovannucci EL, Fuchs CS, Ogino S. A prospective cohort study shows unique epigenetic, genetic, and prognostic features of synchronous colorectal cancers. *Gastroenterology* 2009; **137**: 1609-1620.e1-3 [PMID: 19686742 DOI: 10.1053/j.gastro.2009.08.002]

70 **Pritchard CC**, Grady WM. Colorectal cancer molecular biology moves into clinical practice. *Gut* 2011; **60**: 116-129 [PMID: 20921207 DOI: 10.1136/gut.2009.206250]

71 **Howard ML**, Greene FL. The effect of preoperative endoscopy on recurrence and survival following surgery for colorectal carcinoma. *Am Surg* 1990; **56**: 124-127 [PMID: 2316931]

72 **Sun L**, Wu H, Guan YS. Colonography by CT, MRI and PET/CT combined with conventional colonoscopy in colorectal cancer screening and staging. *World J Gastroenterol* 2008; **14**: 853-863 [PMID: 18240342 DOI: 10.3748/wjg.14.853]

73 **Levine MS**, Yee J. History, evolution, and current status of radiologic imaging tests for colorectal cancer screening. *Radiology* 2014; **273**: S160-S180 [PMID: 25340435 DOI: 10.1148/radiol.14140531]

74 **Park SH**, Lee JH, Lee SS, Kim JC, Yu CS, Kim HC, Ye BD, Kim MJ, Kim AY, Ha HK. CT colonography for detection and characterisation of synchronous proximal colonic lesions in patients with stenosing colorectal cancer. *Gut* 2012; **61**: 1716-1722 [PMID: 22115824 DOI: 10.1136/gutjnl-2011-301135]

75 **Nishikawa T**, Ishihara S, Hata K, Murono K, Yasuda K, Otani K, Tanaka T, Kiyomatsu T, Kawai K, Nozawa H, Yamaguchi H, Watanabe T. Short-term outcomes of open versus laparoscopic surgery in elderly patients with colorectal cancer. *Surg Endosc* 2016; **30**: 5550-5557 [PMID: 27752818 DOI: 10.1007/s00464-016-4921-y]

76 **Heald RJ**, Ryall RD. Recurrence and survival after total mesorectal excision for rectal cancer. *Lancet* 1986; **1**: 1479-1482 [PMID: 2425199 DOI: 10.1016/S0140-6736(86)91510-2]

77 **Nagtegaal ID**, Quirke P. What is the role for the circumferential margin in the modern treatment of rectal cancer? *J Clin Oncol* 2008; **26**: 303-312 [PMID: 18182672 DOI: 10.1200/JCO.2007.12.7027]

78 **Caricato M**, Borzomati D, Ausania F, Valeri S, Rosignoli A, Coppola R. Prognostic factors after surgery for locally recurrent rectal cancer: an overview. *Eur J Surg Oncol* 2006; **32**: 126-132 [PMID: 16377120 DOI: 10.1016/j.ejso.2005.11.001]

79 **Belghiti J**, Hiramatsu K, Benoist S, Massault P, Sauvanet A, Farges O. Seven hundred forty-seven hepatectomies in the 1990s: an update to evaluate the actual risk of liver resection. *J Am Coll Surg* 2000; **191**: 38-46 [PMID: 10898182 DOI: 10.1016/S1072-7515(00)00261-1]

80 **Poon RT**, Ng KK, Lam CM, Ai V, Yuen J, Fan ST, Wong J. Learning curve for radiofrequency ablation of liver tumors: prospective analysis of initial 100 patients in a tertiary institution. *Ann Surg* 2004; **239**: 441-449 [PMID: 15024304 DOI: 10.1097/01.sla.0000118565.21298.0a]

81 **Brouquet A**, Mortenson MM, Vauthey JN, Rodriguez-Bigas MA, Overman MJ, Chang GJ, Kopetz S, Garrett C, Curley SA, Abdalla EK. Surgical strategies for synchronous colorectal liver metastases in 156 consecutive patients: classic, combined or reverse strategy? *J Am Coll Surg* 2010; **210**: 934-941 [PMID: 20510802 DOI: 10.1016/j.jamcollsurg.2010.02.039]

82 **Mayo SC**, Pulitano C, Marques H, Lamelas J, Wolfgang CL, de Saussure W, Choti MA, Gindrat I, Aldrighetti L, Barrosso E, Mentha G, Pawlik TM. Surgical management of patients with synchronous colorectal liver metastasis: a multicenter international analysis. *J Am Coll Surg* 2013; **216**: 707-716; discussion 716-718 [PMID: 23433970 DOI: 10.1016/j.jamcollsurg.2012.12.029]

83 **De Rosa A**, Gomez D, Brooks A, Cameron IC. "Liver-first" approach for synchronous colorectal liver metastases: is this a justifiable approach? *J Hepatobiliary Pancreat Sci* 2013; **20**: 263-270 [PMID: 23325126 DOI: 10.1007/s00534-012-0583-x]

84 **Valdimarsson VT**, Syk I, Lindell G, Norén A, Isaksson B, Sandström P, Rizell M, Ardnor B, Sturesson C. Outcomes of liver-first strategy and classical strategy for synchronous colorectal liver metastases in Sweden. *HPB* (Oxford) 2018; **20**: 441-447 [PMID: 29242035 DOI: 10.1016/j.hpb.2017.11.004]

85 **Buchs NC**, Ris F, Majno PE, Andres A, Cacheux W, Gervaz P, Roth AD, Terraz S, Rubbia-Brandt L, Morel P, Mentha G, Toso C. Rectal outcomes after a liver-first treatment of patients with stage IV rectal cancer. *Ann Surg Oncol* 2015; **22**: 931-937 [PMID: 25201505 DOI: 10.1245/s10434-014-4069-8]

86 **Abbott DE**, Cantor SB, Hu CY, Aloia TA, You YN, Nguyen S, Chang GJ. Optimizing clinical and economic outcomes of surgical therapy for patients with colorectal cancer and synchronous liver metastases. *J Am Coll Surg* 2012; **215**: 262-270 [PMID: 22560316 DOI: 10.1016/j.jamcollsurg.2012.03.021]

87 **Denstman F**. An approach to the newly diagnosed colorectal cancer patient with synchronous stage 4 disease. *Surg Oncol Clin N Am* 2014; **23**: 151-160 [PMID: 24267171 DOI: 10.1016/j.soc.2013.09.013]

88 **Cloyd JM**, Aloia TA. Hammer versus Swiss Army knife: Developing a strategy for the management of bilobar colorectal liver metastases. *Surgery* 2017; **162**: 12-17 [PMID: 28109616 DOI: 10.1016/j.surg.2016.11.035]

89 **Kokudo N**, Imamura H, Sugawara Y, Sakamoto Y, Yamamoto J, Seki M, Makuuchi M. Surgery for multiple hepatic colorectal metastases. *J Hepatobiliary Pancreat Surg* 2004; **11**: 84-91 [PMID: 15127269 DOI: 10.1007/s00534-002-0754-2]

90 **Yamamoto J**, Saiura A, Koga R, Seki M, Ueno M, Oya M, Azekura K, Seto Y, Ohyama S, Fukunaga S, Yamaguchi T, Kokudo N, Makuuchi M, Muto T. Surgical treatment for metastatic malignancies. Nonanatomical resection of liver metastasis: indications and outcomes. *Int J Clin Oncol* 2005; **10**: 97-102 [PMID: 15864694 DOI: 10.1007/s10147-004-0481-6]

91 **De Raffele E**, Mirarchi M, Vaccari S, Cuicchi D, Lecce F, Dalla Via B, Cola B. Intermittent clamping of the hepatic pedicle in simultaneous ultrasonography-guided liver resection and colorectal resection with intestinal anastomosis: is it safe? *Int J Colorectal Dis* 2014; **29**: 1517-1525 [PMID: 25185843 DOI: 10.1007/s00384-014-2004-x]

92 **Allard MA**, Adam R, Giuliante F, Lapointe R, Hubert C, Ijzermans JNM, Mirza DF, Elias D, Laurent C, Gruenberger T, Poston G, Letoublon C, Isoniemi H, Lucidi V, Popescu I, Figueras J. Long-term outcomes of patients with 10 or more colorectal liver metastases. *Br J Cancer* 2017; **117**: 604-611 [PMID: 28728167 DOI: 10.1038/bjc.2017.218]

93 **Narita M**, Oussoultzoglou E, Fuchshuber P, Pessaux P, Chenard MP, Rosso E, Nobili C, Jaeck D, Bachellier P. What is a safe future liver remnant size in patients undergoing major hepatectomy for colorectal liver metastases and treated by intensive preoperative chemotherapy? *Ann Surg Oncol* 2012; **19**: 2526-2538 [PMID: 22395987 DOI: 10.1245/s10434-012-2274-x]

94 **Khan AZ**, Morris-Stiff G, Makuuchi M. Patterns of chemotherapy-induced hepatic injury and their implications for patients undergoing liver resection for colorectal liver metastases. *J Hepatobiliary Pancreat Surg* 2009; **16**: 137-144 [PMID: 19093069 DOI: 10.1007/s00534-008-0016-z]

95 **Pessaux P**, Chenard MP, Bachellier P, Jaeck D. Consequences of chemotherapy on resection of colorectal liver metastases. *J Visc Surg* 2010; **147**: e193-e201 [PMID: 20655821 DOI: 10.1016/j.jviscsurg.2010.06.004]

96 **Zhao J**, van Mierlo KMC, Gómez-Ramírez J, Kim H, Pilgrim CHC, Pessaux P, Rensen SS, van der Stok EP, Schaap FG, Soubrane O, Takamoto T, Viganò L, Winkens B, Dejong CHC, Olde Damink SWM; Chemotherapy-Associated Liver Injury (CALI) consortium. Systematic review of the influence of chemotherapy-associated liver injury on outcome after partial hepatectomy for colorectal liver metastases. *Br J Surg* 2017; **104**: 990-1002 [PMID: 28542731 DOI: 10.1002/bjs.10572]

97 **Lehmann K**, Rickenbacher A, Weber A, Pestalozzi BC, Clavien PA. Chemotherapy before liver resection of colorectal metastases: friend or foe? *Ann Surg* 2012; **255**: 237-247 [PMID: 22041509 DOI: 10.1097/SLA.0b013e3182356236]

98 **van der Pool AE**, de Wilt JH, Lalmahomed ZS, Eggermont AM, Ijzermans JN, Verhoef C. Optimizing the outcome of surgery in patients with rectal cancer and synchronous liver metastases. *Br J Surg* 2010; **97**: 383-390 [PMID: 20101594 DOI: 10.1002/bjs.6947]

99 **Lykoudis PM**, O'Reilly D, Nastos K, Fusai G. Systematic review of surgical management of synchronous colorectal liver metastases. *Br J Surg* 2014; **101**: 605-612 [PMID: 24652674 DOI: 10.1002/bjs.9449]

100 **Kelly ME**, Spolverato G, Lê GN, Mavros MN, Doyle F, Pawlik TM, Winter DC. Synchronous colorectal liver metastasis: a network meta-analysis review comparing classical, combined, and liver-first surgical strategies. *J Surg Oncol* 2015; **111**: 341-351 [PMID: 25363294 DOI: 10.1002/jso.23819]

101 **Baltatzis M**, Chan AK, Jegatheeswaran S, Mason JM, Siriwardena AK. Colorectal cancer with synchronous hepatic metastases: Systematic review of reports comparing synchronous surgery with sequential bowel-first or liver-first approaches. *Eur J Surg Oncol* 2016; **42**: 159-165 [PMID: 26733368 DOI: 10.1016/j.ejso.2015.11.002]

102 **Nordlinger B**, Sorbye H, Glimelius B, Poston GJ, Schlag PM, Rougier P, Bechstein WO, Primrose JN, Walpole ET, Finch-Jones M, Jaeck D, Mirza D, Parks RW, Collette L, Praet M, Bethe U, Van Cutsem E, Scheithauer W, Gruenberger T; EORTC Gastro-Intestinal Tract Cancer Group; Cancer Research UK; Arbeitsgruppe Lebermetastasen und-tumoren in der Chirurgischen Arbeitsgemeinschaft Onkologie (ALM-CAO); Australasian Gastro-Intestinal Trials Group (AGITG); Fédération Francophone de Cancérologie Digestive (FFCD). Perioperative chemotherapy with FOLFOX4 and surgery versus surgery alone for resectable liver metastases from colorectal cancer (EORTC Intergroup trial 40983): a randomised controlled trial. *Lancet* 2008; **371**: 1007-1016 [PMID: 18358928 DOI: 10.1016/S0140-6736(08)60455-9]

103 **Nordlinger B**, Sorbye H, Glimelius B, Poston GJ, Schlag PM, Rougier P, Bechstein WO, Primrose JN, Walpole ET, Finch-Jones M, Jaeck D, Mirza D, Parks RW, Mauer M, Tanis E, Van Cutsem E, Scheithauer W, Gruenberger T; EORTC Gastro-Intestinal Tract Cancer Group; Cancer Research UK; Arbeitsgruppe Lebermetastasen und–tumoren in der Chirurgischen Arbeitsgemeinschaft Onkologie (ALM-CAO); Australasian Gastro-Intestinal Trials Group (AGITG); Fédération Francophone de Cancérologie Digestive (FFCD). Perioperative FOLFOX4 chemotherapy and surgery versus surgery alone for resectable liver metastases from colorectal cancer (EORTC 40983): long-term results of a randomised, controlled, phase 3 trial. *Lancet Oncol* 2013; **14**: 1208-1215 [PMID: 24120480 DOI: 10.1016/S1470-2045(13)70447-9]

104 **Chua TC**, Saxena A, Liauw W, Kokandi A, Morris DL. Systematic review of randomized and nonrandomized trials of the clinical response and outcomes of neoadjuvant systemic chemotherapy for resectable colorectal liver metastases. *Ann Surg Oncol* 2010; **17**: 492-501 [PMID: 19856028 DOI: 10.1245/s10434-009-0781-1]

105 **Bonney GK**, Coldham C, Adam R, Kaiser G, Barroso E, Capussotti L, Laurent C, Verhoef C, Nuzzo G, Elias D, Lapointe R, Hubert C, Lopez-Ben S, Krawczyk M, Mirza DF; LiverMetSurvey International Registry Working Group. Role of neoadjuvant chemotherapy in resectable synchronous colorectal liver metastasis; An international multi-center data analysis using LiverMetSurvey. *J Surg Oncol* 2015; **111**: 716-724 [PMID: 25864987 DOI: 10.1002/jso.23899]

106 **Primrose J**, Falk S, Finch-Jones M, Valle J, O'Reilly D, Siriwardena A, Hornbuckle J, Peterson M, Rees M, Iveson T, Hickish T, Butler R, Stanton L, Dixon E, Little L, Bowers M, Pugh S, Garden OJ, Cunningham D, Maughan T, Bridgewater J. Systemic chemotherapy with or without cetuximab in patients with resectable colorectal liver metastasis: the New EPOC randomised controlled trial. *Lancet Oncol* 2014; **15**: 601-611 [PMID: 24717919 DOI: 10.1016/S1470-2045(14)70105-6]

107 **Charnsangavej C**, Clary B, Fong Y, Grothey A, Pawlik TM, Choti MA. Selection of patients for resection of hepatic colorectal metastases: expert consensus statement. *Ann Surg Oncol* 2006; **13**: 1261-1268 [PMID: 16947009 DOI: 10.1245/s10434-006-9023-y]

108 **Ekberg H**, Tranberg KG, Andersson R, Lundstedt C, Hägerstrand I, Ranstam J, Bengmark S. Determinants of survival in liver resection for colorectal secondaries. *Br J Surg* 1986; **73**: 727-731 [PMID: 3756436 DOI: 10.1002/bjs.1800730917]

109 **Cauchy F**, Soubrane O, Belghiti J. Liver resection for HCC: patient's selection and controversial scenarios. *Best Pract Res Clin Gastroenterol* 2014; **28**: 881-896 [PMID: 25260315 DOI: 10.1016/j.bpg.2014.08.013]

110 **Makuuchi M**, Hasegawa H, Yamazaki S. Ultrasonically guided subsegmentectomy. *Surg Gynecol Obstet* 1985; **161**: 346-350 [PMID: 2996162]

111 **Makuuchi M**, Hasegawa H, Yamazaki S, Takayasu K. Four new hepatectomy procedures for resection of the right hepatic vein and preservation of the inferior right hepatic vein. *Surg Gynecol Obstet* 1987; **164**: 68-72 [PMID: 3026059]

112 **Gozzetti G**, Mazziotti A, Cavallari A, Bellusci R, Bolondi L, Grigioni W, Bragaglia R, Grazi GL, De Raffele E. Clinical experience with hepatic resections for hepatocellular carcinoma in patients with cirrhosis. *Surg Gynecol Obstet* 1988; **166**: 503-510 [PMID: 2836959]

113 **Torzilli G**, Montorsi M, Donadon M, Palmisano A, Del Fabbro D, Gambetti A, Olivari N, Makuuchi M. "Radical but conservative" is the main goal for ultrasonography-guided liver resection: prospective validation of this approach. *J Am Coll Surg* 2005; **201**: 517-528 [PMID: 16183489 DOI: 10.1016/j.jamcollsurg.2005.04.026]

114 **Torzilli G**, Donadon M, Marconi M, Botea F, Palmisano A, Del Fabbro D, Procopio F, Montorsi M. Systematic extended right posterior sectionectomy: a safe and effective alternative to right hepatectomy. *Ann Surg* 2008; **247**: 603-611 [PMID: 18362622 DOI: 10.1097/SLA.0b013e31816387d7]

115 **Knijn N**, de Ridder JA, Punt CJ, de Wilt JH, Nagtegaal ID. Histopathological evaluation of resected colorectal cancer liver metastases: what should be done? *Histopathology* 2013; **63**: 149-156 [PMID: 23763641 DOI: 10.1111/his.12124]

116 **Sasaki A**, Aramaki M, Kawano K, Yasuda K, Inomata M, Kitano S. Prognostic significance of intrahepatic lymphatic invasion in patients with hepatic resection due to metastases from colorectal carcinoma. *Cancer* 2002; **95**: 105-111 [PMID: 12115323 DOI: 10.1002/cncr.10655]

117 **Korita PV**, Wakai T, Shirai Y, Sakata J, Takizawa K, Cruz PV, Ajioka Y, Hatakeyama K. Intrahepatic lymphatic invasion independently predicts poor survival and recurrences after hepatectomy in patients with colorectal carcinoma liver metastases. *Ann Surg Oncol* 2007; **14**: 3472-3480 [PMID: 17828431 DOI: 10.1245/s10434-007-9594-2]

118 **Lupinacci RM**, Mello ES, Pinheiro RS, Marques G, Coelho FF, Kruger JA, Perini MV, Herman P. Intrahepatic lymphatic invasion but not vascular invasion is a major prognostic factor after resection of colorectal cancer liver metastases. *World J Surg* 2014; **38**: 2089-2096 [PMID: 24663482 DOI: 10.1007/s00268-014-2511-5]

119 **Wakai T**, Shirai Y, Sakata J, Valera VA, Korita PV, Akazawa K, Ajioka Y, Hatakeyama K. Appraisal of 1 cm hepatectomy margins for intrahepatic micrometastases in patients with colorectal carcinoma liver metastasis. *Ann Surg Oncol* 2008; **15**: 2472-2481 [PMID: 18594929 DOI: 10.1245/s10434-008-0023-y]

120 **Yamamoto J**, Sugihara K, Kosuge T, Takayama T, Shimada K, Yamasaki S, Sakamoto M, Hirohashi S. Pathologic support for limited hepatectomy in the treatment of liver metastases from colorectal cancer. *Ann Surg* 1995; **221**: 74-78 [PMID: 7826164 DOI: 10.1097/00000658-199501000-00009]

121 **Sui CJ**, Cao L, Li B, Yang JM, Wang SJ, Su X, Zhou YM. Anatomical versus nonanatomical resection of colorectal liver metastases: a meta-analysis. *Int J Colorectal Dis* 2012; **27**: 939-946 [PMID: 22215149 DOI: 10.1007/s00384-011-1403-5]

122 **Moris D**, Ronnekleiv-Kelly S, Rahnemai-Azar AA, Felekouras E, Dillhoff M, Schmidt C, Pawlik TM. Parenchymal-Sparing Versus Anatomic Liver Resection for Colorectal Liver Metastases: a Systematic Review. *J Gastrointest Surg* 2017; **21**: 1076-1085 [PMID: 28364212 DOI: 10.1007/s11605-017-3397-y]

123 **Kokudo N**, Tada K, Seki M, Ohta H, Azekura K, Ueno M, Matsubara T, Takahashi T, Nakajima T, Muto T. Anatomical major resection versus nonanatomical limited resection for liver metastases from colorectal carcinoma. *Am J Surg* 2001; **181**: 153-159 [PMID: 11425058 DOI: 10.1016/S0002-9610(00)00560-2]

124 **Mise Y**, Aloia TA, Brudvik KW, Schwarz L, Vauthey JN, Conrad C. Parenchymal-sparing Hepatectomy in Colorectal Liver Metastasis Improves Salvageability and Survival. *Ann Surg* 2016; **263**: 146-152 [PMID: 25775068 DOI: 10.1097/SLA.0000000000001194]

125 **Hosokawa I**, Allard MA, Mirza DF, Kaiser G, Barroso E, Lapointe R, Laurent C, Ferrero A, Miyazaki M, Adam R. Outcomes of parenchyma-preserving hepatectomy and right hepatectomy for solitary small colorectal liver metastasis: A LiverMetSurvey study.

Surgery 2017; **162**: 223-232 [PMID: 28434557 DOI: 10.1016/j.surg.2017.02.012]
126 **Karanjia ND**, Lordan JT, Quiney N, Fawcett WJ, Worthington TR, Remington J. A comparison of right and extended right hepatectomy with all other hepatic resections for colorectal liver metastases: a ten-year study. *Eur J Surg Oncol* 2009; **35**: 65-70 [PMID: 18222623 DOI: 10.1016/j.ejso.2007.12.002]
127 **Lordan JT**, Roberts JK, Hodson J, Isaac J, Muiesan P, Mirza DF, Marudanayagam R, Sutcliffe RP. Case-controlled study comparing peri-operative and cancer-related outcomes after major hepatectomy and parenchymal sparing hepatectomy for metastatic colorectal cancer. *HPB* (Oxford) 2017; **19**: 688-694 [PMID: 28495437 DOI: 10.1016/j.hpb.2017.04.007]
128 **Matsuki R**, Mise Y, Saiura A, Inoue Y, Ishizawa T, Takahashi Y. Parenchymal-sparing hepatectomy for deep-placed colorectal liver metastases. *Surgery* 2016; **160**: 1256-1263 [PMID: 27521044 DOI: 10.1016/j.surg.2016.06.041]
129 **Bhutiani N**, Philips P, Martin RC 2nd, Scoggins CR. Impact of surgical margin clearance for resection of secondary hepatic malignancies. *J Surg Oncol* 2016; **113**: 289-295 [PMID: 26662026 DOI: 10.1002/jso.24107]
130 **Kokudo N**, Miki Y, Sugai S, Yanagisawa A, Kato Y, Sakamoto Y, Yamamoto J, Yamaguchi T, Muto T, Makuuchi M. Genetic and histological assessment of surgical margins in resected liver metastases from colorectal carcinoma: minimum surgical margins for successful resection. *Arch Surg* 2002; **137**: 833-840 [PMID: 12093342 DOI: 10.1001/archsurg.137.7.833]
131 **Holdhoff M**, Schmidt K, Diehl F, Aggrawal N, Angenendt P, Romans K, Edelstein DL, Torbenson M, Kinzler KW, Vogelstein B, Choti MA, Diaz LA Jr. Detection of tumor DNA at the margins of colorectal cancer liver metastasis. *Clin Cancer Res* 2011; **17**: 3551-3557 [PMID: 21531819 DOI: 10.1158/1078-0432.CCR-10-3087]
132 **Yokoyama N**, Shirai Y, Ajioka Y, Nagakura S, Suda T, Hatakeyama K. Immunohistochemically detected hepatic micrometastases predict a high risk of intrahepatic recurrence after resection of colorectal carcinoma liver metastases. *Cancer* 2002; **94**: 1642-1647 [PMID: 11920523 DOI: 10.1002/cncr.10422]
133 **Wakai T**, Shirai Y, Sakata J, Kameyama H, Nogami H, Iiai T, Ajioka Y, Hatakeyama K. Histologic evaluation of intrahepatic micrometastases in patients treated with or without neoadjuvant chemotherapy for colorectal carcinoma liver metastasis. *Int J Clin Exp Pathol* 2012; **5**: 308-314 [PMID: 22670174]
134 **Yamamoto J**, Shimada K, Kosuge T, Yamasaki S, Sakamoto M, Fukuda H. Factors influencing survival of patients undergoing hepatectomy for colorectal metastases. *Br J Surg* 1999; **86**: 332-337 [PMID: 10201774 DOI: 10.1046/j.1365-2168.1999.01030.x]
135 **Okano K**, Yamamoto J, Kosuge T, Yamamoto S, Sakamoto M, Nakanishi Y, Hirohashi S. Fibrous pseudocapsule of metastatic liver tumors from colorectal carcinoma. Clinicopathologic study of 152 first resection cases. *Cancer* 2000; **89**: 267-275 [PMID: 10918155 DOI: 10.1002/1097-0142(20000715)89:2<267::AID-CNCR10>3.0.CO;2-1]
136 **Mentha G**, Terraz S, Morel P, Andres A, Giostra E, Roth A, Rubbia-Brandt L, Majno P. Dangerous halo after neoadjuvant chemotherapy and two-step hepatectomy for colorectal liver metastases. *Br J Surg* 2009; **96**: 95-103 [PMID: 19109800 DOI: 10.1002/bjs.6436]
137 **Busquets J**, Pelaez N, Alonso S, Grande L. The study of cavitational ultrasonically aspirated material during surgery for colorectal liver metastases as a new concept in resection margin. *Ann Surg* 2006; **244**: 634-635 [PMID: 16998378 DOI: 10.1097/01.sla.0000239631.74713.b5]
138 **Pawlik TM**, Scoggins CR, Zorzi D, Abdalla EK, Andres A, Eng C, Curley SA, Loyer EM, Muratore A, Mentha G, Capussotti L, Vauthey JN. Effect of surgical margin status on survival and site of recurrence after hepatic resection for colorectal metastases. *Ann Surg* 2005; **241**: 715-722, discussion 722-discussion 724 [PMID: 15849507]
139 **Nuzzo G**, Giuliante F, Ardito F, Vellone M, Giovannini I, Federico B, Vecchio FM. Influence of surgical margin on type of recurrence after liver resection for colorectal metastases: a single-center experience. *Surgery* 2008; **143**: 384-393 [PMID: 18291260 DOI: 10.1016/j.surg.2007.09.038]
140 **Muratore A**, Ribero D, Zimmitti G, Mellano A, Langella S, Capussotti L. Resection margin and recurrence-free survival after liver resection of colorectal metastases. *Ann Surg Oncol* 2010; **17**: 1324-1329 [PMID: 19847565 DOI: 10.1245/s10434-009-0770-4]
141 **de Haas RJ**, Wicherts DA, Flores E, Azoulay D, Castaing D, Adam R. R1 resection by necessity for colorectal liver metastases: is it still a contraindication to surgery? *Ann Surg* 2008; **248**: 626-637 [PMID: 18936576 DOI: 10.1097/SLA.0b013e31818a07f1]
142 **Tranchart H**, Chirica M, Faron M, Balladur P, Lefevre LB, Svrcek M, de Gramont A, Tiret E, Paye F. Prognostic impact of positive surgical margins after resection of colorectal cancer liver metastases: reappraisal in the era of modern chemotherapy. *World J Surg* 2013; **37**: 2647-2654 [PMID: 23982776 DOI: 10.1007/s00268-013-2186-3]
143 **Hamady ZZ**, Lodge JP, Welsh FK, Toogood GJ, White A, John T, Rees M. One-millimeter cancer-free margin is curative for colorectal liver metastases: a propensity score case-match approach. *Ann Surg* 2014; **259**: 543-548 [PMID: 23732261 DOI: 10.1097/SLA.0b013e3182902b6e]
144 **Sadot E**, Groot Koerkamp B, Leal JN, Shia J, Gonen M, Allen PJ, DeMatteo RP, Kingham TP, Kemeny N, Blumgart LH, Jarnagin WR, D Angelica MI. Resection margin and survival in 2368 patients undergoing hepatic resection for metastatic colorectal cancer: surgical technique or biologic surrogate? *Ann Surg* 2015; **262**: 476-485; discussion 483-485 [PMID: 26258316 DOI: 10.1097/SLA.0000000000001427]
145 **Truant S**, Séquier C, Leteurtre E, Boleslawski E, Elamrani M, Huet G, Duhamel A, Hebbar M, Pruvot FR. Tumour biology of colorectal liver metastasis is a more important factor in survival than surgical margin clearance in the era of modern chemotherapy regimens. *HPB* (Oxford) 2015; **17**: 176-184 [PMID: 25041611 DOI: 10.1111/hpb.12316]
146 **Ayez N**, Lalmahomed ZS, Eggermont AM, Ijzermans JN, de Jonge J, van Montfort K, Verhoef C. Outcome of microscopic incomplete resection (R1) of colorectal liver metastases in the era of neoadjuvant chemotherapy. *Ann Surg Oncol* 2012; **19**: 1618-1627 [PMID: 22006375 DOI: 10.1245/s10434-011-2114-4]
147 **Andreou A**, Aloia TA, Brouquet A, Dickson PV, Zimmitti G, Maru DM, Kopetz S, Loyer EM, Curley SA, Abdalla EK, Vauthey JN. Margin status remains an important determinant of survival after surgical resection of colorectal liver metastases in the era of modern chemotherapy. *Ann Surg* 2013; **257**: 1079-1088 [PMID: 23426338 DOI: 10.1097/SLA.0b013e318283a4d1]
148 **Miller CL**, Taylor MS, Qadan M, Deshpande V, Worthington S, Smalley R, Collura C, Ryan DP, Allen JN, Blaszkowsky LS, Clark JW, Murphy JE, Parikh AR, Berger D, Tanabe KK, Lillemoe KD, Ferrone CR. Prognostic Significance of Surgical Margin Size After Neoadjuvant FOLFOX and/or FOLFIRI for Colorectal Liver Metastases. *J Gastrointest Surg* 2017; **21**: 1831-1840 [PMID: 28884391 DOI: 10.1007/s11605-017-3557-0]
149 **Ng JK**, Urbanski SJ, Mangat N, McKay A, Sutherland FR, Dixon E, Dowden S, Ernst S, Bathe OF. Colorectal liver metastases contract centripetally with a response to chemotherapy: a histomorphologic study. *Cancer* 2008; **112**: 362-371 [PMID: 18041069 DOI: 10.1002/cncr.23184]
150 **Torzilli G**, Viganò L, Gatti A, Costa G, Cimino M, Procopio F, Donadon M, Del Fabbro D. Twelve-year experience of "radical but conservative" liver surgery for colorectal metastases: impact on surgical practice and oncologic efficacy. *HPB* (Oxford) 2017; **19**: 775-784 [PMID: 28625391 DOI: 10.1016/j.hpb.2017.05.006]
151 **Viganò L**, Procopio F, Cimino MM, Donadon M, Gatti A, Costa G, Del Fabbro D, Torzilli G. Is Tumor Detachment from Vascular Structures Equivalent to R0 Resection in Surgery for Colorectal Liver Metastases? An Observational Cohort. *Ann Surg Oncol* 2016; **23**: 1352-1360 [PMID: 26714946 DOI: 10.1245/s10434-015-5009-y]
152 **Memeo R**, de Blasi V, Adam R, Goéré D, Piardi T, Lermite E, Turrini O, Navarro F, de'Angelis N, Cunha AS, Pessaux P;

French Colorectal Liver Metastases Working Group, Association Française de Chirurgie (AFC). Margin Status is Still an Important Prognostic Factor in Hepatectomies for Colorectal Liver Metastases: A Propensity Score Matching Analysis. *World J Surg* 2018; **42**: 892-901 [PMID: 28929341 DOI: 10.1007/s00268-017-4229-7]

153 **Are C**, Gonen M, Zazzali K, Dematteo RP, Jarnagin WR, Fong Y, Blumgart LH, D'Angelica M. The impact of margins on outcome after hepatic resection for colorectal metastasis. *Ann Surg* 2007; **246**: 295-300 [PMID: 17667509 DOI: 10.1097/SLA.0b013e31811ea962]

154 **Dhir M**, Lyden ER, Wang A, Smith LM, Ullrich F, Are C. Influence of margins on overall survival after hepatic resection for colorectal metastasis: a meta-analysis. *Ann Surg* 2011; **254**: 234-242 [PMID: 21694583 DOI: 10.1097/SLA.0b013e318223c609]

155 **Liu W**, Sun Y, Zhang L, Xing BC. Negative surgical margin improved long-term survival of colorectal cancer liver metastases after hepatic resection: a systematic review and meta-analysis. *Int J Colorectal Dis* 2015; **30**: 1365-1373 [PMID: 26198997 DOI: 10.1007/s00384-015-2323-6]

156 **Margonis GA**, Sergentanis TN, Ntanasis-Stathopoulos I, Andreatos N, Tzanninis IG, Sasaki K, Psaltopoulou T, Wang J, Buettner S, Papalois AE, He J, Wolfgang CL, Pawlik TM, Weiss MJ. Impact of Surgical Margin Width on Recurrence and Overall Survival Following R0 Hepatic Resection of Colorectal Metastases: A Systematic Review and Meta-analysis. *Ann Surg* 2018; **267**: 1047-1055 [PMID: 29189379 DOI: 10.1097/SLA.0000000000002552]

157 **Adson MA**, van Heerden JA, Adson MH, Wagner JS, Ilstrup DM. Resection of hepatic metastases from colorectal cancer. *Arch Surg* 1984; **119**: 647-651 [PMID: 6732473 DOI: 10.1001/archsurg.1984.01390180015003]

158 **Hughes KS**, Rosenstein RB, Songhorabodi S, Adson MA, Ilstrup DM, Fortner JG, Maclean BJ, Foster JH, Daly JM, Fitzherbert D. Resection of the liver for colorectal carcinoma metastases. A multi-institutional study of long-term survivors. *Dis Colon Rectum* 1988; **31**: 1-4 [PMID: 3366020 DOI: 10.1007/BF02552560]

159 **Scheele J**, Stang R, Altendorf-Hofmann A, Paul M. Resection of colorectal liver metastases. *World J Surg* 1995; **19**: 59-71 [PMID: 7740812 DOI: 10.1007/BF00316981]

160 **Weber SM**, Jarnagin WR, DeMatteo RP, Blumgart LH, Fong Y. Survival after resection of multiple hepatic colorectal metastases. *Ann Surg Oncol* 2000; **7**: 643-650 [PMID: 11034240 DOI: 10.1007/s10434-000-0643-3]

161 **Tanaka K**, Adam R, Shimada H, Azoulay D, Lévi F, Bismuth H. Role of neoadjuvant chemotherapy in the treatment of multiple colorectal metastases to the liver. *Br J Surg* 2003; **90**: 963-969 [PMID: 12905549 DOI: 10.1002/bjs.4160]

162 **Adam R**, Laurent A, Azoulay D, Castaing D, Bismuth H. Two-stage hepatectomy: A planned strategy to treat irresectable liver tumors. *Ann Surg* 2000; **232**: 777-785 [PMID: 11088072 DOI: 10.1097/00000658-200012000-00006]

163 **Azoulay D**, Castaing D, Smail A, Adam R, Cailliez V, Laurent A, Lemoine A, Bismuth H. Resection of nonresectable liver metastases from colorectal cancer after percutaneous portal vein embolization. *Ann Surg* 2000; **231**: 480-486 [PMID: 10749607 DOI: 10.1097/00000658-200004000-00005]

164 **Jaeck D**, Oussoultzoglou E, Rosso E, Greget M, Weber JC, Bachellier P. A two-stage hepatectomy procedure combined with portal vein embolization to achieve curative resection for initially unresectable multiple and bilobar colorectal liver metastases. *Ann Surg* 2004; **240**: 1037-1049; discussion 1049-1051 [PMID: 15570209 DOI: 10.1097/01.sla.0000145965.86383.89]

165 **Torzilli G**, Procopio F, Botea F, Marconi M, Del Fabbro D, Donadon M, Palmisano A, Spinelli A, Montorsi M. One-stage ultrasonographically guided hepatectomy for multiple bilobar colorectal metastases: a feasible and effective alternative to the 2-stage approach. *Surgery* 2009; **146**: 60-71 [PMID: 19541011 DOI: 10.1016/j.surg.2009.02.017]

166 **Abdalla EK**, Vauthey JN, Ellis LM, Ellis V, Pollock R, Broglio KR, Hess K, Curley SA. Recurrence and outcomes following hepatic resection, radiofrequency ablation, and combined resection/ablation for colorectal liver metastases. *Ann Surg* 2004; **239**: 818-825; discussion 825-827 [PMID: 15166961 DOI: 10.1097/01.sla.0000128305.90650.71]

167 **Lee H**, Heo JS, Cho YB, Yun SH, Kim HC, Lee WY, Choi SH, Choi DW. Hepatectomy vs radiofrequency ablation for colorectal liver metastasis: a propensity score analysis. *World J Gastroenterol* 2015; **21**: 3300-3307 [PMID: 25805937 DOI: 10.3748/wjg.v21.i11.3300]

168 **van Amerongen MJ**, Jenniskens SFM, van den Boezem PB, Fütterer JJ, de Wilt JHW. Radiofrequency ablation compared to surgical resection for curative treatment of patients with colorectal liver metastases - a meta-analysis. *HPB* (Oxford) 2017; **19**: 749-756 [PMID: 28687147 DOI: 10.1016/j.hpb.2017.05.011]

169 **Karanicolas PJ**, Jarnagin WR, Gonen M, Tuorto S, Allen PJ, DeMatteo RP, D'Angelica MI, Fong Y. Long-term outcomes following tumor ablation for treatment of bilateral colorectal liver metastases. *JAMA Surg* 2013; **148**: 597-601 [PMID: 23699996 DOI: 10.1001/jamasurg.2013.1431]

170 **Evrard S**, Poston G, Kissmeyer-Nielsen P, Diallo A, Desolneux G, Brouste V, Lalet C, Mortensen F, Stättner S, Fenwick S, Malik H, Konstantinidis I, DeMatteo R, D'Angelica M, Allen P, Jarnagin W, Mathoulin-Pelissier S, Fong Y. Combined ablation and resection (CARe) as an effective parenchymal sparing treatment for extensive colorectal liver metastases. *PLoS One* 2014; **9**: e114404 [PMID: 25485541 DOI: 10.1371/journal.pone.0114404]

171 **Philips P**, Groeschl RT, Hanna EM, Swan RZ, Turaga KK, Martinie JB, Iannitti DA, Schmidt C, Gamblin TC, Martin RC. Single-stage resection and microwave ablation for bilobar colorectal liver metastases. *Br J Surg* 2016; **103**: 1048-1054 [PMID: 27191368 DOI: 10.1002/bjs.10159]

172 **Faitot F**, Faron M, Adam R, Elias D, Cimino M, Cherqui D, Vibert E, Castaing D, Cunha AS, Goéré D. Two-stage hepatectomy versus 1-stage resection combined with radiofrequency for bilobar colorectal metastases: a case-matched analysis of surgical and oncological outcomes. *Ann Surg* 2014; **260**: 822-827; discussion 827-828 [PMID: 25379853 DOI: 10.1097/SLA.0000000000000976]

173 **Saiura A**, Yamamoto J, Hasegawa K, Koga R, Sakamoto Y, Hata S, Makuuchi M, Kokudo N. Liver resection for multiple colorectal liver metastases with surgery up-front approach: bi-institutional analysis of 736 consecutive cases. *World J Surg* 2012; **36**: 2171-2178 [PMID: 22547015 DOI: 10.1007/s00268-012-1616-y]

174 **Viganò L**, Capussotti L, Majno P, Toso C, Ferrero A, De Rosa G, Rubbia-Brandt L, Mentha G. Liver resection in patients with eight or more colorectal liver metastases. *Br J Surg* 2015; **102**: 92-101 [PMID: 25451181 DOI: 10.1002/bjs.9680]

175 **Abbott AM**, Parsons HM, Tuttle TM, Jensen EH. Short-term outcomes after combined colon and liver resection for synchronous colon cancer liver metastases: a population study. *Ann Surg Oncol* 2013; **20**: 139-147 [PMID: 22825774 DOI: 10.1245/s10434-012-2515-z]

176 **Zalinski S**, Mariette C, Farges O; SFCD-ACHBT evaluation committee : A. Alves, I. Baum-gaertner, C. Cabral, J. Carles, C. Diana, O. Dubreuil, D. Fuks, D. Goere, M. Karoui, J. Lefevre, P. Pessaux, G. Schmidt, O. Turrini, E. Vibert, J-C. Weber; French Society of Gastrointestinal Surgery (SFCD); Association of Hepatobiliary Surgery and Liver Transplantation (ACHBT). Management of patients with synchronous liver metastases of colorectal cancer. Clinical practice guidelines. Guidelines of the French society of gastrointestinal surgery (SFCD) and of the association of hepatobiliary surgery and liver transplantation (ACHBT). Short version. *J Visc Surg* 2011; **148**: e171-e182 [PMID: 21703959 DOI: 10.1016/j.jviscsurg.2011.05.015]

177 **Qureshi MS**, Goldsmith PJ, Maslekar S, Prasad KR, Botterill ID. Synchronous resection of colorectal cancer and liver metastases: comparative views of colorectal and liver surgeons. *Colorectal Dis* 2012; **14**: e477-e485 [PMID: 22340783 DOI: 10.1111/j.1463-1318.2012.02992.x]

178 **Tanaka K**, Shimada H, Matsuo K, Nagano Y, Endo I, Sekido H, Togo S. Outcome after simultaneous colorectal and hepatic resection for colorectal cancer with synchronous metastases. *Surgery* 2004;

136: 650-659 [PMID: 15349115 DOI: 10.1016/j.surg.2004.02.012]
179 **Fahy BN**, Fischer CP. Synchronous resection of colorectal primary and hepatic metastasis. *J Gastrointest Oncol* 2012; **3**: 48-58 [PMID: 22811869 DOI: 10.3978/j.issn.2078-6891.2012.004]
180 **Minagawa M**, Yamamoto J, Miwa S, Sakamoto Y, Kokudo N, Kosuge T, Miyagawa S, Makuuchi M. Selection criteria for simultaneous resection in patients with synchronous liver metastasis. *Arch Surg* 2006; **141**: 1006-1012; discussion 1013 [PMID: 17043279 DOI: 10.1001/archsurg.141.10.1006]
181 **Yoshioka R**, Hasegawa K, Mise Y, Oba M, Aoki T, Sakamoto Y, Sugawara Y, Sunami E, Watanabe T, Kokudo N. Evaluation of the safety and efficacy of simultaneous resection of primary colorectal cancer and synchronous colorectal liver metastases. *Surgery* 2014; **155**: 478-485 [PMID: 24439744 DOI: 10.1016/j.surg.2013.10.015]

P- Reviewer: Ciocalteu A, Mu YP **S- Editor**: Gong ZM
L- Editor: Filipodia **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 317-327

DOI: 10.4251/wjgo.v10.i10.317

ISSN 1948-5204 (online)

MINIREVIEWS

Histo-molecular oncogenesis of pancreatic cancer: From precancerous lesions to invasive ductal adenocarcinoma

Giulio Riva, Antonio Pea, Camilla Pilati, Giulia Fiadone, Rita Teresa Lawlor, Aldo Scarpa, Claudio Luchini

Giulio Riva, Giulia Fiadone, Aldo Scarpa, Claudio Luchini, Department of Diagnostics and Public Health, Section of Pathology, University and Hospital Trust of Verona, Verona 37134, Italy

Antonio Pea, Department of Surgery, University and Hospital trust of Verona, Verona 37134, Italy

Camilla Pilati, Personalized Medicine, Pharmacogenomics, Therapeutic Optimization, Paris-Descartes University, Paris 75006, France

Rita Teresa Lawlor, ARC-Net Research Center, University and Hospital Trust of Verona, Verona 37134, Italy

ORCID number: Giulio Riva (0000-0001-6636-0605); Antonio Pea (0000-0002-0509-6756); Camilla Pilati (0000-0002-1781-5180); Giulia Fiadone (0000-0003-2351-789X); Rita Teresa Lawlor (0000-0003-3160-0634); Aldo Scarpa (0000-0003-1678-739X); Claudio Luchini (0000-0003-4901-4908).

Author contributions: Luchini C, Scarpa A and Lawlor RT conceived and designed the study; Riva G, Pea A, Lawlor RT and Luchini C performed the literature review; all authors analyzed and interpreted literature; Riva G, Pea A, Scarpa A and Luchini C wrote the manuscript; all authors edited and approved the manuscript in its present form.

Supported by the Associazione Italiana Ricerca sul Cancro, No. 12182; and Cassini Project.

Conflict-of-interest statement: No potential conflict of interest.

Manuscript source: Invited Manuscript

Correspondence to: Claudio Luchini, MD, PhD, Assistant Professor, Department of Diagnostics and Public Health, University and Hospital Trust of Verona, Piazzale L.A. Scuro, 10, Verona 37134, Italy. claudio.luchini@univr.it
Telephone: +39-45-8124835
Fax: +39-45-8027136

Received: May 30, 2018
Peer-review started: May 30, 2018
First decision: July 3, 2018
Revised: July 13, 2018
Accepted: August 12, 2018
Article in press: August 13, 2018
Published online: October 15, 2018

Abstract

Pancreatic cancer is a lethal malignancy, whose precursor lesions are pancreatic intraepithelial neoplasm, intraductal papillary mucinous neoplasm, intraductal tubulopapillary neoplasm, and mucinous cystic neoplasm. To better understand the biology of pancreatic cancer, it is fundamental to know its precursors and to study the mechanisms of carcinogenesis. Each of these precursors displays peculiar histological features, as well as specific molecular alterations. Starting from such pre-invasive lesions, this review aims at summarizing the most important aspects of carcinogenesis of pancreatic cancer, with a specific focus on the recent advances and the future perspectives of the research on this lethal tumor type.

Key words: Oncogenesis; Intraductal papillary mucinous neoplasm; Mucinous cystic neoplasm; Pancreatic ductal adenocarcinoma; Pancreatic intraepithelial neoplasm; *KRAS*; Carcinogenesis; Pancreatic cancer; Intraductal tubulopapillary neoplasm

Core tip: Pancreatic intraepithelial neoplasm, intraductal papillary mucinous neoplasm, intraductal tubulopapillary neoplasm, and mucinous cystic neoplasm are precursor lesions of invasive pancreatic cancer. Each of these precursors displays peculiar histological and molecular features, which have been summarized in this review along with the most important aspects of pancreatic carcinogenesis. The most recent advances and the future perspectives of the research on this topic have also been highlighted.

Riva G, Pea A, Pilati C, Fiadone G, Lawlor RT, Scarpa A, Luchini C. Histo-molecular oncogenesis of pancreatic cancer: From precancerous lesions to invasive ductal adenocarcinoma. *World J Gastrointest Oncol* 2018; 10(10): 317-327 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/317.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.317

INTRODUCTION

Precursor lesions of pancreatic cancer

Precursor lesions of pancreatic ductal adenocarcinoma (PDAC) are non-invasive lesions, which can progress to infiltrating carcinoma[1]. Following the 2010 World Health Organization classification, different consensus conferences and international meetings have provided the basis for the current view of the definition and classification of pancreatic precursor lesions[2-4].

One major issue in classification is in regards to the grading of dysplasia of pre-invasive lesions, which should be restricted to the two categories of "low-grade" and "high-grade" with the elimination of the poorly reproducible intermediate entity of "moderate dysplasia"[5]. Furthermore, small intraductal papillary mucinous lesions (PanIN) ranging from 0.5 to 1.0 cm would be better defined as "incipient intraductal papillary mucinous neoplasm (IPMN)"[5].

Three PDAC precursors have been recognized: PanIN, IPMN, and mucinous cystic neoplasm (MCN). In addition, intratubular papillary neoplasms (ITPNs) display the features of pre-invasive lesion. Each of these entities has distinct clinical, histological, and molecular profiles. Through a multi-step carcinogenesis, with the accumulation of cellular and molecular alterations, each of these precursors may lead to the development of invasive ductal adenocarcinoma.

CLINICAL FEATURES OF PRECURSOR LESIONS OF PANCREATIC CANCER

PanIN

PanIN represents the most common PDAC precursor, affecting both men and women equally. Its incidence increases directly with age[5]. The strict correlation with PDAC is suggested first of all by the fact that these lesions can be found in more than 80% of pancreas with invasive carcinoma[1,6], and by a reported multifocality in patients with PDAC familial history[7,8]. Usually, due to their small size (by definition < 0.5 cm), these lesions are classically found incidentally during histological examination and are not associated with clinical symptoms or specific signs. From the radiological point of view, PanINs are more often associated with acinar atrophy and/or fibrosis, but this correlation is not specific[9,10].

IPMN

IPMNs are grossly visible lesions with intraductal growth, papillary architecture, and mucous producing cells. The first definition of IPMNs was reported in 1994[11]. The median age of IPMN patients ranges from 60 to 66 years. They are more common in men than in women (ratio: 3/1.3 in Europe, 3/2.1 in United States, and 3/1.8 in Asia) and arise most frequently in the proximal pancreatic head and the "uncinatus" process[12]. Although rare, IPMN involving more than a pancreatic segment or even the entire pancreas have been described (Figure 1)[12]. It is estimated that IPMNs may require up to six years to become invasive, although such estimation may be affected by multiple biases, including the specific IPMN histotype[13-15]. Similarly to PanINs, IPMNs are found more frequently in patients with PDAC familial history, thus highlighting the importance of a genetic predisposition to carcinogenesis of IPMN patients. In fact, such precursors have been found also in the context of multi-organ syndromes such as Peutz-Jeghers, familial adenomatous polyposis, Lynch, and McCune-Albright syndromes[16-18]. Moreover, patients with IPMNs have an increased risk of developing other extrapancreatic cancers[19].

One of the most important distinctions between IPMNs and PanINs is the possibility that IPMNs may be detected with imaging techniques. Patients with IPMNs should be followed-up according to specific protocols based on radiological examination[20]. The typical intraductal growth of IPMNs leads to cystic dilation of the pancreatic tree ducts[21]. Based on their location, IPMNs may be classified from the topographic point of view in: (1) main duct IPMNs where Wirsung's duct is involved; (2) branch duct IPMNs in the case of the involvement of secondary ducts; or (3) mixed IPMNs in the case of contemporaneous involvement of the main and the branch ducts[1,20,21]. Although this distinction cannot always be confirmed by histopathological examination due to some branch duct IPMNs displaying some degree of Wirsung's duct involvement[22], this topographic definition has an important clinical impact. Indeed, main duct IPMNs are more often associated with PDAC development and patients with this type of lesion must follow stringent surveillance protocols[20]. IPMN-associated carcinomas usually display a better prognosis than conventional PDACs[1].

MCN

MCNs are typically reported in perimenopausal women, with few cases described in men[4]. They usually arise

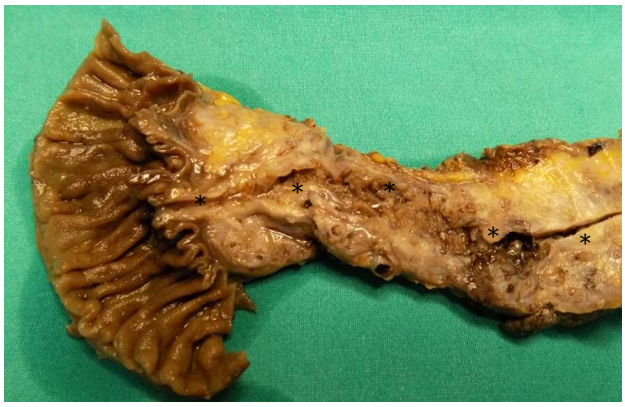


Figure 1 Extensive involvement of the pancreas by an intraductal papillary mucinous neoplasm. This neoplasm involves almost all the pancreatic ductal tree. The asterisks indicate Wirsung's duct along its course.

in the distal part of the pancreas (body and/or tail), and by definition do not communicate with the pancreatic ductal tree[1]. It has been hypothesized that the females may be predisposed to MCNs due to embryogenesis or by a carcinogenetic process stimulated by female hormones[23,24]. This theory is also corroborated by their histological aspect because under the mucinous, non-papillary epithelium is a classic ovarian-like stroma[1]. The mean age of patients with MCNs is about 44 years. The mean age of patients with MCNs with an associated adenocarcinoma is about 55 years[20]. This observation is in line with the status of MCN as a PDAC precursor lesion. The association with PDAC is present in up to one third of MCNs[25,26]. In contrast to PanINs, but similar to IPMNs, MCNs can be detected by imaging. MCN patients must undergo strict follow-up or pancreatic resection because there is a non-negligible risk of PDAC development[21].

ITPN

ITPNs are rare intraductal neoplasms of the pancreas composed of mucinous cells displaying a tubule-papillary architecture[1,5]. The incidence is similar in women and men[1]. These lesions are more commonly located in the head of the pancreas[1], and their symptoms are unspecific, including undefined abdominal pain and vomiting. Notably, about 40% of ITPNs harbor an associated invasive carcinoma[21]. PDACs arising in association with ITPNs usually have a better prognosis than that of conventional PDACs with a 5-year survival rate of more than 30%[21].

HISTOPATHOLOGY OF PRECURSOR LESIONS OF PANCREATIC CANCER

PanIN

PanINs are non-infiltrating microscopic intraductal lesions with a diameter < 0.5 cm[1,3]. From the histological point of view, they are composed of cuboid to columnar mucinous cells with varying degrees of dysplasia, reflecting the different degrees of cytologic and/or architectural atypia[1,3]. In the vast majority of cases, PanINs show gastric/foveolar differentiation[21]. Hruban *et al*[4] classified PanINs into a three-tiered scale, based on the degree of dysplasia. In this scheme, PanIN-1 shows low-grade dysplasia, PanIN-2 shows intermediate dysplasia and PanIN-3 shows high-grade dysplasia characterized by marked cell atypia, presence of mitotic figures, loss of polarity, and complex architecture. To improve inter-observer agreement and in order to report only the most important histological information, a recent consensus suggested a new classification system, distinguishing low-grade PanINs, which includes the previously called PanIN-1 and PanIN-2, and high-grade PanINs that includes PanIN-3 (Figure 2)[5]. In high-grade PanINs, cribriform structures, atypical mitosis, tufting of epithelial cells in the lumen, and even necrosis may be present, but in case these features are concomitant with a PDAC, the most important differential diagnosis of high-grade PanINs is with non-dysplastic ducts, which have been colonized by PDAC cells[27]. Notably, high-grade PanINs have been reported almost exclusively in association with an infiltrating PDAC[1,21]. However, a recent report pointed out that high-grade dysplasia PanINs may be found without concomitant infiltrating PDAC, and, when they involve the main duct, they may cause stenosis with extensive upstream duct dilation[28]. At the immunohistochemical level, PanINs show an increased expression of mucin 1 and mucin 5AC (MUC1 and MUC5AC) and a decreased expression of mucin 6 (MUC6)[29-32].

IPMN

IPMNs are non-infiltrating neoplasms > 1.0 cm with intraductal growth composed of mucinous cells with a papillary architecture[1,5]. Lesions with such features but with a size > 0.5 cm but < 1.0 cm should be classified as "incipient IPMN"[5]. Similarly to PanINs, a recent consensus suggested grading IPMNs into low-grade and high-grade and to avoid the intermediate-grade dysplasia, which should be included into the low-grade category[5]. IPMNs can be classified not only basing on topography (main duct, branch duct, or mixed type), but also from the histological point of view, based on the histotype of the predominant epithelium, which also influences their biological behavior: Gastric, pancreatobiliary, intestinal, and oncocytic[1,33-41] (Figure 3).

Gastric-type IPMNs usually do not involve the Wirsung's duct. They are composed of cells with the features of the gastric foveolar epithelium. There is a single layer of mucinous cells with polarized nuclei located at the basis of the cells. Usually this epithelium is associated with low-grade dysplasia. It can show a mixture of papillary, pseudopapillary, and flat structures[1]. When high-grade dysplasia is present in a gastric-type IPMN, with complex structure and atypical cells, the lesion becomes histologically very similar to a pancreatobiliary-type IPMN[33]. Questions are still open if these aspects represent different degrees of gastric-epithelial dyspla-

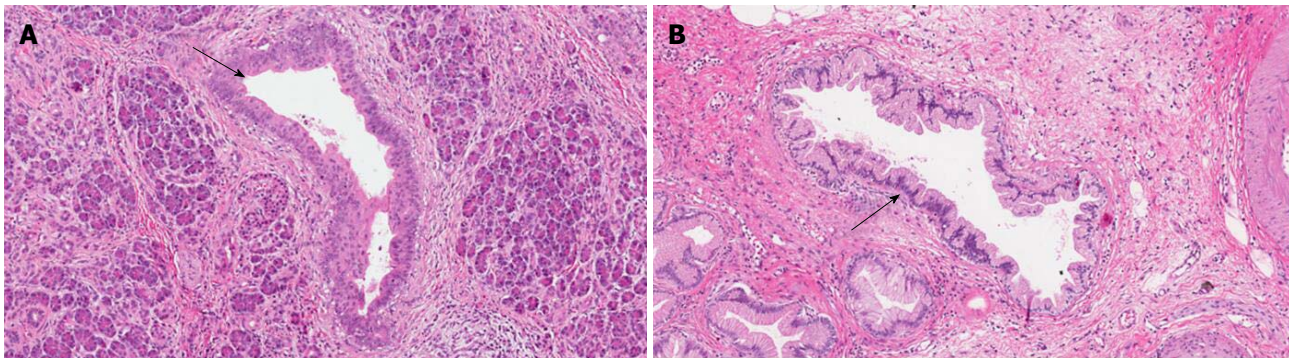


Figure 2 Pancreatic intraepithelial neoplasm precursor lesions. A: High-grade pancreatic intraepithelial neoplasm (PanIN); B: Low-grade PanIN. Original magnification: × 10. Black arrows indicate ducts involved by PanINs.

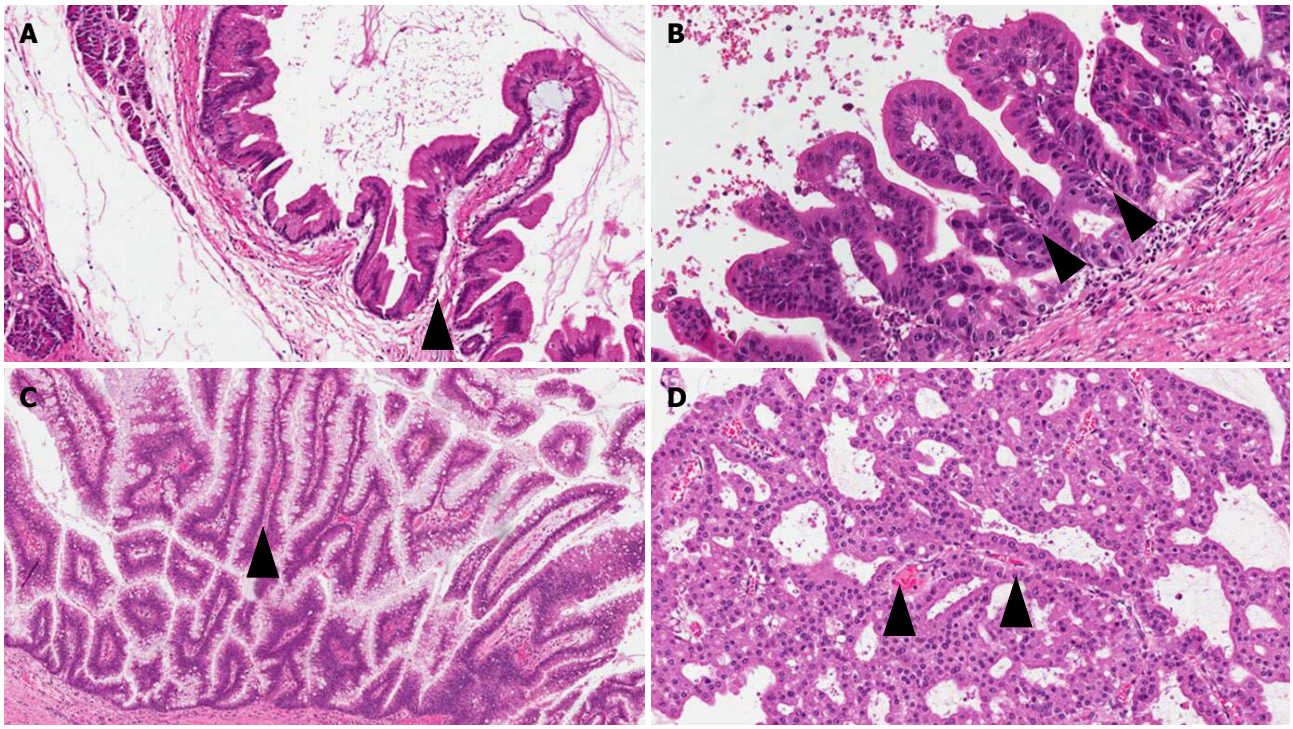


Figure 3 The four different types of intraductal papillary mucinous neoplasm. A: Gastric; B: Pancreatobiliary; C: Intestinal; D: Oncocytic. Original magnification: A and C: × 10; B and D: × 20. Black arrowheads indicate the fibro-vascular axis of the papillary structures.

sia, or could represent intratumor heterogeneity of IPMNs with low-grade gastric epithelium and high-grade pancreatobiliary epithelium coexisting in the same lesion[33-36].

Pancreatobiliary IPMNs usually involve the Wirsung's duct. They are composed of irregular cells usually with enlarged nuclei and prominent nucleoli. Typically, the dysplasia in this type of lesion is high-grade. Among the different IPMN subtypes, they have the highest risk to progress into PDAC[21,33-41]. Indeed, a recent meta-analysis including 14 studies for a total of 1617 patients, showed that pancreatobiliary IPMNs are associated with the most aggressive behavior, while gastric IPMNs display the lowest risk of cancer progression[37].

Intestinal IPMNs usually involve Wirsung's duct. They are histologically similar to villous adenoma of the large bowel. Their most evident morphological feature is represented by the presence of goblet cells, the papillae are long and sometimes branching, and the nuclei of the cells are hyperchromatic, elongated, and show different degrees of pseudostratification[1,21]. Although their risk is lower than pancreaticobiliary type, intestinal IPMNs can progress into invasive adenocarcinoma as well. Interestingly, the latter is not usually represented by a conventional adenocarcinoma but by a colloid carcinoma (Figure 4), which displays a better prognosis than conventional PDAC[1].

Oncocytic IPMNs are rare lesions, which may involve both main and branch ducts, or even the entire pancreatic ductal tree. They are composed of cells with a typical eosinophilic and granular cytoplasm due to the abundance of accumulated mitochondria[32]. Not only

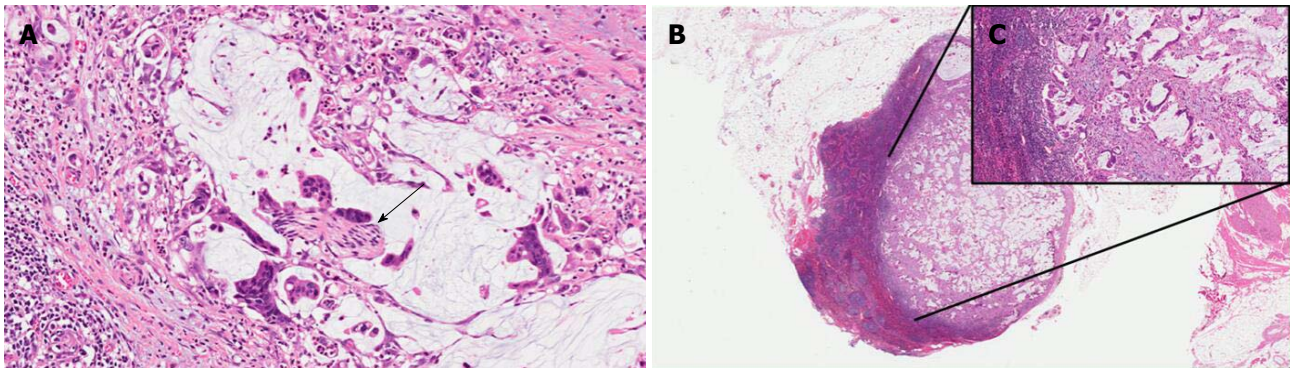


Figure 4 Colloid carcinoma with perineural invasion (A, black arrow) and nodal metastasis (B: low magnification; C: higher magnification of the same metastasis). Original magnification: A: × 10; B: × 4; C: × 20.

is the cytological appearance peculiar, but so is the architecture. Oncocytic IPMN form arborizing papillae, lined by one to five layers of cuboidal cells. A specific feature is represented by punched-out spaces in the epithelium[1,21,32].

The best strategy for pathologists to classify IPMN histotypes is coupling morphology with immunohistochemistry, particularly in the case of high-grade dysplasia. Immunohistochemistry based on mucin staining appears of great importance in this setting (Table 1)[1,33-41]. However, even with this integrated approach about 25% of cases are difficult to classify, mainly due to the presence of phenotypic heterogeneity or dedifferentiated areas[42].

MCN

MCNs are composed of columnar cells with abundant mucin located in the luminal part of the cells. The dysplasia of MCNs should be classified with a two-tiered scale (MCNs with low-grade including the previously called intermediate dysplasia, *vs* MCNs with high-grade dysplasia), following the recommendation of the latest consensus conference[5]. MCNs with low-grade dysplasia show mild cell atypia and lack of architectural complexity. MCNs with high-grade dysplasia are composed of atypical cells often with enlarged nuclei and multi-layer stratification (Figure 5). The diagnostic clues for the diagnosis of MCNs are represented by the lack of communication with the pancreatic ductal tree (always present in IPMNs), and the presence of an ovarian-like stroma located under the mucinous epithelium (Figure 5)[1,5,43]. These stromal cells usually exhibit immunostaining for progesterone and estrogen receptors as well as for alfa-inhibin[44,45]. In the case of an associated invasive adenocarcinoma, the latter is usually represented by a conventional PDAC[21].

ITPN

ITPNs are composed of relatively uniform and cuboidal cells, without a significant amount of mucin, arranged in densely packed tubules and back-to-back glands, with a typical intraductal, tubulopapillary growth (Figure 6)[1]. In this type of lesion, extracellular mucin production may be lacking or very focal with less common cyst formation as a direct consequence. An intestinal-type necrosis may also be present[46]. ITPNs are typically negative for MUC5AC, while MUC6 is often strongly positive (Table 1)[1,46]. Because of their potential progression to invasiveness as well as for their non-negligible association with PDAC, ITPNs are also considered a PDAC precursor lesion[1,21].

MOLECULAR PROFILES OF PRECURSOR LESIONS OF PANCREATIC CANCER

The study of the molecular landscape of PDAC precursor lesions has generated a growing body of knowledge, very useful not only to the comprehension of its oncogenesis but also to plan future strategies for their early detection. From the molecular point of view, *KRAS*, *TP53*, *CDKN2A,* and *SMAD4* represent the four major driver genes of PDAC[1,27], and it is of great interest the timing in which its precursors acquire alterations in such genes during their specific carcinogenesis. The most important aspect in this process, which is common to each precursor, is that a *KRAS* mutation is a fundamental and early event.

PanIN

The generally accepted definition of PanIN as a true precursor lesion of PDAC has been necessarily confirmed through their molecular characterization. Seminal research on this topic has showed that there is molecular evidence of the progression of PanIN towards PDAC. Early lesions (low-grade PanINs) display *KRAS* somatic mutations, and high-grade PanINs harbor *CDKN2A*, *TP53,* and *SMAD4* mutations[47-53]. In PanIN carcinogenesis, *TP53* and *SMAD4* inactivation appear as the latest events[53].

IPMN

A recent whole-exome sequencing study on IPMNs has showed an average of 26 mutated genes per case[54]. The most frequently mutated genes in IPMNs are *GNAS* and *KRAS*, which are altered in up to 60%

Table 1 Immunohistochemical markers for intraductal papillary mucinous neoplasm/intraductal tubulo-papillary neoplasm histopathological classification

Type of lesion	Subtype	MUC1	MUC2	MUC5AC	MUC6	CDX2
IPMN	G	Negative	Negative	Positive[1]	Negative	Negative
	PB	Positive[1]	Negative	Positive[1]	Positive	Negative
	INT	Negative	Positive[1]	Positive[1]	Negative	Positive[1]
	ONC	Positive	Negative	Positive	Positive[1]	Negative
ITPN		Positive	Negative	Negative	Positive[1]	Negative

IPMN: Intraductal papillary mucinous neoplasm; ITPN: Intraductal tubulo-papillary neoplasm; G: Gastric; PB: Pancreaticobiliary; INT: Intestinal; ONC: Oncocytic. [1]The positivity of a marker is very intense at the immunohistochemical level (++).

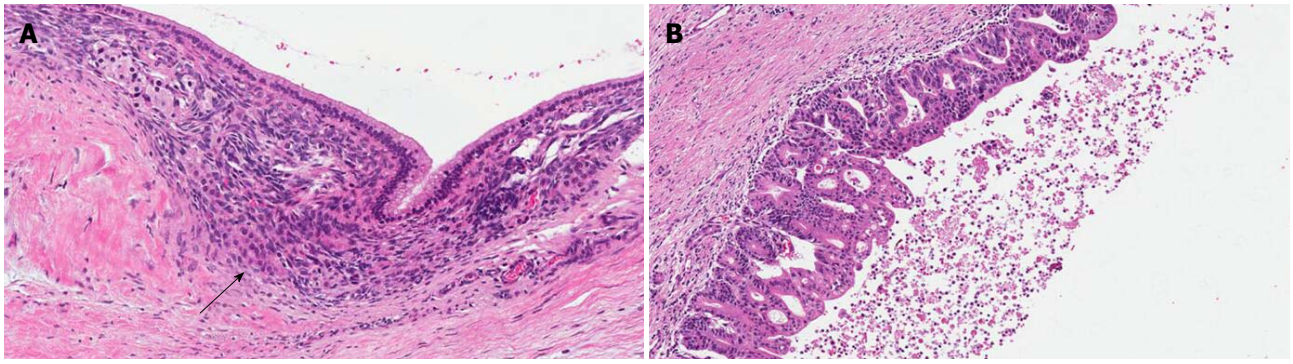


Figure 5 Mucinous cystic neoplasm precursor lesions. A: Low-grade mucinous cystic neoplasm (MCN); B: High-grade MCN. The black arrow indicates the ovarian-like stroma, a typical component of this type of lesion. Original magnification: × 10.

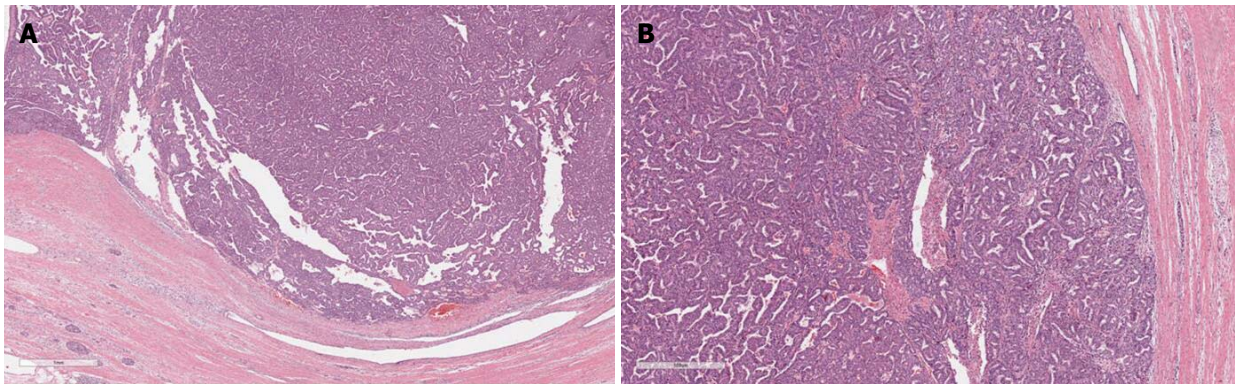


Figure 6 Intratubular papillary neoplasm precursor lesions. A: Low magnification showing an extensive intraductal growth; B: Higher magnification. Original magnification A: × 1; B: × 4.

and to 80% of cases, respectively[54,55]. Notably, recent studies pointed out that the carcinogenesis of IPMNs may follow two distinct pathways: The first, linked to *GNAS* mutations, are intestinal IPMNs progressing to colloid adenocarcinomas, and the second, linked to *KRAS* mutations, are typical of pancreatobiliary IPMNs and leads to conventional PDAC[56-59]. Another frequently mutated gene in IPMNs is *RNF43*, an E3 ubiquitin-protein ligase, which functions as a negative regulator of the Wnt-signaling pathway[54,60]. Lastly, *BRAF*, *TP53,* and *SMAD4* mutations can be found in IPMN with high-grade dysplasia. *TP53* and *SMAD4* mutations, similar to high-grade PanINs, are the latest molecular events in IPMN carcinogenesis[60].

MCN

A recent whole-exome sequencing study of MCNs revealed an average of 16 somatic mutations per case[54], and compared to IPMNs there was a lower percentage of cases with loss of heterozygosity events, a molecular feature associated to poor prognosis[54,61]. The fewer number of mutations and chromosomal alterations in MCNs could explain the lower frequency of progression to PDAC of this type of precursor when compared with IPMNs. In MCNs, somatic mutations involving the four classic PDAC driver genes (*KRAS*, *CDKN2A*, *TP53*, and

SMAD4) and *RNF43* have also been reported[54].

ITPN

This type of lesion has a peculiar molecular profile. Particularly, mutations involving *KRAS, NRAS,* and *GNAS* are very rare[60,62,63]. At the same time, *PIK3CA* mutations and *AKT* alterations (and consequently the involvement of the druggable mTOR pathway) are seen in 21% to 27% of ITPN cases[62,63].

RECENT ADVANCES AND FUTURE PERSPECTIVES ON PANCREATIC CARCINOGENESIS

Recent molecular advances in pancreatic carcinogenesis have given new interesting insights into the biological behavior of PDAC. The study of the genetic landscape of its precursor lesions has highlighted important implications for the early detection and for the clinical management of patients with pre-invasive and invasive pancreatic tumors.

PanIN

The most recent advances on the genetics of PanINs come from the study by Hosoda *et al*[64] of a series of isolated PanINs, *i.e.* those occurring in the absence of a concomitant PDAC. Whole-exome or targeted sequencing of 23 isolated high-grade PanINs found that *KRAS* mutations were present in the vast majority of lesions (> 90%), and *CDKN2A* and *RFN43* mutations were relatively frequent (about 20%-25% of cases), but other genes previously considered important in high-grade PanINs, *i.e., TP53*, *SMAD4*, *GNAS*, *ARID1A*, *PIK3CA,* and *TGFBR2* were very rarely mutated or not altered[64]. In the same study, 16 adjacent low-grade PanINs were sequenced showing very frequent *KRAS* mutations (> 90% of cases) and lack of mutations in *TP53*, *CDKN2A,* and *SMAD4* tumor suppressor genes[64]. The main conclusion of this paper was that mutations of *TP53* and *SMAD4* are events mainly associated with invasive PDAC and not with PanIN precursor lesions. Also the inactivation of chromatin remodeler genes, such as *ARID1A* tumor suppressor gene, previously thought to be important in PDAC and other invasive malignancies[65-68], appeared to be associated with infiltrating cancers rather than precursor lesions in the pancreas. The refinement of our knowledge on the morphological and molecular alterations of PanINs should be taken into account by future researchers in order to improve the possibilities of PDAC early detection.

IPMN

The clinical management of IPMNs has changed in the last decade. The most recent guidelines indicate the need of combining clinical and radiological information in order to define the best therapeutic choice. Particular features, whose presence has different implications, have been distinguished in IPMN patients and indicated as "high-risk stigmata" and "worrisome features"[69]. The "high-risk stigmata" are represented by: (1) obstructive jaundice in a patient with cystic lesion of the head of the pancreas; (2) enhancing mural nodule > 5 mm; and (3) main pancreatic duct > 10 mm. The "worrisome features" comprise of: (a) clinical pancreatitis; (b) cyst > 3 cm, (c) enhancing mural nodule < 5 mm; (d) thickened/enhancing cyst walls; (e) main duct size 5–9 mm; (f) abrupt change in caliber of pancreatic duct with distal pancreatic atrophy; (g) lymphadenopathy; (h) increased serum level of CA19-9; and (i) cyst growth rate > 5 mm/2 years[69]. Thus, patients with IPMN should be followed-up with a stringent protocol, which integrates imaging (endoscopic ultrasonography, computed tomography, and magnetic resonance) and clinical data, on the basis of their importance and their specific risk of progression to invasive cancer. From the molecular point of view, the recent advances in this field have provided interesting information from the genetic analysis of cyst fluids[70]. Future protocols should integrate clinical-radiological information with molecular data, to obtain for each patient an integrated estimation of the risk of PDAC development. This approach, however, should also take into account the issue of field-effect carcinogenesis of PDAC. Indeed, IPMNs and PDACs are not necessarily genetically related as recently reported by Felsenstein *et al*[71], who demonstrated that about 20% of coexisting IPMNs and PDACs are molecularly unrelated, indicating the possibility of PDAC development independent from a coexisting IPMN. The main implication of this research regards the strategy of surveillance of patients with IPMN[72]. Also, the local recurrence of IPMN or PDAC after pancreatic resection for a IPMN may be genetically unrelated, highlighting the existence of a field-effect carcinogenesis of PDAC[73].

MCN and ITPN

The clinical management of patients with MCNs and ITPNs should take into account recent molecular knowledge. MCNs and ITPNs represent true PDAC precursor lesions, thus an integrated approach with clinical-radiological information and molecular data should be implemented in order to define stringent protocols for the surveillance of low-risk subjects as well as precise parameters indicating the need of pancreatic resection in high-risk patients.

Cell of origin of pancreatic cancer

Another recent fascinating advance in pancreatic carcinogenesis are the putative cells of origin of this tumor type. Indeed, recent evidence from engineered mice-models suggests that PanIN development seems to be the result of the transdifferentiation of acinar cells, while IPMNs seem to arise from the progenitor niche of the pancreatic ductal epithelium[74-76]. These new concepts have totally changed the previous convictions, which indicated the differentiated ductal cells as the progenitor of

Table 2 Precursor lesions and their most important histopathological and molecular features

Precursor lesions	Main histopathological features	Molecular hallmarks
PanIN	Non-infiltrating lesions involving pancreatic ducts and < 0.5 cm, composed of cuboid to columnar mucinous cells, with two degrees of dysplasia: (1) Low-grade PanINs include the previously called PanIN-1 and PanIN-2; and (2) high-grade PanINs include PanIN-3	*KRAS* somatic mutations are early molecular events (Low-grade PanINs); *CDKN2A*, *TP53*, and *SMAD4* mutations are late molecular events (High-grade PanINs)
IPMN	Non-infiltrating intraductal neoplasms > 1.0 cm composed of mucinous cells with papillary architecture. IPMNs have two degrees of dysplasia: (1) Low-grade IPMNs; and (2) High-grade IPMNs. IPMNs can be classified based on topography (main duct, branch duct or mixed) and also on histology (gastric, pancreaticobiliary, intestinal, or oncocytic type, see Table 1)	*GNAS* and *KRAS* are altered in up to 60% and to 80% of IPMNs, respectively There are two possible carcinogenetic processes: (1) *GNAS* mutations cause progression to colloid carcinomas; and (2) *KRAS* mutations lead to conventional PDAC. Other frequently mutated genes in IPMNs are *RNF43*, *BRAF*, *TP53*, and *SMAD4*
MCN	Composed of columnar cells with abundant mucin located in the upper part. There are two degrees of dysplasia: (1) Low-grade MCN; and (2) High-grade MCN. The histopathologic clues for MCN diagnosis are the lack of communication with the pancreatic ductal tree and the presence of an ovarian-like stroma under the mucinous epithelium	There are fewer mutations and chromosomal alterations in MCNs compared with other precursors, and this could explain the lower frequency of progression of MCN to PDAC. Frequently altered genes are *KRAS*, *CDKN2A*, *TP53*, *SMAD4*, and *RNF43*
ITPN	Composed of uniform cuboidal cells without a significant amount of mucin, arranged in densely packed tubules and back-to-back glands with a typical intraductal, tubulopapillary growth	*PIK3CA* mutations and *AKT* alterations are frequently seen in ITPNs. Mutations involving *KRAS*, *NRAS*, and *GNAS* are very rare in ITPNs

PanIN: Pancreatic intraepithelial neoplasm; IPMN: Intraductal papillary mucinous neoplasm; MCN: Mucinous cystic neoplasm; ITPN: Intratubular papillary neoplasm; PDAC: Pancreatic ductal adenocarcinoma.

PDAC. Understanding these two different pathways of PDAC carcinogenesis, one starting from acinar epithelium and one from ductal epithelium, could also partly explain the different biological behaviors of PanINs and IPMNs and their progression into an overt PDAC[74-76].

CONCLUSION

The histopathological and molecular features of PDAC precursor lesions have been summarized in Table 2 to provide a complete vision on this important topic. They represent a fundamental issue for the comprehension of PDAC carcinogenesis and its biological behavior. Only an integrated approach coupling histopathology and molecular analysis may guarantee a decisive step for the early detection of PDAC and to design more effective therapeutic strategies.

REFERENCES

1 **Bosman FT**, Carneiro F, Hruban RH, Theise ND. WHO classification of tumours of the digestive system. Lyon: IARC Press, 2010

2 **Hruban RH**, Takaori K, Klimstra DS, Adsay NV, Albores-Saavedra J, Biankin AV, Biankin SA, Compton C, Fukushima N, Furukawa T, Goggins M, Kato Y, Klöppel G, Longnecker DS, Lüttges J, Maitra A, Offerhaus GJ, Shimizu M, Yonezawa S. An illustrated consensus on the classification of pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasms. *Am J Surg Pathol* 2004; **28**: 977-987 [PMID: 15252303 DOI: 10.1097/01.pas.0000126675.59108.80]

3 **Furukawa T**, Klöppel G, Volkan Adsay N, Albores-Saavedra J, Fukushima N, Horii A, Hruban RH, Kato Y, Klimstra DS, Longnecker DS, Lüttges J, Offerhaus GJ, Shimizu M, Sunamura M, Suriawinata A, Takaori K, Yonezawa S. Classification of types of intraductal papillary-mucinous neoplasm of the pancreas: a consensus study. *Virchows Arch* 2005; **447**: 794-799 [PMID: 16088402 DOI: 10.1007/s00428-005-0039-7]

4 **Hruban RH**, Adsay NV, Albores-Saavedra J, Compton C, Garrett ES, Goodman SN, Kern SE, Klimstra DS, Klöppel G, Longnecker DS, Lüttges J, Offerhaus GJ. Pancreatic intraepithelial neoplasia: a new nomenclature and classification system for pancreatic duct lesions. *Am J Surg Pathol* 2001; **25**: 579-586 [PMID: 11342768 DOI: 10.1097/00000478-200105000-00003]

5 **Basturk O**, Hong SM, Wood LD, Adsay NV, Albores-Saavedra J, Biankin AV, Brosens LA, Fukushima N, Goggins M, Hruban RH, Kato Y, Klimstra DS, Klöppel G, Krasinskas A, Longnecker DS, Matthaei H, Offerhaus GJ, Shimizu M, Takaori K, Terris B, Yachida S, Esposito I, Furukawa T; Baltimore Consensus Meeting. A Revised Classification System and Recommendations From the Baltimore Consensus Meeting for Neoplastic Precursor Lesions in the Pancreas. *Am J Surg Pathol* 2015; **39**: 1730-1741 [PMID: 26559377 DOI: 10.1097/PAS.0000000000000533]

6 **Andea A**, Sarkar F, Adsay VN. Clinicopathological correlates of pancreatic intraepithelial neoplasia: a comparative analysis of 82 cases with and 152 cases without pancreatic ductal adenocarcinoma. *Mod Pathol* 2003; **16**: 996-1006 [PMID: 14559982 DOI: 10.1097/01.MP.0000087422.24733.62]

7 **Brune K**, Abe T, Canto M, O'Malley L, Klein AP, Maitra A, Volkan Adsay N, Fishman EK, Cameron JL, Yeo CJ, Kern SE, Goggins M, Hruban RH. Multifocal neoplastic precursor lesions associated with lobular atrophy of the pancreas in patients having a strong family history of pancreatic cancer. *Am J Surg Pathol* 2006; **30**: 1067-1076 [PMID: 16931950]

8 **Shi C**, Klein AP, Goggins M, Maitra A, Canto M, Ali S, Schulick R, Palmisano E, Hruban RH. Increased Prevalence of Precursor Lesions in Familial Pancreatic Cancer Patients. *Clin Cancer Res* 2009; **15**: 7737-7743 [PMID: 19996207 DOI: 10.1158/1078-0432.CCR-09-0004]

9 **Takaori K**, Matsusue S, Fujikawa T, Kobashi Y, Ito T, Matsuo Y, Oishi H, Takeda H. Carcinoma in situ of the pancreas associated with localized fibrosis: a clue to early detection of neoplastic lesions arising from pancreatic ducts. *Pancreas* 1998; **17**: 102-105 [PMID: 9667529 DOI: 10.1097/00006676-199807000-00015]

10 **Detlefsen S**, Sipos B, Feyerabend B, Klöppel G. Pancreatic fibrosis associated with age and ductal papillary hyperplasia. *Virchows Arch* 2005; **447**: 800-805 [PMID: 16021508 DOI: 10.1007/s00428-005-0032-1]

11 **Morohoshi T**, Kanda M, Asanuma K, Klöppel G. Intraductal papillary neoplasms of the pancreas. A clinicopathologic study of six patients. *Cancer* 1989; **64**: 1329-1335 [PMID: 2548703 DOI: 10.1002/1097-0142(19890915)64:6<1329::AID-CNCR28-20640627>3.0.CO;2-S]

12 **Ingkakul T**, Warshaw AL, Fernández-Del Castillo C. Epidemiology of intraductal papillary mucinous neoplasms of the pancreas: sex differences between 3 geographic regions. *Pancreas* 2011; **40**: 779-780 [PMID: 21673537 DOI: 10.1097/MPA.0b013e31821f27fb]

13 **Ingkakul T**, Sadakari Y, Ienaga J, Satoh N, Takahata S, Tanaka M. Predictors of the presence of concomitant invasive ductal carcinoma in intraductal papillary mucinous neoplasm of the pancreas. *Ann Surg* 2010; **251**: 70-75 [PMID: 20009749 DOI: 10.1097/SLA.0b013e3181c5ddc3]

14 **Chari ST**, Yadav D, Smyrk TC, DiMagno EP, Miller LJ, Raimondo M, Clain JE, Norton IA, Pearson RK, Petersen BT, Wiersema MJ, Farnell MB, Sarr MG. Study of recurrence after surgical resection of intraductal papillary mucinous neoplasm of the pancreas. *Gastroenterology* 2002; **123**: 1500-1507 [PMID: 12404225 DOI: 10.1053/gast.2002.36552]

15 **Salvia R**, Fernández-del Castillo C, Bassi C, Thayer SP, Falconi M, Mantovani W, Pederzoli P, Warshaw AL. Main-duct intraductal papillary mucinous neoplasms of the pancreas: clinical predictors of malignancy and long-term survival following resection. *Ann Surg* 2004; **239**: 678-685; discussion 685-687 [PMID: 15082972 DOI: 10.1097/01.sla.0000124386.54496.15]

16 **Sparr JA**, Bandipalliam P, Redston MS, Syngal S. Intraductal papillary mucinous neoplasm of the pancreas with loss of mismatch repair in a patient with Lynch syndrome. *Am J Surg Pathol* 2009; **33**: 309-312 [PMID: 18987546 DOI: 10.1097/PAS.0b013e3181882c3d]

17 **Su GH**, Hruban RH, Bansal RK, Bova GS, Tang DJ, Shekher MC, Westerman AM, Entius MM, Goggins M, Yeo CJ, Kern SE. Germline and somatic mutations of the STK11/LKB1 Peutz-Jeghers gene in pancreatic and biliary cancers. *Am J Pathol* 1999; **154**: 1835-1840 [PMID: 10362809 DOI: 10.1016/S0002-9440(10)65440-5]

18 **Gaujoux S**, Chanson P, Bertherat J, Sauvanet A, Ruszniewski P. Hepato-pancreato-biliary lesions are present in both Carney complex and McCune Albright syndrome: comments on P. Salpea and C. Stratakis. *Mol Cell Endocrinol* 2014; **382**: 344-345 [PMID: 24161590 DOI: 10.1016/j.mce.2013.10.020]

19 **Panic N**, Macchini F, Solito S, Boccia S, Leoncini E, Larghi A, Berretti D, Pevere S, Vadala S, Marino M, Zilli M, Bulajic M. Prevalence of Extrapancreatic Malignancies Among Patients With Intraductal Papillary Mucinous Neoplasms of the Pancreas. *Pancreas* 2018; **47**: 721-724 [PMID: 29771766 DOI: 10.1097/MPA.0000000000001072]

20 **Tanaka M**. Intraductal Papillary Mucinous Neoplasm of the Pancreas as the Main Focus for Early Detection of Pancreatic Adenocarcinoma. *Pancreas* 2018; **47**: 544-550 [PMID: 29702531 DOI: 10.1097/MPA.0000000000001047]

21 **Noë M**, Brosens LA. Pathology of Pancreatic Cancer Precursor Lesions. *Surg Pathol Clin* 2016; **9**: 561-580 [PMID: 27926360 DOI: 10.1016/j.path.2016.05.004]

22 **Fritz S**, Klauss M, Bergmann F, Strobel O, Schneider L, Werner J, Hackert T, Büchler MW. Pancreatic main-duct involvement in branch-duct IPMNs: an underestimated risk. *Ann Surg* 2014; **260**: 848-55; discussion 855-856 [PMID: 25379856 DOI: 10.1097/SLA.0000000000000980]

23 **Zamboni G**, Scarpa A, Bogina G, Iacono C, Bassi C, Talamini G, Sessa F, Capella C, Solcia E, Rickaert F, Mariuzzi GM, Klöppel G. Mucinous cystic tumors of the pancreas: clinicopathological features, prognosis, and relationship to other mucinous cystic tumors. *Am J Surg Pathol* 1999; **23**: 410-422 [PMID: 10199470 DOI: 10.1097/00000478-199904000-00005]

24 **Ridder GJ**, Maschek H, Flemming P, Nashan B, Klempnauer J. Ovarian-like stroma in an invasive mucinous cystadenocarcinoma of the pancreas positive for inhibin. A hint concerning its possible histogenesis. *Virchows Arch* 1998; **432**: 451-454 [PMID: 9645445 DOI: 10.1007/s004280050190]

25 **Reddy RP**, Smyrk TC, Zapiach M, Levy MJ, Pearson RK, Clain JE, Farnell MB, Sarr MG, Chari ST. Pancreatic mucinous cystic neoplasm defined by ovarian stroma: demographics, clinical features, and prevalence of cancer. *Clin Gastroenterol Hepatol* 2004; **2**: 1026-1031 [PMID: 15551256 DOI: 10.1016/S1542-3565(04)00450-1]

26 **Baker ML**, Seeley ES, Pai R, Suriawinata AA, Mino-Kenudson M, Zamboni G, Klöppel G, Longnecker DS. Invasive mucinous cystic neoplasms of the pancreas. *Exp Mol Pathol* 2012; **93**: 345-349 [PMID: 22902940 DOI: 10.1016/j.yexmp.2012.07.005]

27 **Luchini C**, Capelli P, Scarpa A. Pancreatic Ductal Adenocarcinoma and Its Variants. *Surg Pathol Clin* 2016; **9**: 547-560 [PMID: 27926359 DOI: 10.1016/j.path.2016.05.003]

28 **Yokode M**, Akita M, Fujikura K, Kim MJ, Morinaga Y, Yoshikawa S, Terada T, Matsukiyo H, Tajiri T, Abe-Suzuki S, Itoh T, Hong SM, Zen Y. High-grade PanIN presenting with localised stricture of the main pancreatic duct: A clinicopathological and molecular study of 10 cases suggests a clue for the early detection of pancreatic cancer. *Histopathology* 2018; **73**: 247-258 [PMID: 29660164 DOI: 10.1111/his.13629]

29 **Nagata K**, Horinouchi M, Saitou M, Higashi M, Nomoto M, Goto M, Yonezawa S. Mucin expression profile in pancreatic cancer and the precursor lesions. *J Hepatobiliary Pancreat Surg* 2007; **14**: 243-254 [PMID: 17520199 DOI: 10.1007/s00534-006-1169-2]

30 **Adsay NV**, Merati K, Andea A, Sarkar F, Hruban RH, Wilentz RE, Goggins M, Iocobuzio-Donahue C, Longnecker DS, Klimstra DS. The dichotomy in the preinvasive neoplasia to invasive carcinoma sequence in the pancreas: differential expression of MUC1 and MUC2 supports the existence of two separate pathways of carcinogenesis. *Mod Pathol* 2002; **15**: 1087-1095 [PMID: 12379756 DOI: 10.1097/01.MP.0000028647.98725.8B]

31 **Moriya T**, Kimura W, Semba S, Sakurai F, Hirai I, Ma J, Fuse A, Maeda K, Yamakawa M. Biological similarities and differences between pancreatic intraepithelial neoplasias and intraductal papillary mucinous neoplasms. *Int J Gastrointest Cancer* 2005; **35**: 111-119 [PMID: 15879625 DOI: 10.1385/IJGC:35:2:111]

32 **Basturk O**, Khayyata S, Klimstra DS, Hruban RH, Zamboni G, Coban I, Adsay NV. Preferential expression of MUC6 in oncocytic and pancreatobiliary types of intraductal papillary neoplasms highlights a pyloropancreatic pathway, distinct from the intestinal pathway, in pancreatic carcinogenesis. *Am J Surg Pathol* 2010; **34**: 364-370 [PMID: 20139757 DOI: 10.1097/PAS.0b013e3181cf8bb6]

33 **Furukawa T**, Hatori T, Fujita I, Yamamoto M, Kobayashi M, Ohike N, Morohoshi T, Egawa S, Unno M, Takao S, Osako M, Yonezawa S, Mino-Kenudson M, Lauwers GY, Yamaguchi H, Ban S, Shimizu M. Prognostic relevance of morphological types of intraductal papillary mucinous neoplasms of the pancreas. *Gut* 2011; **60**: 509-516 [PMID: 21193453 DOI: 10.1136/gut.2010.210567]

34 **Mino-Kenudson M**, Fernández-del Castillo C, Baba Y, Valsangkar NP, Liss AS, Hsu M, Correa-Gallego C, Ingkakul T, Perez Johnston R, Turner BG, Androutsopoulos V, Deshpande V, McGrath D, Sahani DV, Brugge WR, Ogino S, Pitman MB, Warshaw AL, Thayer SP. Prognosis of invasive intraductal papillary mucinous neoplasm depends on histological and precursor epithelial subtypes. *Gut* 2011; **60**: 1712-1720 [PMID: 21508421 DOI: 10.1136/gut.2010.232272]

35 **Distler M**, Kersting S, Niedergethmann M, Aust DE, Franz M, Rückert F, Ehehalt F, Pilarsky C, Post S, Saeger HD, Grützmann R. Pathohistological subtype predicts survival in patients with intraductal papillary mucinous neoplasm (IPMN) of the pancreas. *Ann Surg* 2013; **258**: 324-330 [PMID: 23532107 DOI: 10.1097/SLA.0b013e318287ab73]

36 **Kim J**, Jang KT, Mo Park S, Lim SW, Kim JH, Lee KH, Lee JK, Heo JS, Choi SH, Choi DW, Rhee JC, Lee KT. Prognostic relevance of pathologic subtypes and minimal invasion in intraductal papillary mucinous neoplasms of the pancreas. *Tumour Biol* 2011; **32**: 535-542 [PMID: 21190101 DOI: 10.1007/s13277-010-0148-z]

37 **Koh YX**, Zheng HL, Chok AY, Tan CS, Wyone W, Lim TK, Tan DM, Goh BK. Systematic review and meta-analysis of the spectrum and outcomes of different histologic subtypes of noninvasive and invasive intraductal papillary mucinous neoplasms. *Surgery* 2015; **157**: 496-509 [PMID: 25656693 DOI: 10.1016/j.surg.2014.08.098]

38 **Nakata K**, Ohuchida K, Aishima S, Sadakari Y, Kayashima T,

Miyasaka Y, Nagai E, Mizumoto K, Tanaka M, Tsuneyoshi M, Oda Y. Invasive carcinoma derived from intestinal-type intraductal papillary mucinous neoplasm is associated with minimal invasion, colloid carcinoma, and less invasive behavior, leading to a better prognosis. *Pancreas* 2011; **40**: 581-587 [PMID: 21499213 DOI: 10.1097/MPA.0b013e318214fa86]

39 **Sadakari Y**, Ohuchida K, Nakata K, Ohtsuka T, Aishima S, Takahata S, Nakamura M, Mizumoto K, Tanaka M. Invasive carcinoma derived from the nonintestinal type intraductal papillary mucinous neoplasm of the pancreas has a poorer prognosis than that derived from the intestinal type. *Surgery* 2010; **147**: 812-817 [PMID: 20060146 DOI: 10.1016/j.surg.2009.11.011]

40 **Yonezawa S**, Higashi M, Yamada N, Yokoyama S, Goto M. Significance of mucin expression in pancreatobiliary neoplasms. *J Hepatobiliary Pancreat Sci* 2010; **17**: 108-124 [PMID: 19787286 DOI: 10.1007/s00534-009-0174-7]

41 **Kobayashi M**, Fujinaga Y, Ota H. Reappraisal of the Immunophenotype of Pancreatic Intraductal Papillary Mucinous Neoplasms (IPMNs)-Gastric Pyloric and Small Intestinal Immunophenotype Expression in Gastric and Intestinal Type IPMNs-. *Acta Histochem Cytochem* 2014; **47**: 45-57 [PMID: 25221363 DOI: 10.1267/ahc.13027]

42 **Schaberg KB**, DiMaio MA, Longacre TA. Intraductal Papillary Mucinous Neoplasms Often Contain Epithelium From Multiple Subtypes and/or Are Unclassifiable. *Am J Surg Pathol* 2016; **40**: 44-50 [PMID: 26469398 DOI: 10.1097/PAS.0000000000000528]

43 **Tanaka M**, Fernández-del Castillo C, Adsay V, Chari S, Falconi M, Jang JY, Kimura W, Levy P, Pitman MB, Schmidt CM, Shimizu M, Wolfgang CL, Yamaguchi K, Yamao K; International Association of Pancreatology. International consensus guidelines 2012 for the management of IPMN and MCN of the pancreas. *Pancreatology* 2012; **12**: 183-197 [PMID: 22687371 DOI: 10.1016/j.pan.2012.04.004]

44 **Shiono S**, Suda K, Nobukawa B, Arakawa A, Yamasaki S, Sasahara N, Hosokawa Y, Suzuki F. Pancreatic, hepatic, splenic, and mesenteric mucinous cystic neoplasms (MCN) are lumped together as extra ovarian MCN. *Pathol Int* 2006; **56**: 71-77 [PMID: 16445818 DOI: 10.1111/j.1440-1827.2006.01926.x]

45 **Izumo A**, Yamaguchi K, Eguchi T, Nishiyama K, Yamamoto H, Yonemasu H, Yao T, Tanaka M, Tsuneyoshi M. Mucinous cystic tumor of the pancreas: immunohistochemical assessment of "ovarian-type stroma". *Oncol Rep* 2003; **10**: 515-525 [PMID: 12684617]

46 **Yamaguchi H**, Shimizu M, Ban S, Koyama I, Hatori T, Fujita I, Yamamoto M, Kawamura S, Kobayashi M, Ishida K, Morikawa T, Motoi F, Unno M, Kanno A, Satoh K, Shimosegawa T, Orikasa H, Watanabe T, Nishimura K, Ebihara Y, Koike N, Furukawa T. Intraductal tubulopapillary neoplasms of the pancreas distinct from pancreatic intraepithelial neoplasia and intraductal papillary mucinous neoplasms. *Am J Surg Pathol* 2009; **33**: 1164-1172 [PMID: 19440145 DOI: 10.1097/PAS.0b013e3181a162e5]

47 **Klimstra DS**, Longnecker DS. K-ras mutations in pancreatic ductal proliferative lesions. *Am J Pathol* 1994; **145**: 1547-1550 [PMID: 7992857]

48 **Lemoine NR**, Jain S, Hughes CM, Staddon SL, Maillet B, Hall PA, Klöppel G. Ki-ras oncogene activation in preinvasive pancreatic cancer. *Gastroenterology* 1992; **102**: 230-236 [PMID: 1309358 DOI: 10.1016/0016-5085(92)91805-E]

49 **Cooper CL**, O'Toole SA, Kench JG. Classification, morphology and molecular pathology of premalignant lesions of the pancreas. *Pathology* 2013; **45**: 286-304 [PMID: 23442735 DOI: 10.1097/PAT.0b013e32835f2205]

50 **DiGiuseppe JA**, Hruban RH, Offerhaus GJ, Clement MJ, van den Berg FM, Cameron JL, van Mansfeld AD. Detection of K-ras mutations in mucinous pancreatic duct hyperplasia from a patient with a family history of pancreatic carcinoma. *Am J Pathol* 1994; **144**: 889-895 [PMID: 8178941]

51 **Delpu Y**, Hanoun N, Lulka H, Sicard F, Selves J, Buscail L, Torrisani J, Cordelier P. Genetic and epigenetic alterations in pancreatic carcinogenesis. *Curr Genomics* 2011; **12**: 15-24 [PMID: 21886451 DOI: 10.2174/138920211794520132]

52 **Moskaluk CA**, Hruban RH, Kern SE. p16 and K-ras gene mutations in the intraductal precursors of human pancreatic adenocarcinoma. *Cancer Res* 1997; **57**: 2140-2143 [PMID: 9187111]

53 **Brosens LA**, Hackeng WM, Offerhaus GJ, Hruban RH, Wood LD. Pancreatic adenocarcinoma pathology: changing "landscape". *J Gastrointest Oncol* 2015; **6**: 358-374 [PMID: 26261723 DOI: 10.3978/j.issn.2078-6891.2015.032]

54 **Wu J**, Jiao Y, Dal Molin M, Maitra A, de Wilde RF, Wood LD, Eshleman JR, Goggins MG, Wolfgang CL, Canto MI, Schulick RD, Edil BH, Choti MA, Adsay V, Klimstra DS, Offerhaus GJ, Klein AP, Kopelovich L, Carter H, Karchin R, Allen PJ, Schmidt CM, Naito Y, Diaz LA Jr, Kinzler KW, Papadopoulos N, Hruban RH, Vogelstein B. Whole-exome sequencing of neoplastic cysts of the pancreas reveals recurrent mutations in components of ubiquitin-dependent pathways. *Proc Natl Acad Sci USA* 2011; **108**: 21188-21193 [PMID: 22158988 DOI: 10.1073/pnas.1118046108]

55 **Wu J**, Matthaei H, Maitra A, Dal Molin M, Wood LD, Eshleman JR, Goggins M, Canto MI, Schulick RD, Edil BH, Wolfgang CL, Klein AP, Diaz LA Jr, Allen PJ, Schmidt CM, Kinzler KW, Papadopoulos N, Hruban RH, Vogelstein B. Recurrent GNAS mutations define an unexpected pathway for pancreatic cyst development. *Sci Transl Med* 2011; **3**: 92ra66 [PMID: 21775669 DOI: 10.1126/scitranslmed.3002543]

56 **Tan MC**, Basturk O, Brannon AR, Bhanot U, Scott SN, Bouvier N, LaFemina J, Jarnagin WR, Berger MF, Klimstra D, Allen PJ. GNAS and KRAS Mutations Define Separate Progression Pathways in Intraductal Papillary Mucinous Neoplasm-Associated Carcinoma. *J Am Coll Surg* 2015; **220**: 845-854.e1 [PMID: 25840541 DOI: 10.1016/j.jamcollsurg.2014.11.029]

57 **Hosoda W**, Sasaki E, Murakami Y, Yamao K, Shimizu Y, Yatabe Y. GNAS mutation is a frequent event in pancreatic intraductal papillary mucinous neoplasms and associated adenocarcinomas. *Virchows Arch* 2015; **466**: 665-674 [PMID: 25796395 DOI: 10.1007/s00428-015-1751-6]

58 **Molin MD**, Matthaei H, Wu J, Blackford A, Debeljak M, Rezaee N, Wolfgang CL, Butturini G, Salvia R, Bassi C, Goggins MG, Kinzler KW, Vogelstein B, Eshleman JR, Hruban RH, Maitra A. Clinicopathological correlates of activating GNAS mutations in intraductal papillary mucinous neoplasm (IPMN) of the pancreas. *Ann Surg Oncol* 2013; **20**: 3802-3808 [PMID: 23846778 DOI: 10.1245/s10434-013-3096-1]

59 **Yamada M**, Sekine S, Ogawa R, Taniguchi H, Kushima R, Tsuda H, Kanai Y. Frequent activating GNAS mutations in villous adenoma of the colorectum. *J Pathol* 2012; **228**: 113-118 [PMID: 22374786 DOI: 10.1002/path.4012]

60 **Amato E**, Molin MD, Mafficini A, Yu J, Malleo G, Rusev B, Fassan M, Antonello D, Sadakari Y, Castelli P, Zamboni G, Maitra A, Salvia R, Hruban RH, Bassi C, Capelli P, Lawlor RT, Goggins M, Scarpa A. Targeted next-generation sequencing of cancer genes dissects the molecular profiles of intraductal papillary neoplasms of the pancreas. *J Pathol* 2014; **233**: 217-227 [PMID: 24604757 DOI: 10.1002/path.4344]

61 **Southern JF**, Warshaw AL, Lewandrowski KB. DNA ploidy analysis of mucinous cystic tumors of the pancreas. Correlation of aneuploidy with malignancy and poor prognosis. *Cancer* 1996; **77**: 58-62 [PMID: 8630940 DOI: 10.1002/(SICI)1097-0142(19960101)77:1<58::AID-CNCR11>3.0.CO;2-7]

62 **Yamaguchi H**, Kuboki Y, Hatori T, Yamamoto M, Shiratori K, Kawamura S, Kobayashi M, Shimizu M, Ban S, Koyama I, Higashi M, Shin N, Ishida K, Morikawa T, Motoi F, Unno M, Kanno A, Satoh K, Shimosegawa T, Orikasa H, Watanabe T, Nishimura K, Harada Y, Furukawa T. Somatic mutations in PIK3CA and activation of AKT in intraductal tubulopapillary neoplasms of the pancreas. *Am J Surg Pathol* 2011; **35**: 1812-1817 [PMID: 21945955 DOI: 10.1097/PAS.0b013e31822769a0]

63 **Yamaguchi H**, Kuboki Y, Hatori T, Yamamoto M, Shimizu K, Shiratori K, Shibata N, Shimizu M, Furukawa T. The discrete nature and distinguishing molecular features of pancreatic intraductal tubulopapillary neoplasms and intraductal papillary mucinous neoplasms of the gastric type, pyloric gland variant. *J Pathol* 2013;

231: 335-341 [PMID: 23893889 DOI: 10.1002/path.4242]
64 **Hosoda W**, Chianchiano P, Griffin JF, Pittman ME, Brosens LA, Noë M, Yu J, Shindo K, Suenaga M, Rezaee N, Yonescu R, Ning Y, Albores-Saavedra J, Yoshizawa N, Harada K, Yoshizawa A, Hanada K, Yonehara S, Shimizu M, Uehara T, Samra JS, Gill AJ, Wolfgang CL, Goggins MG, Hruban RH, Wood LD. Genetic analyses of isolated high-grade pancreatic intraepithelial neoplasia (HG-PanIN) reveal paucity of alterations in TP53 and SMAD4. *J Pathol* 2017; **242**: 16-23 [PMID: 28188630 DOI: 10.1002/path.4884]
65 **Luchini C**, Nottegar A. The Roles of Chromatin Remodeling Genes in Pancreatic-Biliary Malignancies. *Crit Rev Oncog* 2017; **22**: 471-479 [PMID: 29604925 DOI: 10.1615/CritRevOncog.2017020587]
66 **Waddell N**, Pajic M, Patch AM, Chang DK, Kassahn KS, Bailey P, Johns AL, Miller D, Nones K, Quek K, Quinn MC, Robertson AJ, Fadlullah MZ, Bruxner TJ, Christ AN, Harliwong I, Idrisoglu S, Manning S, Nourse C, Nourbakhsh E, Wani S, Wilson PJ, Markham E, Cloonan N, Anderson MJ, Fink JL, Holmes O, Kazakoff SH, Leonard C, Newell F, Poudel B, Song S, Taylor D, Waddell N, Wood S, Xu Q, Wu J, Pinese M, Cowley MJ, Lee HC, Jones MD, Nagrial AM, Humphris J, Chantrill LA, Chin V, Steinmann AM, Mawson A, Humphrey ES, Colvin EK, Chou A, Scarlett CJ, Pinho AV, Giry-Laterriere M, Rooman I, Samra JS, Kench JG, Pettitt JA, Merrett ND, Toon C, Epari K, Nguyen NQ, Barbour A, Zeps N, Jamieson NB, Graham JS, Niclou SP, Bjerkvig R, Grützmann R, Aust D, Hruban RH, Maitra A, Iacobuzio-Donahue CA, Wolfgang CL, Morgan RA, Lawlor RT, Corbo V, Bassi C, Falconi M, Zamboni G, Tortora G, Tempero MA; Australian Pancreatic Cancer Genome Initiative, Gill AJ, Eshleman JR, Pilarsky C, Scarpa A, Musgrove EA, Pearson JV, Biankin AV, Grimmond SM. Whole genomes redefine the mutational landscape of pancreatic cancer. *Nature* 2015; **518**: 495-501 [PMID: 25719666 DOI: 10.1038/nature14169]
67 **Luchini C**, Veronese N, Solmi M, Cho H, Kim JH, Chou A, Gill AJ, Faraj SF, Chaux A, Netto GJ, Nakayama K, Kyo S, Lee SY, Kim DW, Yousef GM, Scorilas A, Nelson GS, Köbel M, Kalloger SE, Schaeffer DF, Yan HB, Liu F, Yokoyama Y, Zhang X, Pang D, Lichner Z, Sergi G, Manzato E, Capelli P, Wood LD, Scarpa A, Correll CU. Prognostic role and implications of mutation status of tumor suppressor gene ARID1A in cancer: a systematic review and meta-analysis. *Oncotarget* 2015; **6**: 39088-39097 [PMID: 26384299 DOI: 10.18632/oncotarget.5142]
68 **Luchini C**, Veronese N, Yachida S, Cheng L, Nottegar A, Stubbs B, Solmi M, Capelli P, Pea A, Barbareschi M, Fassan M, Wood LD, Scarpa A. Different prognostic roles of tumor suppressor gene BAP1 in cancer: A systematic review with meta-analysis. *Genes Chromosomes Cancer* 2016; **55**: 741-749 [PMID: 27223342 DOI: 10.1002/gcc.22381]
69 **Tanaka M**, Fernández-Del Castillo C, Kamisawa T, Jang JY, Levy P, Ohtsuka T, Salvia R, Shimizu Y, Tada M, Wolfgang CL. Revisions of international consensus Fukuoka guidelines for the management of IPMN of the pancreas. *Pancreatology* 2017; **17**: 738-753 [PMID: 28735806 DOI: 10.1016/j.pan.2017.07.007]
70 **Springer S**, Wang Y, Dal Molin M, Masica DL, Jiao Y, Kinde I, Blackford A, Raman SP, Wolfgang CL, Tomita T, Niknafs N, Douville C, Ptak J, Dobbyn L, Allen PJ, Klimstra DS, Schattner MA, Schmidt CM, Yip-Schneider M, Cummings OW, Brand RE, Zeh HJ, Singhi AD, Scarpa A, Salvia R, Malleo G, Zamboni G, Falconi M, Jang JY, Kim SW, Kwon W, Hong SM, Song KB, Kim SC, Swan N, Murphy J, Geoghegan J, Brugge W, Fernandez-Del Castillo C, Mino-Kenudson M, Schulick R, Edil BH, Adsay V, Paulino J, van Hooft J, Yachida S, Nara S, Hiraoka N, Yamao K, Hijioka S, van der Merwe S, Goggins M, Canto MI, Ahuja N, Hirose K, Makary M, Weiss MJ, Cameron J, Pittman M, Eshleman JR, Diaz LA Jr, Papadopoulos N, Kinzler KW, Karchin R, Hruban RH, Vogelstein B, Lennon AM. A combination of molecular markers and clinical features improve the classification of pancreatic cysts. *Gastroenterology* 2015; **149**: 1501-1510 [PMID: 26253305 DOI: 10.1053/j.gastro.2015.07.041]
71 **Felsenstein M**, Noë M, Masica DL, Hosoda W, Chianchiano P, Fischer CG, Lionheart G, Brosens LAA, Pea A, Yu J, Gemenetzis G, Groot VP, Makary MA, He J, Weiss MJ, Cameron JL, Wolfgang CL, Hruban RH, Roberts NJ, Karchin R, Goggins MG, Wood LD. IPMNs with co-occurring invasive cancers: neighbours but not always relatives. *Gut* 2018; **67**: 1652-1662 [PMID: 29500184 DOI: 10.1136/gutjnl-2017-315062]
72 **Scarpa A**, Real FX, Luchini C. Genetic unrelatedness of co-occurring pancreatic adenocarcinomas and IPMNs challenges current views of clinical management. *Gut* 2018; **67**: 1561-1563 [PMID: 29661802 DOI: 10.1136/gutjnl-2018-316151]
73 **Pea A**, Yu J, Rezaee N, Luchini C, He J, Dal Molin M, Griffin JF, Fedor H, Fesharakizadeh S, Salvia R, Weiss MJ, Bassi C, Cameron JL, Zheng L, Scarpa A, Hruban RH, Lennon AM, Goggins M, Wolfgang CL, Wood LD. Targeted DNA Sequencing Reveals Patterns of Local Progression in the Pancreatic Remnant Following Resection of Intraductal Papillary Mucinous Neoplasm (IPMN) of the Pancreas. *Ann Surg* 2017; **266**: 133-141 [PMID: 27433916 DOI: 10.1097/SLA.0000000000001817]
74 **Yamaguchi J**, Yokoyama Y, Kokuryo T, Ebata T, Nagino M. Cells of origin of pancreatic neoplasms. *Surg Today* 2018; **48**: 9-17 [PMID: 28260136 DOI: 10.1007/s00595-017-1501-2]
75 **Yamaguchi J**, Mino-Kenudson M, Liss AS, Chowdhury S, Wang TC, Fernández-Del Castillo C, Lillemoe KD, Warshaw AL, Thayer SP. Loss of Trefoil Factor 2 From Pancreatic Duct Glands Promotes Formation of Intraductal Papillary Mucinous Neoplasms in Mice. *Gastroenterology* 2016; **151**: 1232-1244.e10 [PMID: 27523981 DOI: 10.1053/j.gastro.2016.07.045]
76 **Sánchez-Arévalo Lobo VJ**, Fernández LC, Carrillo-de-Santa-Pau E, Richart L, Cobo I, Cendrowski J, Moreno U, Del Pozo N, Megías D, Bréant B, Wright CV, Magnuson M, Real FX. c-Myc downregulation is required for preacinar to acinar maturation and pancreatic homeostasis. *Gut* 2018; **67**: 707-718 [PMID: 28159836 DOI: 10.1136/gutjnl-2016-312306]

P- Reviewer: Guo JC, Yang F, Guo XZ, Mukaida N **S- Editor**: Dou Y
L- Editor: Filipodia **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 328-335

DOI: 10.4251/wjgo.v10.i10.328

ISSN 1948-5204 (online)

MINIREVIEWS

Facing the challenge of venous thromboembolism prevention in patients undergoing major abdominal surgical procedures for gastrointestinal cancer

Aikaterini Mastoraki, Sotiria Mastoraki, Dimitrios Schizas, Raphael Patras, Nikolaos Krinos, Ioannis S Papanikolaou, Andreas Lazaris, Theodore Liakakos, Nikolaos Arkadopoulos

Aikaterini Mastoraki, Raphael Patras, Nikolaos Krinos, Ioannis S Papanikolaou, Nikolaos Arkadopoulos, 4th Department of Surgery, National and Kapodistrian University of Athens, Attikon University Hospital, Athens 12462, Greece

Sotiria Mastoraki, Andreas Lazaris, Department of Vascular Surgery, National and Kapodistrian University of Athens, Attikon University Hospital, Athens 12462, Greece

Dimitrios Schizas, Theodore Liakakos, 1st Department of Surgery, National and Kapodistrian University of Athens, Laikon Hospital, Athens 11527, Greece

ORCID number: Aikaterini Mastoraki (0000-0002-9948-7503); Sotiria Mastoraki (0000-0002-5769-992X); Dimitrios Schizas (0000-0002-7046-0112); Raphael Patras (0000-0002-8988-3879); Nikolaos Krinos (0000-0002-8834-5120); Ioannis S Papanikolaou (0000-0002-7368-6168); Andreas Lazaris (0000-0001-6387-3907); Theodore Liakakos (0000-0003-2289-6242); Nikolaos Arkadopoulos (0000-0002-0355-0417).

Author contributions: All authors equally contributed to this paper with conception and design of the study, literature review and analysis, drafting and critical revision and editing, and final approval of the final version.

Conflict-of-interest statement: No potential conflicts of interest. No financial support.

Manuscript source: Invited manuscript

Correspondence to: Aikaterini Mastoraki, MD, PhD, Academic Research, Doctor, Lecturer, Surgeon, 4th Department of Surgery, National and Kapodistrian University of Athens, Attikon University Hospital, 1 Rimini Street, Chaidari, Athens 12462, Greece. dr_kamast@yahoo.gr
Telephone: +30-69-32577710
Fax: +30-21-32061018

Received: June 20, 2018
Peer-review started: June 21, 2018
First decision: July 19, 2018
Revised: August 22, 2018
Accepted: August 28, 2018
Article in press: August 28, 2018
Published online: October 15, 2018

Abstract

Venous thromboembolism (VTE) refers to a hypercoagulable state that remains an important and preventable factor in the surgical treatment of malignancies. VTE includes two identical entities with regards to deep vein thrombosis and pulmonary embolism. The incidence of VTE after major abdominal interventions for gastrointestinal, hepato-biliary and pancreatic neoplastic disorders is as high as 25% without prophylaxis. Prophylactic use of classic or low-molecular-weight heparin, anti-Xa factors, antithrombotic stocking, intermittent pneumatic compression devices and early mobilization have been described. Nevertheless, thromboprophylaxis is often discontinued after discharge, although a serious risk may persist long after the initial triggering event, as the coagulation system remains active for at least 14 d post-operatively. The aim of this review is to evaluate the results of the current practice of VTE prevention in cancer patients undergoing major abdominal surgical operations, with special attention to adequately elucidated guidelines

and widely accepted protocols. In addition, the recent literature is presented in order to provide an update on the current concepts concerning the surgical management of the disease.

Key words: Deep vein thrombosis; Pulmonary embolism; Gastro-intestinal cancer; Thromboprophylaxis; Venous thromboembolism

Core tip: Venous thromboembolism (VTE) refers to a hypercoagulable state that remains an important and preventable factor in the surgical treatment of malignancies. The incidence of VTE after major interventions for gastro-intestinal, hepatobiliary and pancreatic neoplastic disorders is as high as 25% without prophylaxis. Prophylactic use of classic or low-molecular-weight heparin, anti-Xa factors, antithrombotic stocking, intermittent pneumatic compression devices and early mobilization have been described. The aim of this review is to evaluate the results of the current practice of VTE prevention in cancer patients undergoing major abdominal surgical operations, with attention to adequately elucidated guidelines and widely accepted protocols.

Mastoraki A, Mastoraki S, Schizas D, Patras R, Krinos N, Papanikolaou IS, Lazaris A, Liakakos T, Arkadopoulos N. Facing the challenge of venous thromboembolism prevention in patients undergoing major abdominal surgical procedures for gastrointestinal cancer. *World J Gastrointest Oncol* 2018; 10(10): 328-335 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/328.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.328

INTRODUCTION

Venous thromboembolism (VTE) refers to a hypercoagulable state that remains an important and preventable factor in the surgical treatment of malignancies. VTE includes two identical entities with regards to deep vein thrombosis (DVT) and pulmonary embolism (PE)[1]. The incidence of VTE after major abdominal intervention for gastrointestinal (GI), hepatobiliary and pancreatic (HPB) neoplastic disorders is as high as 25% without prophylaxis[2]. Associated immobility, the Trendelenburg position, abdominal surgical procedure, potential compression of the vena cava, placement of intravenous catheters and chemotherapy have been proposed as major determinants of hypercoagulation and VTE prevalence. Neoadjuvant chemoradiotherapy followed by surgical resection as well as laparoscopic techniques have also been implicated. Recent surveys suggest that mechanical and pharmacological prophylaxis is effective in preventing post-operative VTE[3]. Prophylactic use of classic or low-molecular-weight heparin (LMWH), anti-Xa factors, antithrombotic stocking, intermittent pneumatic compression devices and early mobilization have been alternatively described[4]. Nevertheless, thromboprophylaxis is interrupted early in many cases, while relevant risk may exist long after discharge, as the activation of the coagulation system persists for at least 2 wk post-operatively. In 2007, the American Society of Clinical Oncology (ASCO) suggested an evidence-based clinical practice for the prophylactic and therapeutic approach to VTE. A subsequent update has recently been reported. However, there is still debate about the choice and duration of the appropriate anticoagulation therapeutic approach. Both guidelines recommend consideration of extended prophylaxis in high-risk patients, despite the lack of a relevant, specific definition[5]. The aim of this study was to elucidate the results of the current practice of VTE prevention in cancer patients undergoing major abdominal surgical operations, with special attention to adequately evaluated guidelines and widely accepted protocols. In addition, recent literature is presented to provide an update on current concepts in surgical management of the disease.

HISTOLOGY AND PATHOGENESIS

Although several predisposing factors in DVT have been meticulously investigated, mechanisms of thrombus development remain unclear. The classic Virchow triad refers to the combination of blood flow restriction, a hypercoagulable state and prothrombotic alterations in the vessel wall, and plays a pivotal role in thrombosis initiation[6]. Traditionally, a blood clot contains a mishmash of platelets, red blood cells and fibrin. Arterial clots are usually created under high shear stress after rupture of an atherosclerotic plaque or other vascular destruction. As they are platelet-rich, administration of antiplatelet drugs is often implemented. In contrast, venous clots are fibrin-rich and develop under lower shear stress on the surface of a macroscopically intact endothelium. The therapeutic approach always involves anticoagulant drug administration[7]. Disturbed blood flow remains a significant risk parameter, as it can provoke DVT due to long-term immobilization[8]. Hypoxia activates the endothelium, promotes the release of Weibel-Palade bodies (storage granules in endothelial cells), and facilitates blood coagulation. Weibel-Palade bodies are also responsible for the production of the von Willebrand factor, which has an important pathogenetic role in platelet recruitment.

The blood coagulation cascade is well-defined and divided into the extrinsic and intrinsic pathways. Deficiencies in the anticoagulants antithrombin and proteins C and S constitute significant genetic risk factors that contribute to the development of a hypercoagulable condition. Mild genetic alterations in von Leiden factor, prothrombin G20210A and fibrinogen C10034T predispose patients to decreased fibrinolysis[9]. It is common knowledge that the most frequent site of thrombus formation is the valve pocket sinus due to its vertical blood flow and inadequate oxygen tension. Therefore, small thrombi initiated within the valve pocket develop slowly and extend along the inside of the vein wall, resu-

lting in vascular occlusion. It has been proposed that, under abnormal conditions, tissue factor (TF) is expressed on both circulating leukocytes and activated endothelial cells together with the platelet inhibitors nitric oxide and prostacyclin[10]. In addition, recent surveys demonstrate that neutrophils accelerate thrombosis by releasing serine proteases that inactivate the anticoagulant TF pathway inhibitor, suggesting that interfering with the binding of leukocytes to the activated endothelium may represent a promising therapeutic strategy against DVT. Finally, post-thrombotic syndrome describes chronic venous insufficiency following DVT and is attributed to venous hypertension, which may result from persistent thrombotic occlusion or venous valvular reflux due to a previous thrombotic condition[11]. Additionally, inflammation may contribute to successive venous valvular damage.

CLINICAL PRESENTATION

Considering that VTE encompasses two clinical conditions, including DVT and PE, clinical findings refer to both nosologic entities. DVT typically presents with pain and lower limb oedema, the latter being the most specific symptom. If the thrombus is located in the ileac bifurcation, pelvic veins or the inferior vena cava, bilateral rather than unilateral oedema is usually apparent. Moreover, high partial obstruction often causes moderate oedema imitating that of heart, liver or renal insufficiency[12]. Pain with tenderness occurs in the majority of affected patients. Relevant clinical signs are considered nonspecific and remain independent of the size, location and extent of the thrombus. Warmth of the related limb, locoregional erythema, or discoloration and dilation of superficial veins may also be apparent. Homan's sign (calf pain on dorsiflexion of the foot) also presents in 50% of patients with DVT[13]. Furthermore, DVT should be differentially diagnosed from various other diseases including cellulitis, Baker's cyst, musculoskeletal injury, neoplasm, lymphedema, hematoma, sarcoma, venous or arterial aneurysms, and connective tissue disorders[14]. Finally, a very uncommon but hazardous form of DVT is Phlegmasia Cerulea Dolens, which is the consequence of extensive thrombotic occlusion of the major and collateral veins of a lower extremity, including the ileac and femoral veins. It is characterized by acute onset of pain, oedema, blue discoloration and swelling of the affected limb, which, if left untreated, will result in foot gangrene[15].

As far as PE is concerned, aetiology refers to air, septic and amniotic fluid emboli. Relevant clinical findings may vary from deadly hemodynamic collapse to progressive dyspnoea, and most patients with PE present with obscure symptoms. Taking into consideration the aforementioned clinical evidence, common signs of PE include sudden dyspnoea (73%) that worsens with exertion, pleuritic chest pain (66%) deteriorating with inhalation, or exertion and a productive cough (37%) that may lead to haemoptysis (13%)[16]. Similar findings upon physical examination include tachypnoea, rales, tachycardia, fever, cardiac galloping, lower limb oedema and cyanosis.

DIAGNOSTIC MODALITIES

Several imaging studies have been proposed for DVT diagnosis. Duplex Ultrasonography (B-mode and Doppler) remains the current first line examination performed, due to non-invasiveness and absence of irradiation or contrast material[17]. B-mode is based on the principle that normal venous structures easily collapse with the pressure applied by the transducer, while veins harboring thrombi will not compress and will therefore be visible. The Doppler color-flow imaging technique can reveal the potential adequacy of blood flow in an area where an isoechoic clot might not be depicted. Sensitivity and specificity are as high as 95% in symptomatic patients, but diminish with obesity, small and peripheral thrombi, as well as asymptomatic disease[18].

Venography with pedal vein cannulation, injection of contrast material, and serial limb radiographs remains the diagnostic modality of choice for DVT verification, with sensitivity and specificity reaching 100%. However, this technique is invasive and may induce serious consequences, such as hypersensitivity reactions, superficial phlebitis and renal toxicity. Another modality applied is Impedance Plethysmography, which is sensitive and specific in proximal vein thrombosis. It measures the electrical resistance of the calf, which reflects changes in blood volume[19]. Spiral multidetector-row CT venography from the popliteal fossa provides adequate diagnostic accuracy in association with sonographic assessment. Finally, magnetic resonance imaging remains the modality of choice for suspected ileac vein or inferior vena cava thrombosis, especially when CT venography is contraindicated or technically difficult[20]. Radiolabelled peptides that tend to connect to various thrombus components have also been studied. Apcitide, a technetium-labelled platelet glycoprotein IIb/IIIa receptor antagonist, is proposed for diagnostic investigations of DVT. Diagnostic modalities related to DVT detection are summarized in Table 1.

With regard to PE, common electrocardiographic abnormalities, including tachycardia, nonspecific ST-T disorders, right heart strain, atrial fibrillation and S_1 Q_3 T_3 pattern, are encountered in the minority of affected patients[21]. CT pulmonary angiography (CTPA) is considered as the initial imaging modality of choice for stable patients[22]. CTPA reveals emboli as an intraluminal filling defect after injection of contrast material, is non-invasive and widely available, and provides invaluable information for differential diagnosis[23]. Sensitivity and specificity are disproportionate to the size of the affected pulmonary artery. Nevertheless, PA is the criterion standard for diagnosing PE. With the use of contrast material, a filling defect or a sharp cut-off of the problematic artery is detected in anterior, posterior and lateral studies[24]. Essential to verifying PE, Ventilation/

Table 1 Diagnostic modalities applied for deep vein thrombosis detection

Deep vein thrombosis	U/S (B-mode)	U/S (Doppler)	Venography	Impedance plethysmography	CTV	MRI	Radiolabeled peptides
Mechanism of action	Veins with thrombi do not compress	Absent or abnormal blood flow when a thrombus is present	Pedal vein cannulation and injection of contrast material	Measures electrical resistance of the calf reflecting blood volume change	Spiral multidetector CT venography from popliteal fossa to the pelvis	-	Radiolabeled peptides that bind to various components of a thrombus
Sensitivity and specificity	95%	95%	100%	Sensitive and specific in proximal vein thrombosis	-	-	-
Advantages	Non-invasiveness Absence of radiation or contrast material	Non-invasiveness Absence of radiation or contrast material	High sensitivity and specificity	-	-	Ileac vein or inferior vena cava thrombosis, when CT venography is contraindicated or technically inadequate	Apcitide, a technetium-labeled platelet glycoprotein IIb/IIIa receptor antagonist
Disadvantages	Obesity, small peripheral thrombi, asymptomatic disease	Obesity, small peripheral thrombi asymptomatic disease	Invasiveness Hypersensitivity reactions Renal toxicity	-	Correlation with sonographic findings	-	Expensive

CTV: Computed tomography venography; MRI: Magnetic resonance imaging.

Perfusion Scanning may be used when CTPA or Pulmonary Angiography are contraindicated[25]. Finally, PE demonstrates increased signal intensity within the pulmonary artery during magnetic resonance angiography with intravenous administration of gadolinium. Sensitivity and specificity are high for central, lobar, and segmental emboli, while sub-segmental emboli render magnetic resonance angiography inadequate[26] (Table 2).

THERAPEUTIC APPROACH

VTE remains the second most common cause of death in cancer patients and constitutes an independent prognostic factor for mortality[27]. Moreover, recurrent VTE and major bleeding complications are higher in cancer patients, even if they receive anticoagulation therapy. Patients with upper GI malignancies, such as hepatobiliary and gastroesophageal cancer, are in great danger of VTE, with pancreatic cancer presenting the highest VTE prevalence. In advanced pancreatic cancer patients, relevant risk is as high as 25%, and asymptomatic VTE incidence is up to 60%. GI cancers are frequently treated with antiangiogenic or chemotherapeutic agents, such as cisplatin and irinotecan, which are associated with increased risk for VTE as well as combined neoadjuvant chemoradiotherapy[28]. It is estimated that chemotherapy provokes an inflammatory response due to endothelial disruption. In particular, IL-1 and TNF-a, among other cytokines, diminish the concentration of anticoagulant proteins, such as antithrombin Ⅲ and protein C. The procoagulant reaction is reinforced by increased TF expression, and is maintained for up to 6 mo after induction of chemoradiation, thus implying an increased long-term risk for VTE[29]. Major abdominal cancer surgery is also a risk factor for VTE, even after hospital discharge and discontinuation of the usual perioperative prophylaxis[30]. As for laparoscopic surgery, there is still no consensus as to whether the laparoscopic or open techniques abate morbidity related to VTE. Some investigators state that the risk is lessened due to overall reduction in postoperative morbidity, while others claim that the impact of pneumoperitoneum increases the risk, due to compression of the inferior vena cava and iliac veins. However, existing trials are not adequate to reliably evaluate these findings[31].

Pharmacologic thromboprophylaxis is strongly recommended in patients with GI cancer undergoing major surgery, as risk reduction up to 80% has been proven for VTE. Current guidelines suggest LMWH as the standard of care. ASCO recommendations propose unfractionated heparin, fondaparinux or LMWH as a first-line treatment, unless contraindicated due to high bleeding risk or active bleeding. Prophylactic dosages at levels of 3000-5000 anti-Fxa units per day have proven more effective than and as safe as lower doses. Treatment should begin 12-24 h pre- or 6-24 h postoperatively and last 7-10 d. The combination of pharmacologic and mechanical prophylaxis, such as compression stockings and intermittent pneumatic compression devices, may be more efficient, especially in the high-risk group of patients. Extended thromboprophylaxis up to 28 d should be taken into serious consideration only in high-risk patients who fulfill the following criteria, including cancer-related stage Ⅲ/Ⅳ, upper GI cancer, histological features of adenocarcinoma, thrombocytosis, leucocytosis, elevated D-dimer and CRP. Patient-related factors refer

Table 2 Imaging modalities for pulmonary embolism verification

PE	ECG	CTPA	V/Q Scan	MRA
Findings	Sinus tachycardia Non-specific ST-T disorders S1Q3T3 pattern Atrial fibrillation Right heart strain	Intraluminal filling defect of pulmonary artery after injection of contrast material	Ventilated area not perfused	Increased signal intensity of pulmonary thrombi within pulmonary artery after injection of gadolinium
Advantages	Immediate Costless	Criterion standard for diagnosis	Radiation dose lower than CTPA	High sensitivity and specificity for central, lobar, and segmental emboli
Disadvantages	Low sensitivity and specificity	Invasiveness Hypersensitivity reactions Renal toxicity	-	Inadequate for subsegmental emboli

CTPA: CT pulmonary angiography; V/Q: Ventilation/perfusion; MRA: Magnetic resonance angiography.

to ages older than 60 years, obesity, previous history of VTE, surgery lasting 2 h or longer, prolonged postoperative immobilization, and presence of infection or fever. Treatment-related determinants include chemotherapy, central-line or port catheter, parenteral nutrition and radiation therapy. On the other hand, European Society of Molecular Oncology (ESMO) guidelines do not suggest fondaparinux as the first line of treatment and recommend extended prophylaxis up to 28 d for all cancer patients undergoing abdominal or pelvic surgery. Given these differentiations, the newest ESMO and ASCO guidelines consort with each other. American Society of Haematology and Australian Government National Health and Medical Research Council recommendations coincide with ASCO guidelines, while Mayo Clinic VTE Prevention and Management and German guidelines go along with ESMO proposals.

As far as long-term prevention of VTE is concerned, ASCO guidelines suggest the use of LMWH as the standard of care. If this is unavailable, vitamin K antagonists (VKA) are used. Novel Oral Anticoagulants (NOACs) are currently not suggested for patients with GI cancer and VTE due to the limited data available in patients with malignancy. Treatment should last for 6 mo. ESCO and American Society of Haematology guidelines recommend the use of LMWH for 6 mo. ESCO specifically proposes an initial dose of LMWH 100% for 1 mo and 75%-80% of the initial dose for 5 mo thereafter. Additionally, the Mayo Clinic suggests that anticoagulants should be continued until there is no evidence of active malignancy, either as evidence of imaging or cancer-related treatment, while German guidelines propose LMWH for 3-6 mo and highlight that prophylaxis could last for a lifetime in persistent cancers[32].

Non-vitamin-K NOACs have been introduced in the treatment of VTE associated with GI cancer. As the aforementioned guidelines state, the use of NOACs are currently not recommended due to limited data in cancer patients. On the other hand, available anticoagulants exhibit certain disadvantages. Unfractionated heparin requires platelet monitoring and daily injections, which are highly inconvenient. Also, it is associated with heparin-induced thrombocytopenia, types Ⅰ and Ⅱ, bleeding and osteoporosis. LMWH is contraindicated in renal impairment, adding to its drawbacks. VKA, such as warfarin, require INR monitoring and have multiple drug and food interactions. A narrow therapeutic window, delayed onset of action and bleeding risk render VKA inadequate and inferior to LMWH[28].

The family of NOACs includes dabigatran etexilate, rivaroxaban, apixaban and edoxaban, each one with their own special pharmacokinetics and pharmacodynamics[33]. Dabigatran etexilate is a direct thrombin (factor IIa) inhibitor. It is administered orally and presents a half-life of 12-14 h and a rapid onset of action. Its bioavailability does not exceed 10% (3-7%), and its absorption is facilitated by acids. Its excretion is primarily in urine (80%), so caution is required for patients with renal impairment, as its half-life can be increased up to 34 h. More specifically, it is contraindicated when creatinine clearance (CrCl) is under 30 mL/min. Its clearance is also dependent on the P-glycoprotein transport pathway. In addition, routine monitoring is not required because of predictable pharmacokinetics. In the RE-COVER, RE-SONATE and RE-MEDY phase Ⅲ clinical trials, dabigatran showed non-inferiority to warfarin and superiority to placebo. It is FDA approved for VTE prophylaxis after hip and knee arthroplasty, stroke prevention in patients with non-valvular atrial fibrillation, and VTE treatment. As for adverse effects, dyspepsia is the only one that occurs more frequently with dabigatran than with warfarin[28,33].

Rivaroxaban is a direct inhibitor of factor Xa. It is orally administered and has a half-life of 7-11 h, with a rapid onset of action as well[34]. Its bioavailability is excellent (80%-100%). Significant food interactions have not been reported. It is a substrate of the cytochrome P450 system, especially CYP3A4 and P-glycoprotein, and is excreted by both the renal and hepatic systems, demanding extreme caution in patients with renal or hepatic insufficiency[33]. More specifically, it is contraindicated when CrCl is under 30 mL/min in haemodialysis and in patients with Child-Pugh B or C cirrhosis. Additionally, monitoring is not required. In the EINSTEIN-DVT and EINSTEIN-Extension phase Ⅲ clinical trials, rivaroxaban presented non-inferiority to VKA/LMWH

Table 3 Comparative evaluation of mechanism of action and contra-indications of novel oral anticoagulants

Novel oral anticoagulants	Dabigatran	Rivaroxaban	Apixaban	Edoxaban
Mechanism of action	Direct thrombin (factor IIa) inhibitor	Direct factor Xa inhibitor	Direct factor Xa inhibitor	Direct factor Xa inhibitor
Route of administration	Per os	Per os	Per os	Per os
Half-life	12-14 h	7-11 h	12 h	8-10 h
Bioavailability	3%-7%	80%-100%	50%	62%
Metabolism	P-glycoprotein	P-glycoprotein Cytochrome P450 system (CYP3A4)	P-glycoprotein Cytochrome P450 system (CYP3A4)	P-glycoprotein Cytochrome P450 system (CYP3A4)
Excretion	Urine (80%)	Urine and HBR	Urine (25%)	Primarily: HBR Secondarily: Urine
Contraindication	CrCl < 30 mL/min	CrCl < 30 mL/min Hemodialysis Child Pugh B and C stage cirrhosis	CrCl < 15 mL/min	
FDA approval	VTE prophylaxis after hip and knee arthroplasty, Non valvular atrial fibrillation VTE treatment	VTE prophylaxis after hip and knee arthroplasty, Non valvular atrial fibrillation VTE treatment	Non valvular atrial fibrillation VTE treatment and prevention after major orthopedic surgery	Non valvular atrial fibrillation VTE treatment and prevention after major orthopedic surgery
Clinical trials	Non-inferiority to warfarin Superiority to placebo	Non-inferiority to VKA/ LMWH Superiority to placebo	-	
Dosage	100-150 mg × 2/24 h	10-30 mg × 1/24 h	2.5-5 mg × 2/24 h	15-30 mg × 1/24 h

HBR: Hepatobiliary route; VTE: Venous thromboembolism; VKA: Vitamin K antagonists; LMWH: Low molecular weight heparin.

and superiority to placebo, respectively. It is also FDA approved with the same indications as dabigatran[28].

Apixaban appears to have the same mode of action and route of administration as rivaroxaban. It has a half-life of 12 h, and a bioavailability of about 50%. It is metabolized by P-glycoprotein, the cytochrome 450 system, and the CYP3A4 pathway. Its excretion is in urine (25%), and the drug is contraindicated when CrCl is under 15 mL/min. Edoxaban inhibits factor Xa and is orally administered. It has a half-life of 8-10 h and good bioavailability (62%). It is excreted primarily by the hepatobiliary route and secondarily in the urine. Also, it is metabolized by both P-glycoprotein and the CYP3A4 pathway. As mentioned for the other NOACs, monitoring is not required. Both apixaban and edoxaban are FDA approved for non-valvular atrial fibrillation, VTE treatment and prevention after major orthopaedic surgery[28,33].

As far as dosing frequency is concerned, dabigatran and apixaban require 110-150 mg and 2.5-5 mg twice a day, respectively, while rivaroxaban and edoxaban necessitate 10-30 mg and 15-30 mg once daily, respectively[33]. Apixaban remains the safest of the NOACs, showing reduced risk of major or clinically-relevant minor bleeding at a statistically significant level, with dabigatran taking the second place. Additionally, apixaban and rivaroxaban pose a significantly lower risk for major bleeding compared with LMWH or VKA, a fact which may be of particular clinical importance[35]. In conclusion, differences between doses of dabigatran and apixaban, as well as the correlations between the safety and timing differences between dabigatran or apixaban and rivaroxaban or edoxaban, are summarized in Table 3.

CONCLUSION

VTE refers to both DVT and PE and is highly associated with malignancy, with HPB and gastric cancer ranking first[36]. CTPA is the initial diagnostic modality, while ultrasonography is preferred for DVT[37]. LMWH is used pre- or 6-24 h postoperatively and should continue for 7-10 d. Extension up to 28 d is highly recommended for major abdominal or pelvic surgical procedures[38-42]. NOACs are promised to revolutionize current treatment and bring together efficacy and many benefits for patients. However, the use of NOACs for VTE prophylaxis is certainly debatable. Potential drug interactions with chemotherapeutic components, GI abnormalities, and hepatic and renal insufficiency remain significant determinants of NOAC administration[43-45]. Therefore, bioavailability may not reach desirable levels[46]. The lack of rapid reversal agents also prevents the use of these agents for invasive procedures and thrombocytopenia. Furthermore, cancer patients are at a greater risk of bleeding than non-cancer patients due to chemotherapy-induced thrombocytopenia and antiangiogenic therapy. Moreover, a reduction in circulating proteins and albumins could influence the binding levels of NOACs. Thus, a comparative study of NOACs with the current curative approach, LMWH, may clarify this dispute.

REFERENCES

1 **Larsen AC**, Dabrowski T, Frøkjær JB, Fisker RV, Iyer VV, Møller

BK, Kristensen SR, Thorlacius-Ussing O. Prevalence of venous thromboembolism at diagnosis of upper gastrointestinal cancer. *Br J Surg* 2014; **101**: 246-253 [PMID: 24446107 DOI: 10.1002/bjs.9353]
2 **Larsen AC**, Frøkjær JB, Fisker RV, Iyer V, Mortensen PB, Yilmaz MK, Møller B, Kristensen SR, Thorlacius-Ussing O. Treatment-related frequency of venous thrombosis in lower esophageal, gastro-esophageal and gastric cancer--a clinical prospective study of outcome and prognostic factors. *Thromb Res* 2015; **135**: 802-808 [PMID: 25743885 DOI: 10.1016/j.thromres.2015.01.021]
3 **Davenport DL**, Vargas HD, Kasten MW, Xenos ES. Timing and perioperative risk factors for in-hospital and post-discharge venous thromboembolism after colorectal cancer resection. *Clin Appl Thromb Hemost* 2012; **18**: 569-575 [PMID: 22345485 DOI: 10.1177/1076029611433642]
4 **Holwell A**, McKenzie JL, Holmes M, Woods R, Nandurkar H, Tam CS, Bazargan A. Venous thromboembolism prevention in patients undergoing colorectal surgery for cancer. *ANZ J Surg* 2014; **84**: 284-288 [PMID: 23782713 DOI: 10.1111/ans.12296]
5 **Lyman GH**, Khorana AA, Kuderer NM, Lee AY, Arcelus JI, Balaban EP, Clarke JM, Flowers CR, Francis CW, Gates LE, Kakkar AK, Key NS, Levine MN, Liebman HA, Tempero MA, Wong SL, Prestrud AA, Falanga A; American Society of Clinical Oncology Clinical Practice. Venous thromboembolism prophylaxis and treatment in patients with cancer: American Society of Clinical Oncology clinical practice guideline update. *J Clin Oncol* 2013; **31**: 2189-2204 [PMID: 23669224 DOI: 10.1200/JCO.2013.49.1118]
6 **Heit JA**, Silverstein MD, Mohr DN, Petterson TM, O'Fallon WM, Melton LJ 3rd. Risk factors for deep vein thrombosis and pulmonary embolism: a population-based case-control study. *Arch Intern Med* 2000; **160**: 809-815 [PMID: 10737280 DOI: 10.1001/archinte.160.6.809]
7 **Scurr JH**, Coleridge-Smith PD, Hasty JH. Deep venous thrombosis: a continuing problem. *BMJ* 1988; **297**: 28 [PMID: 3408903 DOI: 10.1136/bmj.297.6640.28]
8 **Lee KW**, Bang SM, Kim S, Lee HJ, Shin DY, Koh Y, Lee YG, Cha Y, Kim YJ, Kim JH, Park DJ, Kim HH, Oh D, Lee JS. The incidence, risk factors and prognostic implications of venous thromboembolism in patients with gastric cancer. *J Thromb Haemost* 2010; **8**: 540-547 [PMID: 20040044 DOI: 10.1111/j.1538-7836.2009.03731.x]
9 **Mandalà M**, Barni S, Prins M, Labianca R, Tondini C, Russo L, Milesi A, Cremonesi M, Zaccanelli M, Regonesi C, Moro C, Falanga A. Acquired and inherited risk factors for developing venous thromboembolism in cancer patients receiving adjuvant chemotherapy: a prospective trial. *Ann Oncol* 2010; **21**: 871-876 [PMID: 19713246 DOI: 10.1093/annonc/mdp354]
10 **Khorana AA**, Francis CW, Menzies KE, Wang JG, Hyrien O, Hathcock J, Mackman N, Taubman MB. Plasma tissue factor may be predictive of venous thromboembolism in pancreatic cancer. *J Thromb Haemost* 2008; **6**: 1983-1985 [PMID: 18795992 DOI: 10.1111/j.1538-7836.2008.03156.x]
11 **Shah MA**, Capanu M, Soff G, Asmis T, Kelsen DP. Risk factors for developing a new venous thromboembolism in ambulatory patients with non-hematologic malignancies and impact on survival for gastroesophageal malignancies. *J Thromb Haemost* 2010; **8**: 1702-1709 [PMID: 20553384 DOI: 10.1111/j.1538-7836.2010.03948.x]
12 **Imberti D**, Agnelli G, Ageno W, Moia M, Palareti G, Pistelli R, Rossi R, Verso M; MASTER Investigators. Clinical characteristics and management of cancer-associated acute venous thromboembolism: findings from the MASTER Registry. *Haematologica* 2008; **93**: 273-278 [PMID: 18223291 DOI: 10.3324/haematol.11458]
13 **Prandoni P**, Lensing AW, Piccioli A, Bernardi E, Simioni P, Girolami B, Marchiori A, Sabbion P, Prins MH, Noventa F, Girolami A. Recurrent venous thromboembolism and bleeding complications during anticoagulant treatment in patients with cancer and venous thrombosis. *Blood* 2002; **100**: 3484-3488 [PMID: 12393647 DOI: 10.1182/blood-2002-01-0108]
14 **Bates SM**, Jaeschke R, Stevens SM, Goodacre S, Wells PS, Stevenson MD, Kearon C, Schunemann HJ, Crowther M, Pauker SG, Makdissi R, Guyatt GH. Diagnosis of DVT: Antithrombotic Therapy and Prevention of Thrombosis, 9th ed: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines. *Chest* 2012; **141**: e351S-e418S [PMID: 22315267 DOI: 10.1378/chest.11-2299]
15 **Palareti G**, Schellong S. Isolated distal deep vein thrombosis: what we know and what we are doing. *J Thromb Haemost* 2012; **10**: 11-19 [PMID: 22082302 DOI: 10.1111/j.1538-7836.2011.04564.x]
16 **Remy-Jardin M**, Pistolesi M, Goodman LR, Gefter WB, Gottschalk A, Mayo JR, Sostman HD. Management of suspected acute pulmonary embolism in the era of CT angiography: a statement from the Fleischner Society. *Radiology* 2007; **245**: 315-329 [PMID: 17848685 DOI: 10.1148/radiol.2452070397]
17 **Beyer-Westendorf J**, Halbritter K, Platzbecker H, Damme U, Neugebauer B, Kuhlisch E, Schellong S. Central adjudication of venous ultrasound in VTE screening trials: reasons for failure. *J Thromb Haemost* 2011; **9**: 457-463 [PMID: 21143379 DOI: 10.1111/j.1538-7836.2010.04166.x]
18 **Righini M**, Le Gal G, Aujesky D, Roy PM, Sanchez O, Verschuren F, Rutschmann O, Nonent M, Cornuz J, Thys F, Le Manach CP, Revel MP, Poletti PA, Meyer G, Mottier D, Perneger T, Bounameaux H, Perrier A. Diagnosis of pulmonary embolism by multidetector CT alone or combined with venous ultrasonography of the leg: a randomised non-inferiority trial. *Lancet* 2008; **371**: 1343-1352 [PMID: 18424324 DOI: 10.1016/S0140-6736(08)60594-2]
19 **Beck-Razi N**, Kuzmin A, Koren D, Sarig G, Brenner B, Haim N, Gaitini D. Asymptomatic deep vein thrombosis in advanced cancer patients: the value of venous sonography. *J Clin Ultrasound* 2010; **38**: 232-237 [PMID: 20461778 DOI: 10.1002/jcu.20691]
20 **Shammas NW**, Rachwan RJ, Daher G, Bou Dargham B. Double Inferior Vena Cava and its Implications During Endovascular and Surgical Interventions: A Word of Caution. *J Invasive Cardiol* 2017; **29**: 51-53 [PMID: 28145872]
21 **Torbicki A**, Perrier A, Konstantinides S, Agnelli G, Galiè N, Pruszczyk P, Bengel F, Brady AJ, Ferreira D, Janssens U, Klepetko W, Mayer E, Remy-Jardin M, Bassand JP; ESC Committee for Practice Guidelines (CPG). Guidelines on the diagnosis and management of acute pulmonary embolism: the Task Force for the Diagnosis and Management of Acute Pulmonary Embolism of the European Society of Cardiology (ESC). *Eur Heart J* 2008; **29**: 2276-2315 [PMID: 18757870 DOI: 10.1093/eurheartj/ehn310]
22 **Qanadli SD**, Hajjam ME, Mesurolle B, Barré O, Bruckert F, Joseph T, Mignon F, Vieillard-Baron A, Dubourg O, Lacombe P. Pulmonary embolism detection: prospective evaluation of dual-section helical CT versus selective pulmonary arteriography in 157 patients. *Radiology* 2000; **217**: 447-455 [PMID: 11058644 DOI: 10.1148/radiology.217.2.r00nv01447]
23 **Winer-Muram HT**, Rydberg J, Johnson MS, Tarver RD, Williams MD, Shah H, Namyslowski J, Conces D, Jennings SG, Ying J, Trerotola SO, Kopecky KK. Suspected acute pulmonary embolism: evaluation with multi-detector row CT versus digital subtraction pulmonary arteriography. *Radiology* 2004; **233**: 806-815 [PMID: 15564410 DOI: 10.1148/radiol.2333031744]
24 **Stein PD**, Fowler SE, Goodman LR, Gottschalk A, Hales CA, Hull RD, Leeper KV Jr, Popovich J Jr, Quinn DA, Sos TA, Sostman HD, Tapson VF, Wakefield TW, Weg JG, Woodard PK; PIOPED II Investigators. Multidetector computed tomography for acute pulmonary embolism. *N Engl J Med* 2006; **354**: 2317-2327 [PMID: 16738268 DOI: 10.1056/NEJMoa052367]
25 **Anderson DR**, Kahn SR, Rodger MA, Kovacs MJ, Morris T, Hirsch A, Lang E, Stiell I, Kovacs G, Dreyer J, Dennie C, Cartier Y, Barnes D, Burton E, Pleasance S, Skedgel C, O'Rouke K, Wells PS. Computed tomographic pulmonary angiography vs ventilation-perfusion lung scanning in patients with suspected pulmonary embolism: a randomized controlled trial. *JAMA* 2007; **298**: 2743-2753 [PMID: 18165667 DOI: 10.1001/jama.298.23.2743]
26 **Perrier A**, Roy PM, Sanchez O, Le Gal G, Meyer G, Gourdier AL, Furber A, Revel MP, Howarth N, Davido A, Bounameaux H. Multidetector-row computed tomography in suspected pulmonary embolism. *N Engl J Med* 2005; **352**: 1760-1768 [PMID: 15858185 DOI: 10.1056/NEJMoa042905]
27 **Khorana AA**, Dalal M, Lin J, Connolly GC. Incidence and pre-

dictors of venous thromboembolism (VTE) among ambulatory high-risk cancer patients undergoing chemotherapy in the United States. *Cancer* 2013; **119**: 648-655 [PMID: 22893596 DOI: 10.1002/cncr.27772]

28 **Martin LK**, Bekaii-Saab T. Management of venous thromboembolism in patients with advanced gastrointestinal cancers: what is the role of novel oral anticoagulants? *Thrombosis* 2012; **2012**: 758385 [PMID: 23024860 DOI: 10.1155/2012/758385]

29 **Larsen AC**, Brøndum Frøkjaer J, Wishwanath Iyer V, Vincents Fisker R, Sall M, Yilmaz MK, Kuno Møller B, Kristensen SR, Thorlacius-Ussing O. Venous thrombosis in pancreaticobiliary tract cancer: outcome and prognostic factors. *J Thromb Haemost* 2015; **13**: 555-562 [PMID: 25594256 DOI: 10.1111/jth.12843]

30 **White RH**. The epidemiology of venous thromboembolism. *Circulation* 2003; **107**: I4-I8 [PMID: 12814979 DOI: 10.1161/01.CIR.0000078468.11849.66]

31 **Kimura Y**, Oki E, Ando K, Saeki H, Kusumoto T, Maehara Y. Incidence of Venous Thromboembolism Following Laparoscopic Surgery for Gastrointestinal Cancer: A Single-Center, Prospective Cohort Study. *World J Surg* 2016; **40**: 309-314 [PMID: 26316113 DOI: 10.1007/s00268-015-3234-y]

32 **Riess H**, Habbel P, Jühling A, Sinn M, Pelzer U. Primary prevention and treatment of venous thromboembolic events in patients with gastrointestinal cancers - Review. *World J Gastrointest Oncol* 2016; **8**: 258-270 [PMID: 26989461 DOI: 10.4251/wjgo.v8.i3.258]

33 **Franchini M**, Bonfanti C, Lippi G. Cancer-associated thrombosis: investigating the role of new oral anticoagulants. *Thromb Res* 2015; **135**: 777-781 [PMID: 25743884 DOI: 10.1016/j.thromres.2015.02.024]

34 **Bott-Kitslaar DM**, Saadiq RA, McBane RD, Loprinzi CL, Ashrani AA, Ransone TR, Wolfgram AA, Berentsen MM, Wysokinski WE. Efficacy and Safety of Rivaroxaban in Patients with Venous Thromboembolism and Active Malignancy: A Single-Center Registry. *Am J Med* 2016; **129**: 615-619 [PMID: 26797081 DOI: 10.1016/j.amjmed.2015.12.025]

35 **Cohen AT**, Hamilton M, Mitchell SA, Phatak H, Liu X, Bird A, Tushabe D, Batson S. Comparison of the Novel Oral Anticoagulants Apixaban, Dabigatran, Edoxaban, and Rivaroxaban in the Initial and Long-Term Treatment and Prevention of Venous Thromboembolism: Systematic Review and Network Meta-Analysis. *PLoS One* 2015; **10**: e0144856 [PMID: 26716830 DOI: 10.1371/journal.pone.0144856]

36 **Bosch DJ**, Van Dalfsen QA, Mul VE, Hospers GA, Plukker JT. Increased risk of thromboembolism in esophageal cancer patients treated with neoadjuvant chemoradiotherapy. *Am J Surg* 2014; **208**: 215-221 [PMID: 24534559 DOI: 10.1016/j.amjsurg.2013.10.031]

37 **Shukla PJ**, Siddachari R, Ahire S, Arya S, Ramani S, Barreto SG, Gupta S, Shrikhande SV, Jagannath P, Desouza LJ. Postoperative deep vein thrombosis in patients with colorectal cancer. *Indian J Gastroenterol* 2008; **27**: 71-73 [PMID: 18695308]

38 **Lyman GH**, Bohlke K, Khorana AA, Kuderer NM, Lee AY, Arcelus JI, Balaban EP, Clarke JM, Flowers CR, Francis CW, Gates LE, Kakkar AK, Key NS, Levine MN, Liebman HA, Tempero MA, Wong SL, Somerfield MR, Falanga A; American Society of Clinical Oncology. Venous thromboembolism prophylaxis and treatment in patients with cancer: american society of clinical oncology clinical practice guideline update 2014. *J Clin Oncol* 2015; **33**: 654-656 [PMID: 25605844 DOI: 10.1200/JCO.2014.59.7351]

39 **Lee AY**. Prevention and treatment of venous thromboembolism in patients with cancer. *Hematology Am Soc Hematol Educ Program* 2014; **2014**: 312-317 [PMID: 25696871 DOI: 10.1182/asheducation-2014.1.312]

40 **Panizo E**, Alfonso A, García-Mouriz A, López-Picazo JM, Gil-Bazo I, Hermida J, Páramo JA, Lecumberri R. Factors influencing the use of thromboprophylaxis in cancer outpatients in clinical practice: A prospective study. *Thromb Res* 2015; **136**: 1145-1148 [PMID: 26475407 DOI: 10.1016/j.thromres.2015.10.015]

41 **Osaki T**, Saito H, Fukumoto Y, Kono Y, Murakami Y, Shishido Y, Kuroda H, Matsunaga T, Sato K, Hirooka Y, Fujiwara Y. Risk and incidence of perioperative deep vein thrombosis in patients undergoing gastric cancer surgery. *Surg Today* 2018; **48**: 525-533 [PMID: 29234961 DOI: 10.1007/s00595-017-1617-4]

42 **Marshall-Webb M**, Bright T, Price T, Thompson SK, Watson DI. Venous thromboembolism in patients with esophageal or gastric cancer undergoing neoadjuvant chemotherapy. *Dis Esophagus* 2017; **30**: 1-7 [PMID: 27878904 DOI: 10.1111/dote.12516]

43 **Alsubaie H**, Leggett C, Lambert P, Park J, Hochman D, Wirtzfeld D, McKay A. Diagnosis of VTE postdischarge for major abdominal and pelvic oncologic surgery: implications for a change in practice. *Can J Surg* 2015; **58**: 305-311 [PMID: 26204144 DOI: 10.1503/cjs.012314]

44 **Cui G**, Wang X, Yao W, Li H. Incidence of postoperative venous thromboembolism after laparoscopic versus open colorectal cancer surgery: a meta-analysis. *Surg Laparosc Endosc Percutan Tech* 2013; **23**: 128-134 [PMID: 23579505 DOI: 10.1097/SLE.0b013e3182827cef]

45 **Cheung HY**, Chung CC, Yau KK, Siu WT, Wong SK, Chiu E, Li MK. Risk of deep vein thrombosis following laparoscopic rectosigmoid cancer resection in chinese patients. *Asian J Surg* 2008; **31**: 63-68 [PMID: 18490217 DOI: 10.1016/S1015-9584(08)60060-3]

46 **Diao D**, Wang Z, Cheng Y, Zhang H, Guo Q, Song Y, Zhu K, Li K, Liu D, Dang C. D-dimer: not just an indicator of venous thrombosis but a predictor of asymptomatic hematogenous metastasis in gastric cancer patients. *PLoS One* 2014; **9**: e101125 [PMID: 24983619 DOI: 10.1371/journal.pone.0101125]

P- Reviewer: Jeong KY, Nakayama Y, Tanabe S **S- Editor**: Ji FF
L- Editor: Filipodia **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 336-343

DOI: 10.4251/wjgo.v10.i10.336

ISSN 1948-5204 (online)

MINIREVIEWS

Role of pre-transplant 18F-FDG PET/CT in predicting hepatocellular carcinoma recurrence after liver transplantation

Onur Yaprak, Sencan Acar, Gokhan Ertugrul, Murat Dayangac

Onur Yaprak, Gokhan Ertugrul, Murat Dayangac, Medipol University Hospital, Center for Organ Transplantation, Istanbul 34214, Turkey

Sencan Acar, Atasehir Memorial Hospital, Center for Organ Transplantation, Istanbul 34758, Turkey

ORCID number: Onur Yaprak (0000-0003-1941-8290); Sencan Acar (0000-0001-8086-0956); Gokhan Ertugrul (0000-0002-8351-4220); Murat Dayangac (0000-0002-1240-7233).

Author contributions: Yaprak O designed the aim of this minireview; Dayangac M, Yaprak O, Acar S and Ertugrul G contributed equally to this work, reviewed the references, generated the tables, and wrote the manuscript.

Conflict-of-interest statement: No conflict of interest.

Manuscript source: Invited manuscript

Correspondence to: Onur Yaprak, MD, Associate Professor, Medipol University Hospital, Center for Organ Transplantation, Goztepe Mah. Metin Sk. No.4, Bagcilar, Istanbul 34214, Turkey. onuryaprak@hotmail.com
Telephone: +90-53-22239566

Received: April 11, 2018
Peer-review started: April 11, 2018
First decision: April 23, 2018
Revised: May 27, 2018
Accepted: June 13, 2018
Article in press: June 14, 2018

Published online: October 15, 2018

Abstract

The last two decades have seen a paradigm shift in the selection of patients with hepatocellular carcinoma (HCC) for liver transplantation. Microvascular invasion and differentiation have been the most significant factors affecting post-transplant recurrence; however, because of inherent disadvantages of pre-transplant biopsy, histological criteria never gained popularity. Recently, the selection criteria evolved from morphological to biological criteria, such as biomarkers and response to loco-regional therapy. With the introduction of multi-modality imaging, combination of computed tomography with nuclear medicine imaging, particularly, 18F-fluorodeoxyglucose positron emission tomography fulfilled an unmet need and rapidly became a critical component of HCC management. This review article will focus on the use of 18F-fluorodeoxyglucose positron emission tomography combined with computed tomography in the pre-transplant evaluation of HCC patients with special discussion on its ability to predict HCC recurrence after liver transplantation.

Key words: 18F-fluorodeoxyglucose positron emission tomography; Hepatocellular carcinoma; Recurrence; Liver transplantation

Core tip: The last two decades have seen a paradigm shift in the selection of patients with hepatocellular carcinoma (HCC) for liver transplantation. With the introduction of multimodality imaging, combination of computed tomography with nuclear medicine imaging

fulfilled an unmet need and rapidly became a critical component of HCC management. This review article will focus on the use of 18F-fluorodeoxyglucose positron emission tomography in the pre-transplant evaluation of HCC patients with special discussion on its ability to predict HCC recurrence after liver transplantation.

Yaprak O, Acar S, Ertugrul G, Dayangac M. Role of pre-transplant 18F-FDG PET/CT in predicting hepatocellular carcinoma recurrence after liver transplantation. *World J Gastrointest Oncol* 2018; 10(10): 336-343 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/336.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.336

INTRODUCTION

Hepatocellular carcinoma (HCC) is the most common type of primary liver cancer. Currently, HCC is the sixth most common cancer with more than a half million new cases diagnosed annually, and it is the second leading cause of cancer-related mortality in the world[1]. The global risk of HCC has been largely associated with hepatitis B and C virus infection. In addition, improved survival from cirrhosis and increasing rates of obesity and non-alcoholic fatty liver disease are expected to contribute to the ever-increasing incidence of HCC[2,3]. Because of the strong link between cirrhosis and HCC, liver transplantation (LT) is the best treatment option, since it removes the tumor and the underlying tumor-generating cirrhosis. Recently, HCC has been reported as the most common indication for LT in the United States[4].

Until the landmark study by Mazzaferro *et al*[5] in 1996, the liberal selection of HCC patients for LT resulted in high recurrence rates and poor survival. With the introduction of Milan criteria (MC), excellent long-term outcomes have been achieved that were not different from those of patients without HCC. The MC have been validated in several studies and widely accepted as the benchmark for selection of patients with HCC for deceased donor LT (DDLT). Subsequent studies searching for more liberal morphological criteria have shown that it was possible to extend the size and number of tumors without compromising post-transplant outcome[6-11] (Table 1). Despite being continually expanded, aforementioned morphological criteria have been criticized for a variety of reasons: they were restrictive and precluded numerous patients who otherwise would have benefited from LT with a low risk of HCC recurrence; they relied solely on tumor burden (defined as the size and number of tumors at a certain point) and excluded the factors related to tumor behavior (*i.e.,* tumor differentiation, molecular markers, and response to bridging therapy); they depended on imaging parameters that were inconsistent: in patients within MC, up to 40% had explant pathology that exceeded the MC, and in those beyond MC, up to 34% had explant pathology that was within the MC[12,13]. An earlier study investigating the correlation between pathologic and radiologic staging according to the morphological criteria have found that the accuracy of imaging classification for both Milan and (University of California San Francisco (UCSF) criteria was only 60%[14].

In patients with HCC, vascular invasion has been defined as one of the major determinants of the outcome after LT[15]. Further studies have shown that tumor differentiation has also been an independent predictor of recurrence and survival after the transplant[16,17]. Despite initial hesitancy against the use of pre-transplant tumor biopsy, Toronto criteria have led the way to the use of histological criteria in selection of patients with HCC for LT[12]. However, pre-transplant tumor biopsy has not gained popularity because of its limitations: In spite of the invasive biopsy procedures, the presence of vascular invasion and tumor differentiation may not be detected reliably; the sensitivity of biopsy varies depending on location of the tumor, needle size, and tumor size. Moreover, preoperative needle biopsy may increase tumor seeding and post-transplant recurrence[18]. Nevertheless, this was the beginning of a new era when there was a shift in selection criteria from morphological to the combination of biological and histomorphological criteria[19].

Meanwhile, major transplant centers in Asia started to expand aggressively the morphological criteria with the addition of biomarkers to the patient selection process. While in the West, alpha-fetoprotein (AFP) has been traditionally used as a reference biomarker to screen and support the diagnosis of HCC; in the East, des-gamma-carboxy prothrombin (DCP) was introduced as a significant marker for assessing the biological behavior of HCC, particularly in Japan. Shirabe *et al*[20] reported that selection of HCC patients for LT might improve with the use of DCP measurement because pre-transplant DCP level has been shown to be a significant predictor of microvascular invasion (MVI).

The utilization of a combination of biological and morphological data has been a perfect fit for living donor LT (LDLT), which was not restricted by deceased donor organ allocation system. The Kyoto group reported their selection criteria to include no more than 10 tumors, all less than 5 cm in diameter with DCP levels less than 400 ng/mL[21], while the Kyushu group suggested more extended criteria to include a tumor size of less than 5 cm and DCP levels less than 300 ng/mL with no limitation on the number of tumors[22]. Both centers achieved outstanding post-transplant outcomes. The criteria that incorporated biomarkers with expanded morphological criteria are shown in Table 2[21-24].

As the selection criteria have been continuously expanded, search for new criteria to predict the biological behavior of HCC also continued. To this end, response to loco-regional therapy (LRT) has been suggested as a surrogate marker of tumor biology[19]. Bridging therapies primarily focused on reducing the tumor burden and has

Table 1 Morphological criteria used in selection of patients with hepatocellular carcinoma for liver transplantation

Ref.	Year	Size and number
Milan[5]	1996	1 lesion ⩽ 5 cm, or 2 to 3 lesions each ⩽ 3
University of California San Francisco[6]	2001	1 lesion ⩽ 6.5 cm, 2-3 lesions each ⩽ 4.5 cm with total tumor diameter ⩽ 8 cm
Tokyo University[8]	2008	Up to 5 tumors, each < 5 cm
Asan Medical Center[9]	2008	The largest tumor diameter < 5 cm, tumor number ⩽ 6
Alberta[10]	2008	Total tumor volume < 115 cm
Valencia[11]	2008	Up to 3 tumors, each < 5 cm, and a cumulative tumor burden ⩽ 10 cm
Up-to-seven[7]	2009	7 as the sum of the size of the largest tumor and total number of tumors

Table 2 The use of biomarkers with expanded morphological criteria

Ref.	Year	No. of patients	Criteria	Overall survival	
				Within criteria	Beyond criteria
Kyoto[21]	2007	136	Up to 10 tumors, all ⩽ 5 cm; DCP ⩽ 400 ng/mL	87% (5-yr)	37% (5-yr)
Kyushu[22]	2007	40	Any number, tumor diameter ⩽ 5 cm; DCP < 300 ng/mL	77% (3-yr)	40% (3-yr)
Seoul[23]	2007	140	Any number, tumor diameter ⩽ 5 cm; AFP ⩽ 400 ng/mL	87% (5-yr)	23% (5-yr)
Hangzhou[24]	2008	195	Total tumor diameter ⩽ 8 cm; or total tumor diameter > 8 cm and grade Ⅰ/Ⅱ and AFP ⩽ 400 ng/mL	71% (5-yr)	19% (5-yr)

Table 3 The criteria used for prediction of biological behavior of hepatocellular carcinoma in the pre-transplant setting

Biomarkers (AFP, DCP)[21-24]
The neutrophil-lymphocyte ratio[27]
Pre-transplant liver biopsy[12]
Response to loco-regional therapy[19]
Test of time (3-mo waiting period)[19,26]
Dynamic evaluation (tumor doubling time and change in AFP)[19]
FDG-PET scan

AFP: Alpha-fetoprotein; DCP: Des-gamma-carboxy prothrombin; FDG-PET: Fluorodeoxyglucose positron emission tomography.

been recommended to downstage the HCC patients who exceeded the morphological selection criteria to within the MC to become eligible for DDLT[25]. In addition, long waiting times for DDLT and high dropout rates have led to an active approach to the treatment of HCC with LRT to prevent progression while awaiting LT. The LRTs have also been used in LDLT to exclude patients with unfavorable tumor behavior, such as the patients who are unresponsive to treatment or those with progression upon observation. The interval between therapy and LT was found to help in identifying the patients who have HCC with poor tumor biology with an increased risk of post-transplant recurrence[26].

Despite the ability of cross-sectional imaging studies to reliably diagnose HCC, neither computed tomography (CT), nor magnetic resonance imaging (MRI) have been instrumental as a marker of tumor biology[27] (Table 3). With the introduction of multimodality imaging, combination of CT with nuclear medicine imaging, particularly 18F-fluorodeoxyglucose positron emission tomography (18F-FDG-PET/CT), fulfilled an unmet need and rapidly became a critical component of HCC management[28]. This review article will focus on the use of 18F-FDG PET/CT in the setting of LT for HCC with special discussion on its ability to predict HCC recurrence after LT.

18F-FDG PET/CT IMAGING IN HCC

The successful application of 18F-FDG to a growing number of oncological indications has led to the widespread use of 18F-FDG-PET/CT in the diagnosis, staging and follow-up of patients with distinct types of cancer. Oncological imaging using 18F-FDG is based on the principle of enhanced glucose metabolism in tumors as compared with normal tissues. However, in normal hepatic parenchyma, where the concentration of glucose-6-phosphatase is high, the rapid clearance of 18F-FDG leads to a reduced discrimination between normal tissue and well-differentiated HCC. Because of the fact that low-grade HCC exhibits a lower FDG avidity, the general reported false-negative rate of 18F-FDG-PET/CT approaches 50% in the imaging of HCC[29]. The 18F-FDG uptake in HCC ranges from 38% to 70% with an overall sensitivity of only about 60%[29-32].

In the liver, PET/CT positivity is determined by examining whether the FDG uptake in tumor is significantly higher than that in the surrounding liver parenchyma. Standardized uptake values (SUV) of the lesions are calculated by plotting a circular region of interest (ROI) at the area of the maximum FDG uptake in the PET images. Numerous studies have defined PET/CT positivity *vs* PET/CT negativity by using the maximum SUV (SUVmax) within ROI. In a retrospective study of 280 patients undergoing LDLT for HCC, Lee *et al*[33] defined the SUVmax values for PET/CT positivity and negativity as 4.46 and 3.08, respectively ($P < 0.001$). However, SUV measurements are prone to be influenced by a variety of factors, including high glucose metabolism in the normal liver tissue, as well as the factors related with scanner and reconstruction parameters. Therefore,

Table 4 The standardized uptake values used to define clinically significant 18F-fluorodeoxyglucose positron emission tomography/computed tomography positivity for hepatocellular carcinoma

Ref.	Year	No. of patients	Study model	SUV values		
				SUVmax	TSUVmax-to-LSUVmax	TSUVmax-to-LSUVmean
Lee *et al*[34]	2009	59	LT	3	1.15	1.35
Song *et al*[35]	2012	83	LRT	4	1.45	1.9
Lee *et al*[36]	2015	280	LDLT	4.4		
Hsu *et al*[37]	2016	147	LDLT	4.8		2
Hong *et al*[38]	2016	123	LDLT		1.1	
Boussouar *et al*[39]	2016	28	LT		1.15	
Bailly *et al*[40]	2016	34	LT		1.15	
Lin *et al*[41]	2017	65	LT	3.8	1.49	1.69

SUV: Standardized uptake values; TSUVmax: Tumor SUVmax; LSUVmax: Normal-liver SUVmax.

many researchers suggested using either tumor SUVmax to normal-liver SUVmax (TSUVmax/LSUVmax) or tumor SUVmax to normal-liver SUVmean (TSUVmax/LSUVmean) values instead of SUVmax to identify PET/CT positivity[34-41] (Table 4).

While 18F-FDG-PET/CT has demonstrated substandard sensitivity in discovering new HCC, it has been useful in detecting extra-hepatic metastases, with detection rates reported as high as 100%[42,43]. 18F-FDG-PET/CT has also been reported to detect post-treatment recurrences earlier and at higher rates than conventional imaging modalities[44]. The sensitivity of 18F-FDG-PET/CT is size-dependent in both extra-hepatic metastases and recurrences. Sugiyama *et al*[42] reported a detection rate of 83% for extra-hepatic metastases > 1 cm, which was only 13% for lesions ≤ 1 cm in diameter. In patients with post-transplant HCC recurrence, Kim *et al*[45] reported that a detection rate of > 90% has been achieved for extra-hepatic metastases when the lesions were larger than 1 cm in diameter. However, 18F-FDG-PET/CT was not able to detect any of the extra-hepatic lesions under 1 cm and demonstrated a low detection rate of less than 10% for intrahepatic recurrences. They reported a detection rate of 100% in bone, 60% in the lungs, and 100% in lymph nodes. 18F-FDG-PET/CT has also been used in the evaluation of patients with unexplained AFP elevation after surgical or interventional treatment[46]. In HCC patients presenting with portal vein thrombosis, 18F-FDG-PET/CT was found more valuable than conventional imaging studies in differential diagnosis of tumor thrombus[47,48].

Considering the limited role of 18F-FDG-PET/CT in the detection of HCC because of its low overall sensitivity, Ho *et al*[49] advocated for the use of 11C-acetate, which showed better detection sensitivity of 87.3% compared to 47.3% using 18F-FDG. In another study from Hong Kong, which evaluated the accuracy of dual-tracer PET/CT in HCC patients who underwent either partial hepatectomy or LT, the sensitivity of 11C-acetate PET/CT was significantly higher than those of 18F-FDG-PET/CT and contrast-enhanced CT for the detection of small HCCs (87.0% *vs* 17.4% and 43.5%, respectively)[50]. Recent studies have concluded that in patients undergoing LT for HCC, although 11C-choline PET had a better detection rate for well-differentiated lesions and the addition of 11C-acetate to 18F-FDG-PET/CT significantly increased the overall sensitivity and specificity for the detection of HCC, the complementary role of 18F-FDG should not be underestimated as a marker of poorly differentiated tumor pathology[51-53].

CORRELATION BETWEEN 18F-FDG PET/CT AND HISTOLOGICAL FINDINGS

In HCC, the growth rate and the activity of glycolytic enzymes are related[54]. Therefore, contrary to well differentiated HCC, poorly differentiated HCC cells have low glucose-6 phosphatase activity and high uptake of 18F-FDG[30]. Recent studies have suggested that maximum standardized uptake values in 18F-FDG PET/CT imaging demonstrated strong correlation with histopathological characteristics of HCC, such as MVI and tumor grade[28,55-57]. The reported accuracy rate of 18F-FDG-PET/CT for detection of MVI invasion and tumor differentiation in HCC ranged between 68.3% to 88.1% and 57.4% to 71.4%, respectively[55].

Considering the risk of tumor seeding and limitations related to multifocality and microscopic heterogeneity within tumor, 18F-FDG-PET/CT is a more valuable tool in the prediction of tumor biology. The maximum standardized uptake value (SUVmax) and ratio of tumor-to-normal liver SUVmax value (SUVmax T/L) have been recognized as objective indices for the definition of 18F-FDG-PET/CT positivity. In a recent study on 65 HCC patients who underwent 18F-FDG-PET/CT before LT, Lin *et al*[41] have found that the SUVmax T/L ratio was an independent predictor of vascular invasion. The optimal cutoff values for SUVmax of the tumor and SUVmax T/L ratio for the prediction of HCC vascular invasion were 3.80 and 1.49, respectively. In another study that reviewed 18F-FDG-PET/CT findings of 34 patients with HCC who underwent LT, Bailly *et al*[40] reported that none of the patients with SUVmax L/T ratio > 1.15 had well differentiated HCC.

A study from Seoul National University investigated the association of the gadoxetic acid-enhanced MR and

Table 5 The use of 18F-fluorodeoxyglucose positron emission tomography/computed tomography in predicting post-transplant hepatocellular carcinoma recurrences

Ref.	Year	Follow-up (mo)	Recurrence		Disease-free survival	Risk of recurrence (95%CI)
			PET/CT (+)	PET/CT (-)		
Yang *et al*[28]	2006	19	13/8	25/3	2-yr, 46.1% *vs* 85.1%	OR = 7.6 (1.9-28.9)
Kornberg *et al*[56]	2009	11.5	19/9	36/1	3-yr, 46.9% *vs* 93.3%	OR = 23.9 (2.1-268.5)
Lee *et al*[34]	2013	26.1	55/22	136/16	3-yr, 57.1% *vs* 86.8%	HR = 3.9 (1.1-13.0)
Hsu *et al*[37]	2016	25.8	30/9	117/9	5-yr, 68.3 *vs* 84.8%	HR = 13.5 (4.7-38.2)
Kornberg *et al*[57]	2017	74	41/24	75/5	5-yr, 38.1% *vs* 93.3%	HR = 22.8 (6.3-83.0)
Ye *et al*[63]	2017	25.7	78/46	25/7	5-yr, 21.9% *vs* 76%	HR = 3.6 (1.3-9.6)

PET/CT: Positron emission tomography/computed tomography.

the 18F-FDG-PET/CT findings with the MVI in patients who underwent LT for HCC[58]. Multivariate analysis revealed that peritumoral enhancement and the ratio of tumor maximum standardized uptake value (SUV) to normal liver mean SUV (TSUVmax/LSUVmean) ⩾ 1.2 had a statistically significant association with MVI, with an odds ratio of 10.6 and 14.2, respectively. With regard to predicting MVI, the sensitivity and specificity was 35.7% and 93.3% for MRI and 64.3% and 86.7% for PET/CT, respectively. For the prediction of MVI, a sensitivity of 78.6% and a specificity of 80% were achieved when both imaging modalities were combined.

CORRELATION BETWEEN 18F-FDG PET/CT AND MORPHOLOGICAL CRITERIA

As the selection criteria for LT shifted towards biological criteria, MC as the current gold standard and other morphological criteria have been challenged with a number of studies using 18F-FDG PET/CT. Kornberg *et al*[59] was the first to investigate the prognostic value of preoperative 18F-FDG PET/CT in liver transplant candidates with HCC. They concluded that PET/CT negative patients with HCC beyond MC might achieve excellent post-transplant disease-free survival (DFS). In a more recent study, they combined the pre-transplant 18F-FDG-PET/CT assessments with Up-to-seven criteria[60]. Among 116 patients with HCC who underwent 18F-FDG-PET/CT prior to LT, 5-year DFS was comparable between patients within Up-to-seven criteria (n = 85) and those beyond Up-to-seven criteria with negative PET/CT (n = 16) (81.0% *vs* 87.1%, P = 0.5).

A Japanese multicenter study including 182 LDLT recipients from 16 Japanese LT centers investigated the significance of pre-transplant 18F-FDG-PET/CT at a much larger scale. While patients beyond MC had a significantly higher recurrence rate at 5 years compared with those within MC (38% *vs* 7%, P < 0.001), a subgroup of "beyond MC" patients with negative PET/CT and low AFP (< 115 ng/mL) showed similar recurrence rate with "within MC" patients (19%, P = 0.1)[61]. Similar data were recently published by the Taiwan group who combined pre-transplant PET/CT results with UCSF criteria for predicting the risk of post-transplant HCC recurrence. In a group of 147 patients with HCC who underwent 18F-FDG-PET/CT and proceeded to LDLT, patients within UCSF criteria and those beyond UCSF criteria with a low FDG uptake had similar post-transplant recurrence rates (3.6% *vs* 11.1%)[37].

Another study from Korea investigated the clinical impact of 18F-FDG-PET/CT in patients undergoing LDLT for advanced HCC, where more than half of the patients were beyond MC. In patients beyond either MC (n = 147) or UCSF (n = 136) criteria, PET/CT negative patients had 5-year DFS rates of 73.3% and 72.8%, respectively. Despite the fact that these figures were significantly lower than those of patients within MC (89.8%), the outcome is highly acceptable when the discussion shifts from "zero recurrence" towards targeting 50% 5-year survival as an acceptable goal in advanced HCC[33].

ROLE OF 18F-FDG PET/CT IN PREDICTING POST-TRANSPLANT HCC RECURRENCE

Seoul National University Hospital was the first to report the effectiveness of pre-transplant 18F-FDG-PET/CT to predict post-transplant HCC recurrence[28]. Further studies have shown that a high 18F-FDG uptake on pre-transplant PET/CT was a strong predictive factor for MVI and tumor recurrence after LT[56,33,62] (Table 5).

In a cohort of 116 liver transplant patients with HCC, Kornberg *et al*[60] reported a 5-year DFS rate of 93.3% in PET/CT negative patients *vs* 38.1% in PET/CT positive patients. PET/CT positive patients showed a recurrence rate of 58.5%, while only 6.7% of the PET/CT negative patients had recurrence. Ye *et al*[63] also investigated the clinical value of pre-transplant PET/CT in the selection and prognostic prediction of patients with advanced

HCC in the LT setting. Patients with a positive 18F-FDG-PET/CT had significantly increased risk of post-transplant recurrence compared to PET/CT negative patients (59.0% *vs* 28.0%, $P = 0.007$). In patients with positive PET/CT, they reported a significantly lower 5-year DFS rate than that of patients with negative PET/CT (76.0% *vs* 21.9%, $P < 0.001$). In another study investigating the role of PET/CT as a prognostic factor for early HCC recurrence after LT, Lee *et al*[62] have shown that median SUVmax of PET/CT-positive tumors in the early, late, and no recurrence groups was 5.2, 3.7, and 3.2, respectively. They concluded that preoperative 18F-FDG-PET/CT was an independent and significant prognostic factor for early tumor recurrence after LT for HCC.

Hong *et al*[38] further developed the concept, hypothesizing that the combination of 18F-FDG PET/CT positivity and serum AFP level might improve the prediction of post-LT outcome for patients with HCC. Using cut-off values of 200 ng/mL for AFP and 1.1 for SUVmax T/L ratio for the definition of "high-risk" HCC, they found that the rate of MVI and poor differentiation was 33% and 92%, respectively in the high-risk group. They reported 5-year DFS rates of 49.1% *vs* 93.4% in PET/CT positive *vs* negative patients and 47.7% *vs* 88.3% in high AFP *vs* low AFP patients. In the high-risk group ($n = 12$), 5-year DFS rate was only 8.4%.

CONCLUSION

In patients with HCC, LT is the best treatment option. The selection criteria for LT have been shifting from morphological to the combination of biological and histomorphological criteria. When combined with serum markers, 18F-FDG-PET/CT represents the "new generation" of biological criteria, which has the potential to further improve the prediction of tumor behavior and to provide a better risk stratification model for HCC.

REFERENCES

1 **Mittal S**, El-Serag HB. Epidemiology of hepatocellular carcinoma: consider the population. *J Clin Gastroenterol* 2013; **47** Suppl: S2-S6 [PMID: 23632345 DOI: 10.1097/MCG.0b013e3182872f29]

2 **El-Serag HB**, Kanwal F. Epidemiology of hepatocellular carcinoma in the United States: where are we? Where do we go? *Hepatology* 2014; **60**: 1767-1775 [PMID: 24839253 DOI: 10.1002/hep.27222]

3 **McGlynn KA**, London WT. The global epidemiology of hepatocellular carcinoma: present and future. *Clin Liver Dis* 2011; **15**: 223-243, vii-vix [PMID: 21689610 DOI: 10.1016/j.cld.2011.03.006]

4 **Yang JD**, Larson JJ, Watt KD, Allen AM, Wiesner RH, Gores GJ, Roberts LR, Heimbach JA, Leise MD. Hepatocellular Carcinoma Is the Most Common Indication for Liver Transplantation and Placement on the Waitlist in the United States. *Clin Gastroenterol Hepatol* 2017; **15**: 767-775.e3 [PMID: 28013117 DOI: 10.1016/j.cgh.2016.11.034]

5 **Mazzaferro V**, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, Montalto F, Ammatuna M, Morabito A, Gennari L. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. *N Engl J Med* 1996; **334**: 693-699 [PMID: 8594428 DOI: 10.1056/NEJM199603143341104]

6 **Yao FY**, Ferrell L, Bass NM, Watson JJ, Bacchetti P, Venook A, Ascher NL, Roberts JP. Liver transplantation for hepatocellular carcinoma: expansion of the tumor size limits does not adversely impact survival. *Hepatology* 2001; **33**: 1394-1403 [PMID: 11391528 DOI: 10.1053/jhep.2001.24563]

7 **Mazzaferro V**, Llovet JM, Miceli R, Bhoori S, Schiavo M, Mariani L, Camerini T, Roayaie S, Schwartz ME, Grazi GL, Adam R, Neuhaus P, Salizzoni M, Bruix J, Forner A, De Carlis L, Cillo U, Burroughs AK, Troisi R, Rossi M, Gerunda GE, Lerut J, Belghiti J, Boin I, Gugenheim J, Rochling F, Van Hoek B, Majno P; Metroticket Investigator Study Group. Predicting survival after liver transplantation in patients with hepatocellular carcinoma beyond the Milan criteria: a retrospective, exploratory analysis. *Lancet Oncol* 2009; **10**: 35-43 [PMID: 19058754 DOI: 10.1016/S1470-2045(08)70284-5]

8 **Sugawara Y**, Kokudo N. Surgical treatment of hepatocellular carcinoma: comparison of resection and transplantation. *Oncology* 2008; **75** Suppl 1: 119-123 [PMID: 19092281 DOI: 10.1159/000173433]

9 **Lee SG**, Hwang S, Moon DB, Ahn CS, Kim KH, Sung KB, Ko GY, Park KM, Ha TY, Song GW. Expanded indication criteria of living donor liver transplantation for hepatocellular carcinoma at one large-volume center. *Liver Transpl* 2008; **14**: 935-945 [PMID: 18581465 DOI: 10.1002/lt.21445]

10 **Toso C**, Trotter J, Wei A, Bigam DL, Shah S, Lancaster J, Grant DR, Greig PD, Shapiro AM, Kneteman NM. Total tumor volume predicts risk of recurrence following liver transplantation in patients with hepatocellular carcinoma. *Liver Transpl* 2008; **14**: 1107-1115 [PMID: 18668667 DOI: 10.1002/lt.21484]

11 **Silva M**, Moya A, Berenguer M, Sanjuan F, López-Andujar R, Pareja E, Torres-Quevedo R, Aguilera V, Montalva E, De Juan M, Mattos A, Prieto M, Mir J. Expanded criteria for liver transplantation in patients with cirrhosis and hepatocellular carcinoma. *Liver Transpl* 2008; **14**: 1449-1460 [PMID: 18825681 DOI: 10.1002/lt.21576]

12 **DuBay D**, Sandroussi C, Sandhu L, Cleary S, Guba M, Cattral MS, McGilvray I, Ghanekar A, Selzner M, Greig PD, Grant DR. Liver transplantation for advanced hepatocellular carcinoma using poor tumor differentiation on biopsy as an exclusion criterion. *Ann Surg* 2011; **253**: 166-172 [PMID: 21294289 DOI: 10.1097/SLA.0b013e31820508f1]

13 **Bruix J**, Llovet JM. Prognostic prediction and treatment strategy in hepatocellular carcinoma. *Hepatology* 2002; **35**: 519-524 [PMID: 11870363 DOI: 10.1053/jhep.2002.32089]

14 **Sotiropoulos GC**, Malagó M, Molmenti E, Paul A, Nadalin S, Brokalaki E, Kühl H, Dirsch O, Lang H, Broelsch CE. Liver transplantation for hepatocellular carcinoma in cirrhosis: is clinical tumor classification before transplantation realistic? *Transplantation* 2005; **79**: 483-487 [PMID: 15729176 DOI: 10.1097/01.TP.0000152801.82734.74]

15 **Hemming AW**, Cattral MS, Reed AI, Van Der Werf WJ, Greig PD, Howard RJ. Liver transplantation for hepatocellular carcinoma. *Ann Surg* 2001; **233**: 652-659 [PMID: 11323504 DOI: 10.1097/00000658-200105000-00009]

16 **Roayaie S**, Schwartz JD, Sung MW, Emre SH, Miller CM, Gondolesi GE, Krieger NR, Schwartz ME. Recurrence of hepatocellular carcinoma after liver transplant: patterns and prognosis. *Liver Transpl* 2004; **10**: 534-540 [PMID: 15048797 DOI: 10.1002/lt.20128]

17 **Cillo U**, Vitale A, Bassanello M, Boccagni P, Brolese A, Zanus G, Burra P, Fagiuoli S, Farinati F, Rugge M, D'Amico DF. Liver transplantation for the treatment of moderately or well-differentiated hepatocellular carcinoma. *Ann Surg* 2004; **239**: 150-159 [PMID: 14745321 DOI: 10.1097/01.sla.0000109146.72827.76]

18 **Clavien PA**, Lesurtel M, Bossuyt PM, Gores GJ, Langer B, Perrier A; OLT for HCC Consensus Group. Recommendations for liver transplantation for hepatocellular carcinoma: an international consensus conference report. *Lancet Oncol* 2012; **13**: e11-e22 [PMID: 22047762 DOI: 10.1016/S1470-2045(11)70175-9]

19 **Cillo U**, Giuliani T, Polacco M, Herrero Manley LM, Crivellari G, Vitale A. Prediction of hepatocellular carcinoma biological behavior in patient selection for liver transplantation. *World J Gastroenterol* 2016; **22**: 232-252 [PMID: 26755873 DOI: 10.3748/wjg.v22.i1.232]

20 **Shirabe K**, Itoh S, Yoshizumi T, Soejima Y, Taketomi A, Aishima S, Maehara Y. The predictors of microvascular invasion in candidates

for liver transplantation with hepatocellular carcinoma-with special reference to the serum levels of des-gamma-carboxy prothrombin. *J Surg Oncol* 2007; **95**: 235-240 [PMID: 17323337 DOI: 10.1002/jso.20655]

21 **Kaido T**, Ogawa K, Mori A, Fujimoto Y, Ito T, Tomiyama K, Takada Y, Uemoto S. Usefulness of the Kyoto criteria as expanded selection criteria for liver transplantation for hepatocellular carcinoma. *Surgery* 2013; **154**: 1053-1060 [PMID: 24074704 DOI: 10.1016/j.surg.2013.04.056]

22 **Uchiyama H**, Itoh S, Yoshizumi T, Ikegami T, Harimoto N, Soejima Y, Harada N, Morita K, Toshima T, Motomura T, Maehara Y. Living donor liver transplantation for hepatocellular carcinoma: results of prospective patient selection by Kyushu University Criteria in 7 years. *HPB* (Oxford) 2017; **19**: 1082-1090 [PMID: 28888776 DOI: 10.1016/j.hpb.2017.08.004]

23 **Kwon CH**, Kim DJ, Han YS, Park JB, Choi GS, Kim SJ, Joh JW, Lee SK. HCC in living donor liver transplantation: can we expand the Milan criteria? *Dig Dis* 2007; **25**: 313-319 [PMID: 17960066 DOI: 10.1159/000106911]

24 **Zheng SS**, Xu X, Wu J, Chen J, Wang WL, Zhang M, Liang TB, Wu LM. Liver transplantation for hepatocellular carcinoma: Hangzhou experiences. *Transplantation* 2008; **85**: 1726-1732 [PMID: 18580463 DOI: 10.1097/TP.0b013e31816b67e4]

25 **Heimbach JK**, Kulik LM, Finn RS, Sirlin CB, Abecassis MM, Roberts LR, Zhu AX, Murad MH, Marrero JA. AASLD guidelines for the treatment of hepatocellular carcinoma. *Hepatology* 2018; **67**: 358-380 [PMID: 28130846 DOI: 10.1002/hep.29086]

26 **Pomfret EA**, Washburn K, Wald C, Nalesnik MA, Douglas D, Russo M, Roberts J, Reich DJ, Schwartz ME, Mieles L, Lee FT, Florman S, Yao F, Harper A, Edwards E, Freeman R, Lake J. Report of a national conference on liver allocation in patients with hepatocellular carcinoma in the United States. *Liver Transpl* 2010; **16**: 262-278 [PMID: 20209641 DOI: 10.1002/lt.21999]

27 **Halazun KJ**, Hardy MA, Rana AA, Woodland DC 4th, Luyten EJ, Mahadev S, Witkowski P, Siegel AB, Brown RS Jr, Emond JC. Negative impact of neutrophil-lymphocyte ratio on outcome after liver transplantation for hepatocellular carcinoma. *Ann Surg* 2009; **250**: 141-151 [PMID: 19561458 DOI: 10.1097/SLA.0b013e3181a77e59]

28 **Yang SH**, Suh KS, Lee HW, Cho EH, Cho JY, Cho YB, Yi NJ, Lee KU. The role of (18)F-FDG-PET imaging for the selection of liver transplantation candidates among hepatocellular carcinoma patients. *Liver Transpl* 2006; **12**: 1655-1660 [PMID: 16964589 DOI: 10.1002/lt.20861]

29 **Khan MA**, Combs CS, Brunt EM, Lowe VJ, Wolverson MK, Solomon H, Collins BT, Di Bisceglie AM. Positron emission tomography scanning in the evaluation of hepatocellular carcinoma. *J Hepatol* 2000; **32**: 792-797 [PMID: 10845666 DOI: 10.1016/S0168-8278(00)80248-2]

30 **Torizuka T**, Tamaki N, Inokuma T, Magata Y, Sasayama S, Yonekura Y, Tanaka A, Yamaoka Y, Yamamoto K, Konishi J. In vivo assessment of glucose metabolism in hepatocellular carcinoma with FDG-PET. *J Nucl Med* 1995; **36**: 1811-1817 [PMID: 7562048]

31 **Talbot JN**, Fartoux L, Balogova S, Nataf V, Kerrou K, Gutman F, Huchet V, Ancel D, Grange JD, Rosmorduc O. Detection of hepatocellular carcinoma with PET/CT: a prospective comparison of 18F-fluorocholine and 18F-FDG in patients with cirrhosis or chronic liver disease. *J Nucl Med* 2010; **51**: 1699-1706 [PMID: 20956466 DOI: 10.2967/jnumed.110.075507]

32 **Blechacz B**, Gores GJ. Positron emission tomography scan for a hepatic mass. *Hepatology* 2010; **52**: 2186-2191 [PMID: 20967825 DOI: 10.1002/hep.24002]

33 **Lee SD**, Kim SH, Kim SK, Kim YK, Park SJ. Clinical Impact of 18F-Fluorodeoxyglucose Positron Emission Tomography/Computed Tomography in Living Donor Liver Transplantation for Advanced Hepatocellular Carcinoma. *Transplantation* 2015; **99**: 2142-2149 [PMID: 25905981 DOI: 10.1097/TP.0000000000000719]

34 **Lee JW**, Paeng JC, Kang KW, Kwon HW, Suh KS, Chung JK, Lee MC, Lee DS. Prediction of tumor recurrence by 18F-FDG PET in liver transplantation for hepatocellular carcinoma. *J Nucl Med* 2009; **50**: 682-687 [PMID: 19372474 DOI: 10.2967/jnumed.108.060574]

35 **Song MJ**, Bae SH, Yoo IeR, Park CH, Jang JW, Chun HJ, Choi BG, Lee HG, Choi JY, Yoon SK. Predictive value of ^{18}F-fluorodeoxyglucose PET/CT for transarterial chemolipiodolization of hepatocellular carcinoma. *World J Gastroenterol* 2012; **18**: 3215-3222 [PMID: 22783045 DOI: 10.3748/wjg.v18.i25.3215]

36 **Lee SD**, Lee B, Kim SH, Joo J, Kim SK, Kim YK, Park SJ. Proposal of new expanded selection criteria using total tumor size and (18)F-fluorodeoxyglucose - positron emission tomography/computed tomography for living donor liver transplantation in patients with hepatocellular carcinoma: The National Cancer Center Korea criteria. *World J Transplant* 2016; **6**: 411-422 [PMID: 27358787 DOI: 10.5500/wjt.v6.i2.411]

37 **Hsu CC**, Chen CL, Wang CC, Lin CC, Yong CC, Wang SH, Liu YW, Lin TL, Lee WF, Lin YH, Chan YC, Wu YJ, Eng HL, Cheng YF. Combination of FDG-PET and UCSF Criteria for Predicting HCC Recurrence After Living Donor Liver Transplantation. *Transplantation* 2016; **100**: 1925-1932 [PMID: 27306534 DOI: 10.1097/TP.0000000000001297]

38 **Hong G**, Suh KS, Suh SW, Yoo T, Kim H, Park MS, Choi Y, Paeng JC, Yi NJ, Lee KW. Alpha-fetoprotein and (18)F-FDG positron emission tomography predict tumor recurrence better than Milan criteria in living donor liver transplantation. *J Hepatol* 2016; **64**: 852-859 [PMID: 26658686 DOI: 10.1016/j.jhep.2015.11.033]

39 **Boussouar S**, Itti E, Lin SJ, Decaens T, Evangelista E, Chiaradia M, Chalaye J, Baranes L, Calderaro J, Laurent A, Pigneur F, Duvoux C, Azoulay D, Costentin C, Rahmouni A, Luciani A. Functional imaging of hepatocellular carcinoma using diffusion-weighted MRI and (18)F-FDG PET/CT in patients on waiting-list for liver transplantation. *Cancer Imaging* 2016; **16**: 4 [PMID: 26883745 DOI: 10.1186/s40644-016-0062-8]

40 **Bailly M**, Venel Y, Orain I, Salamé E, Ribeiro MJ. 18F-FDG PET in Liver Transplantation Setting of Hepatocellular Carcinoma: Predicting Histology? *Clin Nucl Med* 2016; **41**: e126-e129 [PMID: 26545024 DOI: 10.1097/RLU.0000000000001040]

41 **Lin CY**, Liao CW, Chu LY, Yen KY, Jeng LB, Hsu CN, Lin CL, Kao CH. Predictive Value of 18F-FDG PET/CT for Vascular Invasion in Patients With Hepatocellular Carcinoma Before Liver Transplantation. *Clin Nucl Med* 2017; **42**: e183-e187 [PMID: 28114226 DOI: 10.1097/RLU.0000000000001545]

42 **Sugiyama M**, Sakahara H, Torizuka T, Kanno T, Nakamura F, Futatsubashi M, Nakamura S. 18F-FDG PET in the detection of extrahepatic metastases from hepatocellular carcinoma. *J Gastroenterol* 2004; **39**: 961-968 [PMID: 15549449 DOI: 10.1007/s00535-004-1427-5]

43 **Yoon KT**, Kim JK, Kim DY, Ahn SH, Lee JD, Yun M, Rha SY, Chon CY, Han KH. Role of 18F-fluorodeoxyglucose positron emission tomography in detecting extrahepatic metastasis in pretreatment staging of hepatocellular carcinoma. *Oncology* 2007; **72** Suppl 1: 104-110 [PMID: 18087190 DOI: 10.1159/000111715]

44 **Paudyal B**, Oriuchi N, Paudyal P, Tsushima Y, Iida Y, Higuchi T, Hanaoka H, Miyakubo M, Takano A, Ishikita T, Endo K. Early diagnosis of recurrent hepatocellular carcinoma with 18F-FDG PET after radiofrequency ablation therapy. *Oncol Rep* 2007; **18**: 1469-1473 [PMID: 17982632 DOI: 10.3892/or.18.6.1469]

45 **Kim YK**, Lee KW, Cho SY, Han SS, Kim SH, Kim SK, Park SJ. Usefulness 18F-FDG positron emission tomography/computed tomography for detecting recurrence of hepatocellular carcinoma in posttransplant patients. *Liver Transpl* 2010; **16**: 767-772 [PMID: 20517911 DOI: 10.1002/lt.22069]

46 **Chen YK**, Hsieh DS, Liao CS, Bai CH, Su CT, Shen YY, Hsieh JF, Liao AC, Kao CH. Utility of FDG-PET for investigating unexplained serum AFP elevation in patients with suspected hepatocellular carcinoma recurrence. *Anticancer Res* 2005; **25**: 4719-4725 [PMID: 16334166]

47 **Hanajiri K**, Mitsui H, Maruyama T, Kondo Y, Shiina S, Omata M, Nakagawa K. 18F-FDG PET for hepatocellular carcinoma presenting with portal vein tumor thrombus. *J Gastroenterol* 2005; **40**: 1005-1006 [PMID: 16261443 DOI: 10.1007/s00535-005-1667-z]

48 **Kurtovic J**, Van Der Wall H, Riordan SM. FDG PET for discri-

mination between tumor extension and blood thrombus as a cause for portal vein thrombosis in hepatocellular carcinoma: important role in exclusion of transplant candidacy. *Clin Nucl Med* 2005; **30**: 408-410 [PMID: 15891293 DOI: 10.1097/01.rlu.0000162606.83862.a7]

49 **Ho CL**, Yu SC, Yeung DW. 11C-acetate PET imaging in hepatocellular carcinoma and other liver masses. *J Nucl Med* 2003; **44**: 213-221 [PMID: 12571212]

50 **Cheung TT**, Ho CL, Lo CM, Chen S, Chan SC, Chok KS, Fung JY, Yan Chan AC, Sharr W, Yau T, Poon RT, Fan ST. 11C-acetate and 18F-FDG PET/CT for clinical staging and selection of patients with hepatocellular carcinoma for liver transplantation on the basis of Milan criteria: surgeon's perspective. *J Nucl Med* 2013; **54**: 192-200 [PMID: 23321459 DOI: 10.2967/jnumed.112.107516]

51 **Park JW**, Kim JH, Kim SK, Kang KW, Park KW, Choi JI, Lee WJ, Kim CM, Nam BH. A prospective evaluation of 18F-FDG and 11C-acetate PET/CT for detection of primary and metastatic hepatocellular carcinoma. *J Nucl Med* 2008; **49**: 1912-1921 [PMID: 18997056 DOI: 10.2967/jnumed.108.055087]

52 **Wu HB**, Wang QS, Li BY, Li HS, Zhou WL, Wang QY. F-18 FDG in conjunction with 11C-choline PET/CT in the diagnosis of hepatocellular carcinoma. *Clin Nucl Med* 2011; **36**: 1092-1097 [PMID: 22064078 DOI: 10.1097/RLU.0b013e3182335df4]

53 **Yamamoto Y**, Nishiyama Y, Kameyama R, Okano K, Kashiwagi H, Deguchi A, Kaji M, Ohkawa M. Detection of hepatocellular carcinoma using 11C-choline PET: comparison with 18F-FDG PET. *J Nucl Med* 2008; **49**: 1245-1248 [PMID: 18632827 DOI: 10.2967/jnumed.108.052639]

54 **Sweeney MJ**, Ashmore J, Morrıs HP, Weber G. Comparatıve biochemıstry hepatomas. IV. isotope studies of glucose and fructose metabolism in liver tumors of different growth rates. *Cancer Res* 1963; **23**: 995-1002 [PMID: 14050771]

55 **Lee SD**, Kim SH. Role of positron emission tomography/computed tomography in living donor liver transplantation for hepatocellular carcinoma. *Hepatobiliary Surg Nutr* 2016; **5**: 408-414 [PMID: 27826555 DOI: 10.21037/hbsn.2016.08.01]

56 **Kornberg A**, Freesmeyer M, Bärthel E, Jandt K, Katenkamp K, Steenbeck J, Sappler A, Habrecht O, Gottschild D, Settmacher U. 18F-FDG-uptake of hepatocellular carcinoma on PET predicts microvascular tumor invasion in liver transplant patients. *Am J Transplant* 2009; **9**: 592-600 [PMID: 19191771 DOI: 10.1111/j.1600-6143.2008.02516.x]

57 **Kornberg A**, Küpper B, Tannapfel A, Büchler P, Krause B, Witt U, Gottschild D, Friess H. Patients with non-[18 F]fludeoxyglucose-avid advanced hepatocellular carcinoma on clinical staging may achieve long-term recurrence-free survival after liver transplantation. *Liver Transpl* 2012; **18**: 53-61 [PMID: 21850692 DOI: 10.1002/lt.22416]

58 **Ahn SY**, Lee JM, Joo I, Lee ES, Lee SJ, Cheon GJ, Han JK, Choi BI. Prediction of microvascular invasion of hepatocellular carcinoma using gadoxetic acid-enhanced MR and (18)F-FDG PET/CT. *Abdom Imaging* 2015; **40**: 843-851 [PMID: 25253426 DOI: 10.1007/s00261-014-0256-0]

59 **Kornberg A**, Küpper B, Thrum K, Katenkamp K, Steenbeck J, Sappler A, Habrecht O, Gottschild D. Increased 18F-FDG uptake of hepatocellular carcinoma on positron emission tomography independently predicts tumor recurrence in liver transplant patients. *Transplant Proc* 2009; **41**: 2561-2563 [PMID: 19715974 DOI: 10.1016/j.transproceed.2009.06.115]

60 **Kornberg A**, Witt U, Schernhammer M, Kornberg J, Ceyhan GO, Mueller K, Friess H, Thrum K. Combining 18F-FDG positron emission tomography with Up-to-seven criteria for selecting suitable liver transplant patients with advanced hepatocellular carcinoma. *Sci Rep* 2017; **7**: 14176 [PMID: 29074969 DOI: 10.1038/s41598-017-14430-9]

61 **Takada Y**, Kaido T, Shirabe K, Nagano H, Egawa H, Sugawara Y, Taketomi A, Takahara T, Wakabayashi G, Nakanishi C, Kawagishi N, Kenjo A, Gotoh M, Toyoki Y, Hakamada K, Ohtsuka M, Akamatsu N, Kokudo N, Takeda K, Endo I, Takamura H, Okajima H, Wada H, Kubo S, Kuramitsu K, Ku Y, Ishiyama K, Ohdan H, Ito E, Maehara Y, Honda M, Inomata Y, Furukawa H, Uemoto S, Yamaue H, Miyazaki M, Takada T; LTx-PET study group of the Japanese Society of Hepato-Biliary-Pancreatic Surgery and the Japanese Liver Transplantation Society. Significance of preoperative fluorodeoxyglucose-positron emission tomography in prediction of tumor recurrence after liver transplantation for hepatocellular carcinoma patients: a Japanese multicenter study. *J Hepatobiliary Pancreat Sci* 2017; **24**: 49-57 [PMID: 27806426 DOI: 10.1002/jhbp.412]

62 **Lee SD**, Kim SH, Kim YK, Kim C, Kim SK, Han SS, Park SJ. (18)F-FDG-PET/CT predicts early tumor recurrence in living donor liver transplantation for hepatocellular carcinoma. *Transpl Int* 2013; **26**: 50-60 [PMID: 23106431 DOI: 10.1111/j.1432-2277.2012.01572.x]

63 **Ye YF**, Wang W, Wang T, Yu J, Geng L, Yu SF, Yan S, Zheng SS. Role of 18F fludeoxyglucose positron emission tomography in the selection of liver transplantation candidates in patients with hepatocellular carcinoma. *Hepatobiliary Pancreat Dis Int* 2017; **16**: 257-263 [PMID: 28603093 DOI: 10.1016/S1499-3872(17)60011-0]

P- Reviewer: Chen JN, Chok KSH, Chiu KW, Tallon-Aguilar L, Xu X, Yeo W **S- Editor**: Ji FF **L- Editor**: Filipodia **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 344-350

DOI: 10.4251/wjgo.v10.i10.344

ISSN 1948-5204 (online)

ORIGINAL ARTICLE

Basic Study

miR-122-5p as a novel biomarker for alpha-fetoprotein-producing gastric cancer

Suguru Maruyama, Shinji Furuya, Kensuke Shiraishi, Hiroki Shimizu, Hidenori Akaike, Naohiro Hosomura, Yoshihiko Kawaguchi, Hidetake Amemiya, Hiromichi Kawaida, Makoto Sudo, Shingo Inoue, Hiroshi Kono, Daisuke Ichikawa

Suguru Maruyama, Shinji Furuya, Kensuke Shiraishi, Hiroki Shimizu, Hidenori Akaike, Naohiro Hosomura, Yoshihiko Kawaguchi, Hidetake Amemiya, Hiromichi Kawaida, Makoto Sudo, Shingo Inoue, Hiroshi Kono, Daisuke Ichikawa, First Department of Surgery, Faculty of Medicine University of Yamanashi, Yamanashi 409-3898, Japan

ORCID number: Suguru Maruyama (0000-0002-9926-7463); Shinji Furuya (0000-0002-6766-3779); Kensuke Shiraishi (0000-0002-8530-362X); Hiroki Shimizu (0000-0001-8553-6864); Hidenori Akaike (0000-0001-9713-7190); Naohiro Hosomura (0000-0003-0483-3635); Yoshihiko Kawaguchi (0000-0001-8788-9524); Hidetake Amemiya (0000-0002-4320-755X); Hiromichi Kawaida (0000-0003-3507-0167); Makoto Sudo (0000-0002-0200-6460); Shingo Inoue (0000-0002-1968-2182); Hiroshi Kono (0000-0001-6843-0814); Daisuke Ichikawa (0000-0003-0093-2206).

Author contributions: Maruyama S performed the majority of experiments and wrote the manuscript; Furuya S performed the research; Shiraishi K, Akaike H and Kawaguchi Y provided tissue samples and clinical data; Shimizu H, Hosomura N, Amemiya H, Kawaida H, Sudo M, Inoue S and Kono H made substantial contributions to the data analysis and interpretation; Ichikawa D designed the research and helped to draft the manuscript.

Institutional review board statement: This study was approved by the Ethics Committee of Yamanashi University (approved number: 1825) and was performed in accordance with the ethical standards of the Declaration of Helsinki and its later amendments.

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

Manuscript source: Unsolicited manuscript

Correspondence to: Daisuke Ichikawa, MD, PhD, Professor, First Department of Surgery, Faculty of Medicine University of Yamanashi, 1110 Shimokato, Chuou, Yamanashi 409-3898, Japan. dichikawa@yamanashi.ac.jp
Telephone: +81-55-2737390
Fax: +81-55-2737390

Received: July 4, 2018
Peer-review started: July 5, 2018
First decision: July 24, 2018
Revised: August 5, 2018
Accepted: August 30, 2018
Article in press: August 31, 2018
Published online: October 15, 2018

Abstract

AIM

To investigate the clinical utility of alpha-fetoprotein (AFP)-producing gastric cancer (AFPGC)-specific microRNA (miRNA) for monitoring and prognostic prediction of patients.

METHODS

We performed a comprehensive miRNA array-based approach to compare miRNA expression levels between AFP-positive and AFP-negative cells in three patients with primary AFPGC. We next examined the expression levels of the selected miRNAs in five AFPGC and ten non-AFPGC tissue samples by quantitative reverse transcription-polymerase chain reaction to validate their utility. We also investigated the expression levels of the selected miRNA not only in tissue but also in plasma

samples. Moreover, we investigated the relationship between plasma AFP levels and plasma selected miRNA expression levels, and also investigated the correlation of the selected miRNA expression levels and malignant potential.

RESULTS

Among the five miRNAs selected from the miRNA array results, the expression levels of *miR-122-5p* were significantly higher in the AFPGC patients than in the non-AFPGC patients ($P < 0.05$). In tissue samples, *miR-122-5p* expression level tended to be lower in the non-AFPGC tissue than the normal gastric mucosa. Conversely, in the AFPGC tissue, *miR-122-5p* expression level was significantly higher in the AFPGC tissue than both the normal gastric mucosa and the non-AFPGC tissue samples ($P < 0.05$). Plasma *miR-122-5p* expression levels were also significantly higher in the AFPGC patients than the health volunteers and the non-AFPGC patients ($P < 0.05$) and were strongly correlated with plasma AFP levels ($r = 0.7975$, $P < 0.0001$). Moreover, the correlation of *miR-122-5p* expression in tissue samples with malignant potential was stronger than that of plasma AFP level in the AFPGC patients. In contrast, no correlation was found between *miR-122-5p* expression levels and liver metastasis in the non-AFPGC patients.

CONCLUSION

miR-122-5p might be a useful biomarker for early detection and disease monitoring in AFPGC.

Key words: Gastric cancer; Alpha-fetoprotein; Alpha-fetoprotein producing gastric cancer; MicroRNA; *miR-122-5p*

Core tip: We examined the microRNAs (miRNA) expression in alpha-fetoprotein (AFP)-producing gastric cancer (AFPGC) tissue samples using a comprehensive miRNA array-based approach, and also investigated the clinical utility of the identified AFPGC-specific miRNAs. We found the expression of *miR-122-5p* was significantly higher in the AFPGC tissues and plasma samples. Moreover, tissue *miR-122-5p* expression levels exhibited a stronger correlation with malignant potential than plasma AFP level in AFPGC patients. *miR-122-5p* might be a useful biomarker for early detection and disease monitoring in AFPGC.

Maruyama S, Furuya S, Shiraishi K, Shimizu H, Akaike H, Hosomura N, Kawaguchi Y, Amemiya H, Kawaida H, Sudo M, Inoue S, Kono H, Ichikawa D. *miR-122-5p* as a novel biomarker for alpha-fetoprotein-producing gastric cancer. *World J Gastrointest Oncol* 2018; 10(10): 344-350 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/344.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.344

INTRODUCTION

Gastric cancer (GC) is one of the most common solid tumors and is the third leading cause of cancer-related deaths worldwide[1,2]. Despite improvements in treatment approaches, prognosis of patients with advanced GC remains poor even after curative resection.

Among various tumor subtypes, alpha-fetoprotein (AFP)-producing GC (AFPGC) is recognized as one of the most aggressive tumors, with a high propensity for liver metastasis and subsequent poor prognosis compared with other GC subtypes[3-7]. The incidence of AFPGC is low, ranging from 1.3% to 15% of all GCs[8-12]. Therefore, recent comprehensive molecular analyses have not yet referred to this minor subtype.

MicroRNAs (miRNAs) are endogenous, small, non-coding, single-stranded RNAs of 20-25 nucleotides that regulate the expression of target genes at post-transcriptional level by binding to complementary sequences[13]. Various miRNAs were shown to play crucial roles in cancer as well as normal cells were reported to act as tumor suppressors or oncogenes in a cell type-dependent manner in various cancers[14]. In addition, certain miRNAs have been used for cancer detection, monitoring of tumor dynamics, and predicting prognosis and chemoresistance[15-20].

In the present study, we examined the miRNAs expression in AFPGC tissue samples using a comprehensive miRNA array-based approach. We also investigated the clinical utility of the identified AFPGC-specific miRNAs in monitoring and prognostic prediction of patients with AFPGC and evaluated their potential as universal biomarker for liver metastases.

MATERIALS AND METHODS

Patients and samples

A total of 492 patients underwent gastrectomy for GC at the University of Yamanashi Hospital between 2012 and 2018. Tumor specimens and resected lymph nodes obtained at the time of surgery were immediately fixed in 10% neutral-buffered formalin and embedded in paraffin after fixation. None of the patients underwent preoperative chemotherapy or radiotherapy. Tissue samples of all five patients with primary AFPGC and those from ten patients with primary non-AFPGC at various stages as controls from the same cohort were selected. The selected AFPGC samples contained all AFPGC patients who were operated in our hospital.

Pre-operative plasma samples were also obtained from four AFPGC patients and twenty non-AFPGC patients with GC who underwent surgical resection at the University of Yamanashi Hospital between 2017 and 2018. Control plasma samples were collected from 12 healthy adult volunteers. A total of 5 mL blood samples were collected into ethylenediaminetetraacetic acid-coated tubes and immediately spun at 3000 rpm at 4℃ for 10 min to separate serum, which was stored at

-80℃ for further processing. AFPGC was defined based on a plasma AFP level above 10 ng/mL or positive AFP immunoreactivity in tissue samples. This study was approved by the Ethics Committee of Yamanashi University and performed in accordance with the ethical standards of the Declaration of Helsinki and its amendments.

RNA extraction

Formalin-fixed, paraffin-embedded tissue samples were cut into 10-μm-thick sections, and total RNA was extracted from tumor and normal gastric mucosa in each patient using RNeasy FFPE kit (Qiagen, Valencia, CA), according to the manufacturer's protocol. In plasma samples, total RNA was extracted from 100 μL plasma using RNeasy Serum/Plasma kit (Qiagen), according to the manufacturer's protocol.

miRNA microarray analysis

Microarray analyses of the GC tissue samples were performed using 3D-Gene miRNA oligo chips (Toray Industries, Kamakura, Japan), with 2565 genes mounted onto each DNA chip. Results were compared between the AFP-positive and AFP-negative cells among AFPGC patient samples using macro-dissection. Tissue samples from the three AFPGC patients who underwent curative surgery were mixed equally. RNAs were labeled with the 3D-Gene miRNA labeling kit (Toray Industries). Fluorescent signals were scanned using a 3D-Gene scanner 3000 (Toray Industries) and analyzed with the 3D-Gene Extraction software (Toray Industries). In the current study, expression level of each miRNA was normalized using the median signal intensity of the all genes in each chip, and median signal intensity was adjusted to 25.

Quantification of miRNA by quantitative reverse transcription-polymerase chain reaction

Levels of miRNAs were quantified by quantitative reverse transcription-polymerase chain reaction (qRT-PCR) using a Human TaqMan MicroRNA Assay kit (Applied Biosystems, Foster City, CA), according to standard procedures. Reverse transcription was conducted with a TaqMan MicroRNA Reverse Transcription kit (Applied Biosystems). Tissue miRNA levels were normalized to the endogenous control *RNU6B*, and plasma miRNA levels were normalized to a synthetic RNA oligonucleotide, cel-miR-39-3p (Qiagen), by spiking the samples with the oligonucleotide which does not exist in human genome. The following primers were used for the Taqman assay (Thermo Fisher Scientific, CA, United States): human *hsa-miR-122-5p* (cat #002245), *hsa-miR-144-5p* (cat #002148), *hsa-miR-20a-5p* (cat #000580), *hsa-miR-20b-5p* (cat #001014), *hsa-miR-106a-5p* (cat #000578), *RNU6B* (cat #001093), and cel-miR-39-3p (cat #000200). ΔCt values for all miRNAs relative to the control gene RNU6B and cel-miR-39-3p were determined. ΔΔCt values were calculated using mean ΔCt values in non-AFPGC tissue, normal gastric mucosa, or healthy volunteer plasma samples. Plasma *miR-122-5p* expression was calculated using $\log10(2^{-\Delta Ct})$.

Statistical analysis

Statistical significance was determined using GraphPad Prism® version 5 (San Diego, CA). Quantitative values were expressed as means ± SD unless noted otherwise. Statistical significance was evaluated using the Student's *t* test and one-way analysis of variance for each time point, followed by Tukey's *post hoc* test. Pearson's correlation coefficient was determined to assess the correlation between plasma AFP and plasma *miR-122-5p* levels. P values < 0.05 were considered to indicate statistical significance.

RESULTS

Identification of miRNA candidates from a comprehensive miRNA array-based approach in AFPGC tissue

We selected miRNA candidates using a miRNA array-based approach. We compared the expression levels of each miRNA between the AFP-positive and AFP-negative cells in AFPGC patients. Of the 2565 candidates analyzed, we selected the following five miRNAs: *miR-122-5p*, *miR-20a-5p*, *miR-20b-5p*, *miR-106a-5p*, and *miR-144-5p*. The expression levels of these selected miRNAs were significantly different in AFP-positive cells compared with the AFP-negative cells, and the signal intensity of each miRNA was sufficient (Table 1).

Validation of the expression levels of five miRNAs in AFPGC and non-AFPGC tissue samples

We examined the expression levels of the five selected miRNAs in five AFPGC and ten non-AFPGC tissue samples by qRT-PCR to validate their utility (Figure 1). Among these five miRNAs, the expression of *miR-122-5p* was significantly higher in the AFPGC patients than the non-AFPGC patients. Therefore, we selected *miR-122-5p* for further analyses in this study.

miR-122-5p expression levels in tissue and plasma samples

Next, we investigated the expression levels of *miR-122-5p* not only in tissue but also in plasma samples. In tissue samples, *miR-122-5p* expression levels tended to be lower in the non-AFPGC tissue samples than in the normal gastric mucosa samples. Conversely, *miR-122-5p* expression levels were significantly higher in the AFPGC tissue samples compared with the normal gastric mucosa and the non-AFPGC tissue samples (Figure 2A). The plasma expression levels of *miR-122-5p* were also significantly higher in the AFPGC patient samples than the samples from health volunteers and the non-AFPGC patients (Figure 2B).

Table 1 Summary of five miRNA candidates selected by microarray analysis

Gene ID	Signal intensity		Fold change
	AFPGC	Non-AFPGC	AFPGC/non-AFPGC
hsa-miR-122-5p	492	105	4.7
hsa-miR-20a-5p	245	113	2.2
hsa-miR-20b-5p	198	94	2.1
hsa-miR-106a-5p	304	195	1.6
hsa-miR-145-5p	712	1367	0.5

Expression level of each miRNA was normalized using the median signal intensity of the all genes in each chip, and median signal intensity was adjusted to 25. AFPGC: Alpha-fetoprotein-producing gastric cancer.

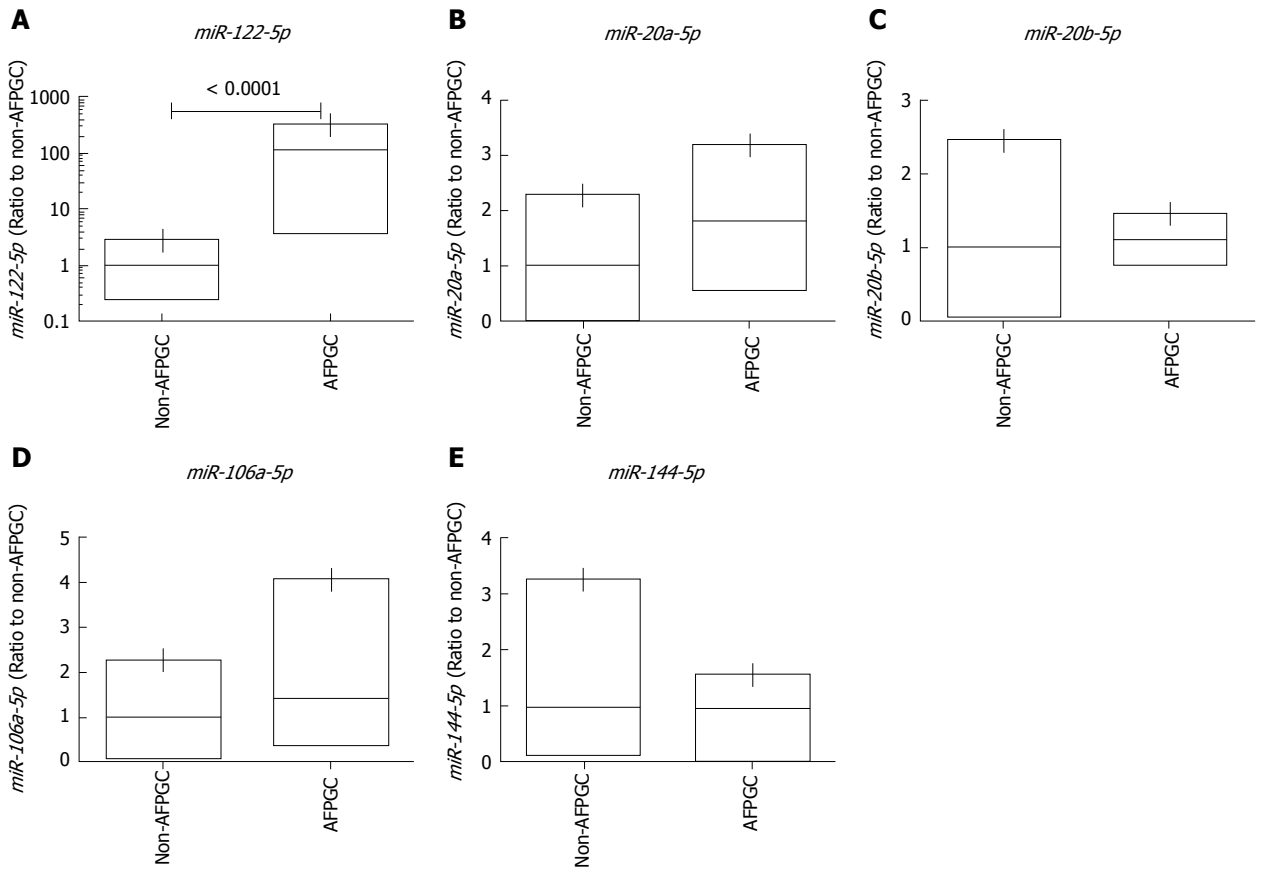


Figure 1 Validation of five microRNAs in non-alpha-fetoprotein producing gastric cancer and alpha-fetoprotein producing gastric cancer tissue samples performed by quantitative reverse transcription-polymerase chain reaction. A: *miR-122-5p*; B: *miR-20a-5p*; C: *miR-20b-5p*; D: *miR-106a-5p*; E: *miR-144-5p*. The lines inside the bow plot represent the average size. AFPGC: Alpha-fetoprotein-producing gastric cancer.

Plasma miR-122-5 levels are strongly correlated with plasma AFP levels in GC patients

We next investigated the relationship between plasma AFP levels and plasma *miR-122-5p* expression levels in the AFPGC and non-AFPGC patients and found that *miR-122-5p* expression level in plasma was strongly correlated with plasma AFP level ($r = 0.7975$, $P < 0.0001$; Figure 3).

Prognostic utility of tissue miR-122-5p expression in AFPGC patients

Figure 4 shows the correlation between malignant potential, all biomarkers, tissue *miR-122-5p* expression, and plasma AFP level in the AFPGC patients. We found that the expression level of *miR-122-5p* in tissue exhibited a stronger correlation with malignant potential (*i.e.*, liver metastasis) than plasma AFP level in the AFPGC patients. Two patients with malignant potential were diagnosed morphologically as poorly differentiated adenocarcinoma and mucinous adenocarcinoma, and the other current alive patients were diagnosed as poorly differentiated adenocarcinoma and hepatoid adenocarcinoma.

DISCUSSION

AFPGC has been reported to be more likely to metastasize to liver and is therefore associated with extremely

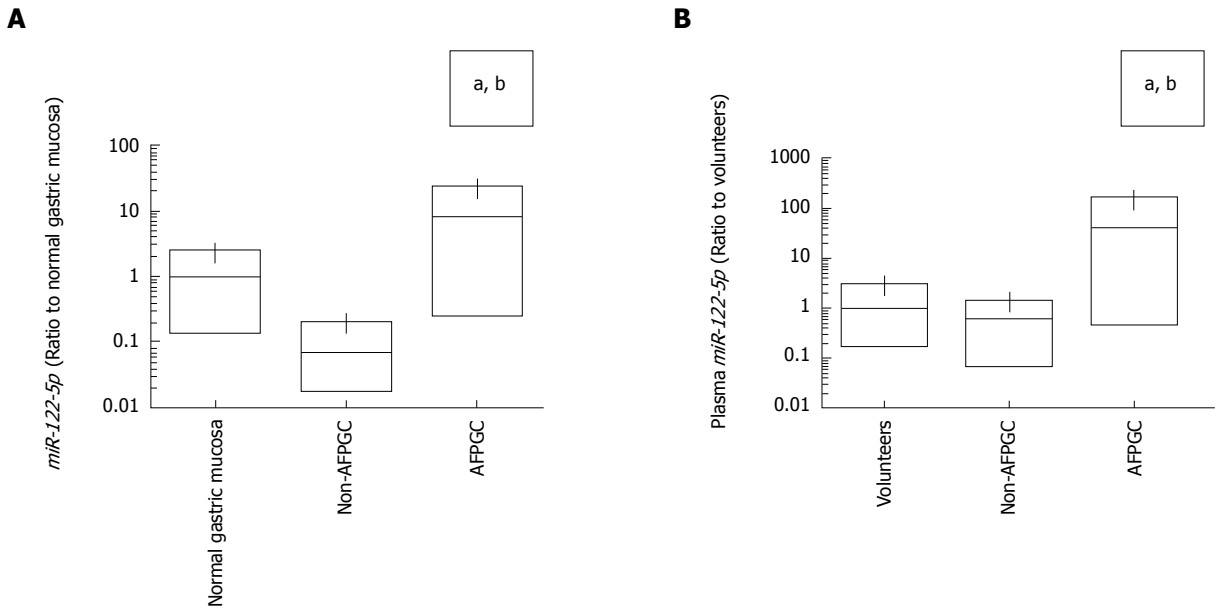


Figure 2 Quantification of *miR-122-5p* expression levels by quantitative reverse transcription-polymerase chain reaction. A: Comparison of *miR-122-5p* expression levels between normal gastric mucosa, non-alpha-fetoprotein-producing gastric cancer (AFPGC) and AFPGC in tissue samples. [a]$P < 0.05$, compared to normal gastric mucosa; [b]$P < 0.05$, compared to non-AFPGC; B: Comparison of *miR-122-5p* expression levels between health volunteers, non-AFPGC and AFPGC in plasma sample. [a]$P < 0.05$, compared to health volunteers; [b]$P < 0.05$, compared to non-AFPGC. The lines inside the bow plot represent the average size.

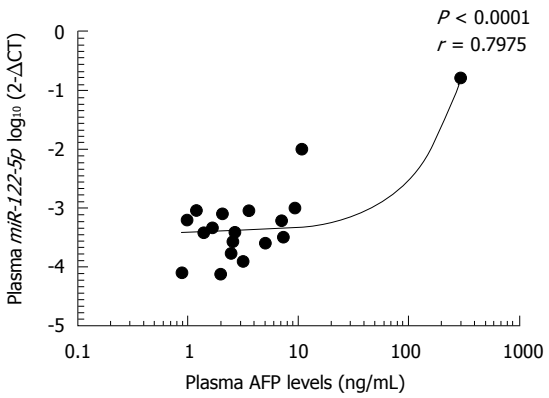


Figure 3 Relationship between plasma alpha-fetoprotein levels and plasma *miR-122-5p* expression levels. Plasma *miR-122-5p* expression level was strongly correlated with plasma alpha-fetoprotein (AFP) levels in gastric cancer patients ($r = 0.7975$, $P < 0.0001$).

poor prognosis[8,11]. However, no genomic analyses have been conducted for AFPGC due to its rarity. Therefore, AFPGC-specific genomic and/or epigenomic alterations are not well known, which have urged us to examine the molecular characteristics specific to AFPGC. The current study investigated the molecular characteristics of AFPGC with a comprehensive analysis, with particular focus on miRNA expression.

The findings of the present study clearly demonstrated that the expression of *miR-122-5p* was significantly higher in the AFPGC tissues than the normal and non-AFPGC tissues. The expression levels of this miRNA were also higher in the plasma samples of patients with AFPGC compared with those of healthy volunteers and non-AFPGC patients and correlated with plasma AFP levels to a certain extent. Interestingly, the tissue expression level of *miR-122-5p* exhibited a stronger correlation with malignant potential than plasma AFP level in AFPGC patients, suggesting that *miR-122-5p* might have utility as a prognostic biomarker especially for liver metastasis in this small GC subgroup.

miR-122-5p expression has been increasingly examined in various normal and cancer tissue types. Several studies reported that *miR-122-5p* was specifically expressed in human liver and that hepatocyte-specific *miR-122-5p* regulated hepatocyte differentiation and metabolism[21-23]. Taken together, AFPGC might show characteristics of hepatocytes not only morphologically but also in its miRNA expression patterns. In fact, AFPGC was not necessarily hepatoid adenocarcinoma in this series, and two patients with aggressive development of liver metastasis were diagnosed morphologically as poorly differentiated adenocarcinoma and mucinous adenocarcinoma. Conversely, *miR-122-5p* was previously shown to function as a tumor suppressor and was reported to be downregulated in several cancer types such as hepatocellular carcinoma[24], non-small-cell lung cancer[25], gallbladder carcinoma[26], bladder cancer[27], and breast cancer[28]. In GC, the expression of *miR-122-5p* was reported to be lower in tumor tissue than the adjacent non-cancerous tissue. Furthermore, several studies reported that *miR-122-5p* inhibited proliferation, migration, and invasion in GC[15,29,30]. It's not known exactly why *miR-122-5p*, which is known as suppressor gene, is higher in AFPGC. Some miRNA was reported that decreased in early cases and elevated again in staged-advanced cases[31]. Therefore, *miR-122-5p* decreased in carcinogenesis might be elevated during tumor evolution

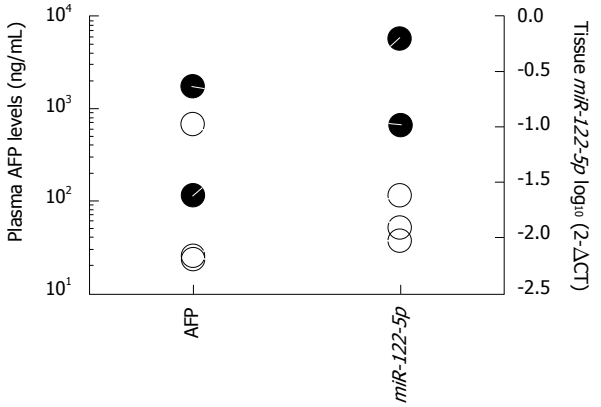


Figure 4 Correlation between malignant potential and tissue *miR-122-5p* expression levels in alpha-fetoprotein-producing gastric cancer patients. White symbols indicate current alive and black symbols indicate current death. AFP: Alpha-fetoprotein.

to AFPGC. However, the exact mechanism is unknown at the present time.

In the current study, we did not see a correlation between *miR-122-5p* in patients with non-AFPGC and development of liver metastasis, suggesting that the mechanism underlying liver metastasis might be distinct between AFPGC and non-AFPGC. We assume that AFPGC is completely different from non-AFPGC, and the mechanism of liver metastasis between AFPGC and non-AFPGC is also distinct.

Several reports demonstrated that the clinical behavior of AFPGC was distinct from that of non-AFPGC[32]. Recently, Lu *et al*[33] demonstrated that AFP contributed to invasion and metastasis directly. We speculate AFPGC has specific ability of liver metastasis, and correlated with *miR-122-5p*. Therefore, *miR-122-5p* might directly facilitate tumor proliferation, migration, and invasion, which raises the possibility of *miR-122-5p* as a potential therapeutic target in AFPGC. However, future studies are warranted to demonstrate the biological function underlying altered expression of *miR-122-5p* in AFPGC. The current study revealed *miR-122-5p* as a potentially useful biomarker for early detection, disease monitoring, and prognostic prediction in patients with AFPGC, which warrant further investigation.

ARTICLE HIGHLIGHTS

Research background

Alpha-fetoprotein (AFP)-producing gastric cancer (AFPGC) is recognized as one of the most aggressive tumors, with a high propensity for liver metastasis and subsequent poor prognosis compared with other GC subtypes. Recent comprehensive molecular analyses have not yet referred to this minor subtype because of its rareness.

Research motivation

To discover universal biomarkers for liver metastasis by researching AFPGC-specific microRNAs (miRNAs).

Research objectives

To investigate the clinical utility of AFPGC-specific miRNA for monitoring and prognostic prediction of patients.

Research methods

We performed a comprehensive miRNA array-based approach to compare miRNA expression levels between AFP-positive and AFP-negative cells, and also investigated the clinical utility of the identified AFPGC-specific miRNAs.

Research results

We found the expression of *miR-122-5p* was significantly higher in the AFPGC tissues than the normal and non-AFPGC tissues. The expression levels of this miRNA were also higher in the plasma samples of patients with AFPGC compared with those of healthy volunteers and non-AFPGC patients and correlated with plasma AFP levels. Moreover, the tissue expression level of *miR-122-5p* exhibited a stronger correlation with malignant potential than plasma AFP level in AFPGC patients.

Research conclusions

miR-122-5p as a potentially useful biomarker for early detection and disease monitoring in patients with AFPGC.

Research perspectives

We identified *miR-122-5p* as AFPGC-specific miRNA. *miR-122-5p* might be a clinical useful biomarker in AFPGC. Although studies are warranted to demonstrate the biological function underlying altered expression of *miR-122-5p* in AFPGC, the *miR-122-5p* might be a potential therapeutic target for liver metastasis in AFPGC.

ACKNOWLEGEMENTS

The Authors are grateful to Motoko Inui and Makiko Mishina for expert technical assistance.

REFERENCES

1 **Kamangar F**, Dores GM, Anderson WF. Patterns of cancer incidence, mortality, and prevalence across five continents: defining priorities to reduce cancer disparities in different geographic regions of the world. *J Clin Oncol* 2006; **24**: 2137-2150 [PMID: 16682732 DOI: 10.1200/jco.2005.05.2308]

2 **Cancer Genome Atlas Research Network**. Comprehensive molecular characterization of gastric adenocarcinoma. *Nature* 2014; **513**: 202-209 [PMID: 25079317 DOI: 10.1038/nature13480]

3 **Bourreille J**, Metayer P, Sauger F, Matray F, Fondimare A. [Existence of alpha feto protein during gastric-origin secondary cancer of the liver]. *Presse Med* 1970; **78**: 1277-1278 [PMID: 5426134]

4 **Chang YC**, Nagasue N, Abe S, Kohno H, Yamanoi A, Uchida M, Nakamura T. [The characters of AFP-producing early gastric cancer]. *Nihon Geka Gakkai Zasshi* 1990; **91**: 1574-1580 [PMID: 1702182]

5 **Motoyama T**, Aizawa K, Watanabe H, Fukase M, Saito K. alpha-Fetoprotein producing gastric carcinomas: a comparative study of three different subtypes. *Acta Pathol Jpn* 1993; **43**: 654-661 [PMID: 7508672 DOI: 10.1111/j.1440-1827.1993.tb02549.x]

6 **Chang YC**, Nagasue N, Abe S, Taniura H, Kumar DD, Nakamura T. Comparison between the clinicopathologic features of AFP-positive and AFP-negative gastric cancers. *Am J Gastroenterol* 1992; **87**: 321-325 [PMID: 1371637]

7 **Koide N**, Nishio A, Igarashi J, Kajikawa S, Adachi W, Amano J. Alpha-fetoprotein-producing gastric cancer: histochemical analysis of cell proliferation, apoptosis, and angiogenesis. *Am J Gastroenterol* 1999; **94**: 1658-1663 [PMID: 10364040 DOI: 10.1111/j.1572-0241.1999.01158.x]

8 **Kono K**, Amemiya H, Sekikawa T, Iizuka H, Takahashi A, Fujii H, Matsumoto Y. Clinicopathologic features of gastric cancers producing alpha-fetoprotein. *Dig Surg* 2002; **19**: 359-365; discussion 365 [PMID: 12435906 DOI: 10.1159/000065838]

9 **Chang YC**, Nagasue N, Kohno H, Taniura H, Uchida M, Yamanoi A, Kimoto T, Nakamura T. Clinicopathologic features and long-

term results of alpha-fetoprotein-producing gastric cancer. *Am J Gastroenterol* 1990; **85**: 1480-1485 [PMID: 1700600]
10 **Chun H**, Kwon SJ. Clinicopathological characteristics of alpha-fetoprotein-producing gastric cancer. *J Gastric Cancer* 2011; **11**: 23-30 [PMID: 22076198 DOI: 10.5230/jgc.2011.11.1.23]
11 **Liu X**, Cheng Y, Sheng W, Lu H, Xu Y, Long Z, Zhu H, Wang Y. Clinicopathologic features and prognostic factors in alpha-fetoprotein-producing gastric cancers: analysis of 104 cases. *J Surg Oncol* 2010; **102**: 249-255 [PMID: 20740583 DOI: 10.1002/jso.21624]
12 **McIntire KR**, Waldmann TA, Moertel CG, Go VL. Serum alpha-fetoprotein in patients with neoplasms of the gastrointestinal tract. *Cancer Res* 1975; **35**: 991-996 [PMID: 46783]
13 **Ha M**, Kim VN. Regulation of microRNA biogenesis. *Nat Rev Mol Cell Biol* 2014; **15**: 509-524 [PMID: 25027649 DOI: 10.1038/nrm3838]
14 **Esquela-Kerscher A**, Slack FJ. Oncomirs - microRNAs with a role in cancer. *Nat Rev Cancer* 2006; **6**: 259-269 [PMID: 16557279 DOI: 10.1038/nrc1840]
15 **Chen Q**, Ge X, Zhang Y, Xia H, Yuan D, Tang Q, Chen L, Pang X, Leng W, Bi F. Plasma miR-122 and miR-192 as potential novel biomarkers for the early detection of distant metastasis of gastric cancer. *Oncol Rep* 2014; **31**: 1863-1870 [PMID: 24481716 DOI: 10.3892/or.2014.3004]
16 **Hiramoto H**, Muramatsu T, Ichikawa D, Tanimoto K, Yasukawa S, Otsuji E, Inazawa J. miR-509-5p and miR-1243 increase the sensitivity to gemcitabine by inhibiting epithelial-mesenchymal transition in pancreatic cancer. *Sci Rep* 2017; **7**: 4002 [PMID: 28638102 DOI: 10.1038/s41598-017-04191-w]
17 **Hiyoshi Y**, Akiyoshi T, Inoue R, Murofushi K, Yamamoto N, Fukunaga Y, Ueno M, Baba H, Mori S, Yamaguchi T. Serum miR-143 levels predict the pathological response to neoadjuvant chemoradiotherapy in patients with locally advanced rectal cancer. *Oncotarget* 2017; **8**: 79201-79211 [PMID: 29108299 DOI: 10.18632/oncotarget.16760]
18 **Imamura T**, Komatsu S, Ichikawa D, Miyamae M, Okajima W, Ohashi T, Kiuchi J, Nishibeppu K, Konishi H, Shiozaki A, Morimura R, Ikoma H, Ochiai T, Okamoto K, Taniguchi H, Otsuji E. Depleted tumor suppressor miR-107 in plasma relates to tumor progression and is a novel therapeutic target in pancreatic cancer. *Sci Rep* 2017; **7**: 5708 [PMID: 28720759 DOI: 10.1038/s41598-017-06137-8]
19 **Matsushita R**, Seki N, Chiyomaru T, Inoguchi S, Ishihara T, Goto Y, Nishikawa R, Mataki H, Tatarano S, Itesako T, Nakagawa M, Enokida H. Tumour-suppressive microRNA-144-5p directly targets CCNE1/2 as potential prognostic markers in bladder cancer. *Br J Cancer* 2015; **113**: 282-289 [PMID: 26057453 DOI: 10.1038/bjc.2015.195]
20 **Yang R**, Fu Y, Zeng Y, Xiang M, Yin Y, Li L, Xu H, Zhong J, Zeng X. Serum miR-20a is a promising biomarker for gastric cancer. *Biomed Rep* 2017; **6**: 429-434 [PMID: 28413641 DOI: 10.3892/br.2017.862]
21 **Roderburg C**, Benz F, Vargas Cardenas D, Koch A, Janssen J, Vucur M, Gautheron J, Schneider AT, Koppe C, Kreggenwinkel K, Zimmermann HW, Luedde M, Trautwein C, Tacke F, Luedde T. Elevated miR-122 serum levels are an independent marker of liver injury in inflammatory diseases. *Liver Int* 2015; **35**: 1172-1184 [PMID: 25039534 DOI: 10.1111/liv.12627]
22 **Koyama S**, Kuragaichi T, Sato Y, Kuwabara Y, Usami S, Horie T, Baba O, Hakuno D, Nakashima Y, Nishino T, Nishiga M, Nakao T, Arai H, Kimura T, Ono K. Dynamic changes of serum microRNA-122-5p through therapeutic courses indicates amelioration of acute liver injury accompanied by acute cardiac decompensation. *ESC Heart Fail* 2017; **4**: 112-121 [PMID: 28451447 DOI: 10.1002/ehf2.12123]
23 **Satishchandran A**, Ambade A, Rao S, Hsueh YC, Iracheta-Vellve A, Tornai D, Lowe P, Gyongyosi B, Li J, Catalano D, Zhong L, Kodys K, Xie J, Bala S, Gao G, Szabo G. MicroRNA 122, Regulated by GRLH2, Protects Livers of Mice and Patients From Ethanol-Induced Liver Disease. *Gastroenterology* 2018; **154**: 238-252.e7 [PMID: 28987423 DOI: 10.1053/j.gastro.2017.09.022]
24 **Wang N**, Wang Q, Shen D, Sun X, Cao X, Wu D. Downregulation of microRNA-122 promotes proliferation, migration, and invasion of human hepatocellular carcinoma cells by activating epithelial-mesenchymal transition. *Onco Targets Ther* 2016; **9**: 2035-2047 [PMID: 27103830 DOI: 10.2147/OTT.S92378]
25 **Qin H**, Sha J, Jiang C, Gao X, Qu L, Yan H, Xu T, Jiang Q, Gao H. miR-122 inhibits metastasis and epithelial-mesenchymal transition of non-small-cell lung cancer cells. *Onco Targets Ther* 2015; **8**: 3175-3184 [PMID: 26604787 DOI: 10.2147/OTT.S91696]
26 **Lu W**, Zhang Y, Zhou L, Wang X, Mu J, Jiang L, Hu Y, Dong P, Liu Y. miR-122 inhibits cancer cell malignancy by targeting PKM2 in gallbladder carcinoma. *Tumour Biol* 2015 [PMID: 26546436 DOI: 10.1007/s13277-015-4308-z]
27 **Wang Y**, Xing QF, Liu XQ, Guo ZJ, Li CY, Sun G. MiR-122 targets VEGFC in bladder cancer to inhibit tumor growth and angiogenesis. *Am J Transl Res* 2016; **8**: 3056-3066 [PMID: 27508026]
28 **Ergün S**, Ulasli M, Igci YZ, Igci M, Kırkbes S, Borazan E, Balik A, Yumrutaş Ö, Camci C, Cakmak EA, Arslan A, Oztuzcu S. The association of the expression of miR-122-5p and its target ADAM10 with human breast cancer. *Mol Biol Rep* 2015; **42**: 497-505 [PMID: 25318895 DOI: 10.1007/s11033-014-3793-2]
29 **Rao M**, Zhu Y, Zhou Y, Cong X, Feng L. MicroRNA-122 inhibits proliferation and invasion in gastric cancer by targeting CREB1. *Am J Cancer Res* 2017; **7**: 323-333 [PMID: 28337380]
30 **Xu X**, Gao F, Wang J, Tao L, Ye J, Ding L, Ji W, Chen X. MiR-122-5p inhibits cell migration and invasion in gastric cancer by down-regulating DUSP4. *Cancer Biol Ther* 2018; **19**: 427-435 [PMID: 29509059 DOI: 10.1080/15384047.2018.1423925]
31 **Cheng H**, Zhang L, Cogdell DE, Zheng H, Schetter AJ, Nykter M, Harris CC, Chen K, Hamilton SR, Zhang W. Circulating plasma MiR-141 is a novel biomarker for metastatic colon cancer and predicts poor prognosis. *PLoS One* 2011; **6**: e17745 [PMID: 21445232 DOI: 10.1371/journal.pone.0017745]
32 **Hirajima S**, Komatsu S, Ichikawa D, Kubota T, Okamoto K, Shiozaki A, Fujiwara H, Konishi H, Ikoma H, Otsuji E. Liver metastasis is the only independent prognostic factor in AFP-producing gastric cancer. *World J Gastroenterol* 2013; **19**: 6055-6061 [PMID: 24106406 DOI: 10.3748/wjg.v19.i36.6055]
33 **Lu S**, Ma Y, Sun T, Ren R, Zhang X, Ma W. Expression of α-fetoprotein in gastric cancer AGS cells contributes to invasion and metastasis by influencing anoikis sensitivity. *Oncol Rep* 2016; **35**: 2984-2990 [PMID: 26986949 DOI: 10.3892/or.2016.4678]

P- Reviewer: Jung Y, Takemura N **S- Editor**: Ji FF
L- Editor: A **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 351-359

DOI: 10.4251/wjgo.v10.i10.351

ISSN 1948-5204 (online)

ORIGINAL ARTICLE

Retrospective Study

Prognostic value of vascular endothelial growth factor receptor 1 and class III β-tubulin in survival for non-metastatic rectal cancer

Xiang-Quan Kong, Yun-Xia Huang, Jin-Luan Li, Xue-Qing Zhang, Qing-Qin Peng, Li-Rui Tang, Jun-Xin Wu

Xiang-Quan Kong, Yun-Xia Huang, Jin-Luan Li, Xue-Qing Zhang, Li-Rui Tang, Jun-Xin Wu, Department of Radiation Oncology, Fujian Medical University Cancer Hospital, Fujian Cancer Hospital, Fuzhou 350014, Fujian Province, China

Qing-Qin Peng, Department of Radiation Oncology, First Hospital of Quanzhou Affiliated to Fujian Medical University, Quanzhou 362000, Fujian Province, China

ORCID number: Xiang-Quan Kong (0000-0002-5215-121X); Yun-Xia Huang (0000-0002-8542-9804); Jin-Luan Li (0000-0002-3533-898X); Xue-Qing Zhang (0000-0002-0213-8041); Qing-Qin Peng (0000-0002-8787-301X); Li-rui Tang (0000-0003-3721-5913); Jun-Xin Wu (0000-0003-1047-2338).

Author contributions: All authors helped to perform the research; Kong XQ wrote the manuscript and performed procedures; Huang YX wrote the manuscript and performed data analysis; Li JL contributed to writing the manuscript and drafting conception; Zhang XQ, Peng QQ and Tang LR contributed to writing the manuscript and data analysis; Wu JX contributed to writing the manuscript, drafting conception and design.

Supported by Fujian Province Natural Science Foundation, Nos. 2016J01437, 2017J01260 and 2018J01266; the Fujian Medical Innovation Project, No. 2015-CX-8; the Peking University Cancer Hospital and Institute, Key Laboratory of Carcinogenesis and Translational Research, Ministry of Education/Beijing (2017 Open Project-9), Joint Funds for the innovation of science and Technology, Fujian Province, No. 2017Y9074.

Institutional review board statement: This study was reviewed and approved by the Ethics Committee of the Fujian Cancer Hospital.

Informed consent statement: Patients were not required to give informed consent to the study due to the retrospective nature of the study involving the review of patient medical records and tumor specimens.

Conflict-of-interest statement: All authors declare no conflicts-of-interest related to this article.

Manuscript source: Unsolicited manuscript

Correspondence to: Jun-Xin Wu, PhD, Attending Doctor, Professor, Department of Radiation Oncology, Fujian Medical University Cancer Hospital, Fujian Cancer Hospital, 420 Fuma Rd, Jinan District, Fuzhou 350014, Fujian Province, China. junxinwufj@aliyun.com
Telephone: +86-591-83660063

Received: June 2, 2018
Peer-review started: June 2, 2018
First decision: July 10, 2018
Revised: July 17, 2018
Accepted: August 26, 2018
Article in press: August 26, 2018
Published online: October 15, 2018

Abstract

AIM

To assess the long-term prognostic value of vascular endothelial growth factor receptor 1 (VEGFR1) and class Ⅲ β-tubulin (TUBB3) mRNA expression in non-metastatic rectal cancer.

METHODS

A total of 75 consecutive patients with non-metastatic rectal cancer from March 2004 to November 2008 were analyzed retrospectively at our institute. The mRNA expressions of VEGFR1 and TUBB3 were detected by multiplex branched DNA liquid-chip technology. The Cut-off Finder application was applied to determine cutoff point of mRNA expression. SPSS software version 22.0 was used for analysis.

RESULTS

The median follow-up was 102.7 mo (range, 6-153.6). The χ^2 and Fisher's exact tests showed that VEGFR1 expression was related to lymph node metastasis (P = 0.013), while no relationships between TUBB3 and clinicopathological features were observed. Univariate analysis showed that T stage, lymph node metastasis, tumor differentiation, VEGFR1 and TUBB3 mRNA expression were correlated to overall survival (OS) (P = 0.048, P = 0.003, P = 0.052, P = 0.003 and P = 0.015, respectively). Also, lymph node metastasis and VEGFR1 expression independently influenced OS by multivariate analysis (P = 0.027 and P = 0.033). VEGFR1 expression was positively correlated with TUBB3 (P = 0.024). The patients with low expression of both TUBB3 and VEGFR1 presented a better OS (P = 0.003). In addition, the receiver operating characteristic analysis suggested that the combination of lymph node metastasis and VEGFR1 had a more favorable prognostic value (P < 0.001).

CONCLUSION

VEGFR1 expression and lymph node metastasis independently and jointly affect survival. Moreover, low expression of VEGFR1 and TUBB3 presented a better OS in patients with non-metastatic rectal cancer, which might serve as a potential prognostic factor.

Key words: Rectal cancer; Class Ⅲ β-tubulin; Vascular endothelial growth factor receptor 1; Overall survival

Core tip: Nowadays, personalized and precision medicine becomes vital in cancer treatment. Herein, we focus on the long-term prognostic value of vascular endothelial growth factor receptor 1 (VEGFR1) and class Ⅲ β-tubulin (TUBB3) mRNA expression in non-metastatic rectal cancer. In the 75 consecutive patients enrolled, we found that VEGFR1 expression and lymph node metastasis were independent factors influencing overall survival, and the combination of them showed a favorable prognostic value. Also, VEGFR1 expression was significantly related to lymph node metastasis. In addition, VEGFR1 expression was positively correlated with TUBB3 expression.

Kong XQ, Huang YX, Li JL, Zhang XQ, Peng QQ, Tang LR, Wu JX. Prognostic value of vascular endothelial growth factor receptor 1 and class III β-tubulin in survival for non-metastatic rectal cancer. *World J Gastrointest Oncol* 2018; 10(10): 351-359 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/351.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.351

INTRODUCTION

Rectal cancer is one of the most diagnosed malignancies among both males and females worldwide with worse outcomes than colon cancer[1,2]. Clinically, patients showed various outcomes to multimodality therapies. Nowadays, personalized and precision medicine has become essential in the treatment of rectal cancer. Recent studies conducted gene expression profiling to predict the response and long-term prognosis of malignancies[3,4]; however, no consensus was achieved on prognostic gene profiling for rectal cancer.

Vascular endothelial growth factor (VEGF) possesses a significant role in angiogenesis by binding to VEGFR1 and VEGFR2, which is required for cancer progression and metastasis[5,6]. A phase Ⅱ trial indicated that VEGF could predict the pathological response to locally advanced rectal cancer patients treated with neoadjuvant cetuximab-based chemoradiation[7]. In addition, class Ⅲ β-tubulin (TUBB3) has been reported to play a critical role in tumor development and malignant transformation as a β-tubulin isotype. The variable levels of expression of the gene have been reported in colon, lung, ovary, kidney, prostate, and throat cancer with solid tumors[8-10]. However, only a few studies focused on its role in rectal cancer.

Herein, our study attempted to explore the potential prognostic value of VEGFR1 and TUBB3 for long-term survival in non-metastatic rectal cancer.

MATERIALS AND METHODS

Patients

Eighty cases of well-preserved formalin-fixed and paraffin embedded tumor tissue specimens that had undergone total mesorectal excision (TME) at the Fujian Cancer Hospital from March 2004 to November 2008 were retrospectively examined. Among these, two patients with previous malignancy and three with distant metastasis were excluded. Finally, 75 patients who fulfilled the following inclusion criteria were enrolled in the study: (1) Pathologically confirmed as primary rectal adenocarcinoma; (2) underwent TME; (3) no evidence of distant metastasis; (4) no previous or concurrent malignancy; and (5) complete follow-up information was obtained.

The variables such as gender, age, preoperative carcino-embryonic antigen (pre-CEA), pre-operative hemoglobin (pre-Hb), distance to the verge, T stage, lymph node metastasis, venous invasion, and tumor differentiation were considered. The T stage and lymph node metastasis were re-diagnosed based on the 8th Edition of the American Joint Committee on Cancer (AJCC)[11].

Treatments and follow-up

All patients underwent TME, including abdominoperineal resection and low anterior resection. Of these, eight cases received neoadjuvant chemoradiotherapy followed by TME. A total of 66 cases received 5-fluorouracil (5-FU)-based chemotherapy. The overall survival (OS)

Table 1 Patient characteristics

Characteristics	Data, *n* (%)
Gender	
Female	36 (48)
Male	39 (52)
Age (yr)	
median (range)	52 (29-74)
≤ 60	58 (77.3)
> 60	17 (22.7)
Pre-CEA (ng/mL)	
≤ 5	36 (63.2)
> 5	21 (36.8)
Pre-Hb (g/L)	
≤ 120	26 (34.7)
> 120	49 (65.3)
Distance to verge (cm)	
≤ 5	46 (61.3)
> 5	29 (38.7)
T stage	
T1 + T2	13 (17.3)
T3 + T4	63 (82.6)
Lymph node metastasis	
Negative	22 (29.3)
Positive	53 (70.6)
Venous invasion	
Negative	68 (90.7)
Positive	7 (9.3)
Tumor differentiation	
Poorly differentiated	20 (26.7)
Moderately-well differentiated	55 (73.3)
Chemotherapy	
No	9 (12)
Yes	66 (88)
TUBB3 expression	
Low-expression	39 (52)
High-expression	36 (48)
VEGFR1 expression	
Low-expression	53 (70.7)
High-expression	22 (29.3)
TUBB3 and VEGFR1	
Both low expression	32 (42.6)
Others	43 (57.3)

Pre-CEA: Preoperative carcino-embryonic antigen; Pre-Hb: Preoperative hemoglobin.

was defined as the duration from the date of diagnosis to the last follow-up or the date of death due to any cause, which was obtained from the medical records and telephonic interviews.

Multiplex branched DNA liquidchip technology

The formalin-fixed and paraffin embedded (FFPE) tumor tissue specimens containing more than 70% of tumor cells were selected. The Multiplex branched DNA liquidchip (MBL) technology (Guangzhou SurExam Bio-Tech Co., Ltd., China) was implemented to determine the mRNA expression levels of VEGFR1 and TUBB3. The FFPE tissue samples were lysed in the presence of proteinase K, at 56℃ for 2 h. Then, the lysate was transferred to a 96-well plate containing the blocking reagent, capture beads with probes for VEGFR1 and TUBB3, and target gene-specific probe sets. The sandwich nucleic acid hybridization was carried out for 16 h. The unbound RNA was removed by three washes with buffer under a vacuum system. The signal bound to the target mRNA was amplified with a streptavidin-conjugated phycoerythrin solution at 50℃ for 30 min. The fluorescence values of the samples were identified and analyzed using Luminex 200 system (Luminex, Austin, TX, United States), which were regarded as the RNA expression levels of each gene. The cutoff point of mRNA expression affecting the survival was determined by the Cutoff Finder application[12].

Statistical analysis

The end point of our analysis was OS. The association of gene expression level and clinicopathological features was studied by the χ^2 and Fisher's exact tests. The association between the mRNA expressions of VEGFR1 and TUBB3 was studied by the Spearman correlation test. The Kaplan–Meier test was used to analyze the OS, and Cox regression model (LR forward) was employed for univariate and multivariate analysis. Receiver operating characteristic (ROC) analysis was employed for assessing the specificity as well as the sensitivity of predicting OS by specific parameters. The statistical significance of area under the ROC (area under curve, AUC) was calculated by Delong's test[13]. *P*-values < 0.05 were deemed significant. The statistical analysis was conducted by SPSS version 22.0 (IBM Corporation, Armonk, NY, United States). The statistical methods of our study were reviewed by Qian-yu Ni from The First Affiliated Hospital of Fujian Medical University.

RESULTS

Patient characteristics

A total of 75 patients were enrolled in the present study. The characteristics of non-metastatic patients are summarized in Table 1. Median follow-up time was 102.7 mo (range: 6.0-153.6). The cohort comprised of 39 (52%) male and 36 (48%) female cases with the median age 52 years (range, 29-74). Among these patients, 21 (36.8%) cases presented pre-CEA records that were higher than 5 ng/mL, while they could not be accessed for 18 cases. In the case of pre-Hb, 26 (34.7%) patients were ≤ 120 g/L and the remaining were > 120 g/L. In terms of the tumor location, 46 (61.3%) patients had low rectal cancer (0-5 cm distance to verge), while the other 29 (38.7%) patients were > 5 cm. In all, 22 (29.3%) with lymph node metastasis positive and 53 (70.6%) were negative. Twenty (26.7%) patients were identified as poorly differentiated and 55 (73.3%) as moderate-to-well differentiated. According to the Cutoff Finder software, 0.0575 and 0.2025 were considered as the optimal cutoff point for the VEGFR1 and TUBB3 expression value, respectively (Figure 1). In addition, 36 (48%) and 22 (29.3%) patients showed a high expression of VEGFR1 and TUBB3, respectively.

Associations between mRNA expression and clinicopathological features

The correlations between VEGFR1/TUBB3 mRNA expre-

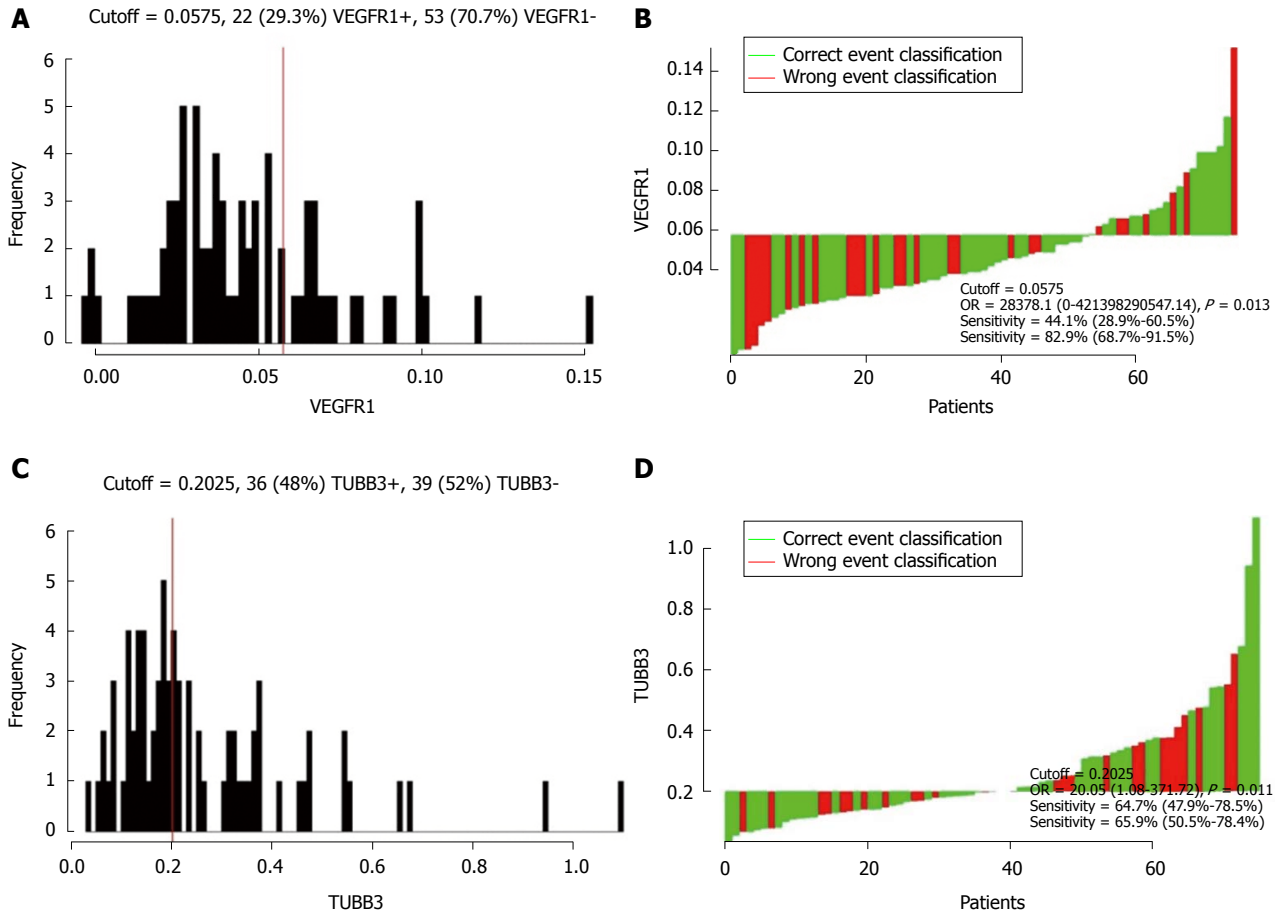


Figure 1 Distribution-based cutoff optimization of vascular endothelial growth factor receptor 1 and class Ⅲ β-tubulin expression value in 75 non-metastatic rectal cancer patients. A: Histograms of vascular endothelial growth factor receptor 1 (VEGFR1) expression value; B: Waterfall plot of optimal dichotomization for VEGFR1 expression value; C: Histograms of class Ⅲ β-tubulin expression value; D: Waterfall plot of optimal dichotomization for VEGFR1 expression value.

ssion and clinicopathological features were analyzed (Table 2). A majority of the patients displayed positive lymph node metastasis in the high-expression group of VEGFR1 (P = 0.013). However, no significant difference was found between the expression level of TUBB3 expression and clinicopathological features (gender, age, pre-CEA, pre-Hb, distance to the verge, T stage, lymph node metastasis and venous invasion, all P > 0.05).

Impact of VEGFR1 and TUBB3 on OS

The Cox regression analysis of OS influencing factors was shown in Table 3. Univariate analysis showed that T stage, lymph node metastasis, tumor differentiation, and VEGFR1 and TUBB3 expression were significantly related to OS (P = 0.048, P = 0.003, P = 0.052, P = 0.003 and P = 0.015, respectively) (Figures 2, 3 A and B). Moreover, Kaplan-Meier analysis showed that the rates of 1-, 3-, and 5-year OS in the TUBB3 low- and high-expression groups were 94.9% *vs* 94.4%, 76.9% *vs* 52.8%, and 71.8% *vs* 47.2%, respectively (P = 0.017). The rates of OS in the VEGFR1 low- and high-expression groups were 98.1% *vs* 86.4%, 77.4% *vs* 36.4%, and 69.8% *vs* 36.4%, respectively (P = 0.003). Moreover, lymph node metastasis (HR = 3.042, 95%CI: 1.137-8.142, P = 0.027) and VEGFR1 (HR = 2.151, 95%CI: 1.062-4.355, P = 0.033) were independent factors influencing OS, as evaluated by the multivariate Cox regression model.

Prognostic value of different combinations on survival

VEGFR1 and TUBB3 expression were positively correlated (P = 0.006, r = 0.315) by the Spearman's correlation test. Both low expression of VEGFR1 and TUBB3 were observed in 32 (42.6%) cases. Moreover, the Kaplan-Meier analysis showed that the 1-, 3-, and 5-year OS of both low-expression patients *vs* others were 96.9% *vs* 93.0%, 84.4% *vs* 53.5%, and 78.1% *vs* 46.5%, respectively (P = 0.003, Figure 3C). Meanwhile, Kaplan-Meier analysis showed that the rates of 1-, 3-, and 5-year OS in positive lymph node metastasis patients with high expression of VEGFR1 *vs* others were 90.0% *vs* 98.2%, 35.0% *vs* 78.2%, and 30.0% *vs* 70.9%, respectively (P < 0.001) (Figure 3D).

Finally, we combined the two independent prognostic factors, lymph node metastasis and VEGFR1 expression, to construct a prognostic model and supplemented the VEGFR1 expression to the lymph node metastasis

Table 2 Correlation between vascular endothelial growth factor receptor 1 and class III β-tubulin expression with clinicopathological features

Parameter	TUBB3			VEGFR1		
	Low (*n*)	High (*n*)	*P*	Low (*n*)	High (*n*)	*P*
Gender			0.426			0.081
Female	17	19		22	14	
Male	22	17		31	8	
Age (yr)			0.31			1
≤ 60	32	26		41	17	
> 60	7	10		12	5	
Pre-CEA			0.203			0.244
≤ 5	20	16		26	10	
> 5	8	13		12	9	
Pre-Hb			0.801			0.206
≤ 120	13	13		16	10	
> 120	26	23		37	12	
Distance to verge (cm)			0.608			0.792
≤ 5	25	21		32	14	
> 5	14	15		21	8	
T stage			0.883			0.744
T1 + T2	7	6		10	3	
T3 + T4	32	30		43	19	
Lymph node metastasis			0.071			0.013
Negative	15	7		20	2	
Positive	24	29		33	20	
Tumor thrombus			0.25			1
Negative	37	31		48	20	
Positive	2	5		5	2	
Tumor differentiation			0.754			0.939
Poorly	11	9		14	6	
Moderately-well	28	27		39	16	
Chemotherapy			0.156			0.051
No	7	2		9	0	
Yes	32	34		44	22	
VEGFR1			0.024			
Low	32	21				
High	7	15				

Pre-CEA: Preoperative carcino-embryonic antigen; Pre-Hb: Preoperative-hemoglobin.

by ROC analysis to assess the improvement of the model for OS. The lymph node metastasis (AUC: 0.688, 95%CI: 0.567–0.808, $P = 0.005$) showed a better prognostic value than VEGFR1 expression (AUC: 0.635, 95%CI: 0.507–0.764, $P = 0.045$). Furthermore, a better prognostic value was shown when combining the lymph node metastasis and VEGFR1 expression (AUC: 0.748, 95%CI: 0.637–0.859, $P < 0.001$) (Figure 4).

DISCUSSION

Firstly, we evaluated the long-term prognostic value of VEGFR1 and TUBB3 expression after the diagnosis of non-metastatic rectal cancer with a median follow-up of 102 mo. Here, we found that VEGFR1 and TUBB3 expression affected OS in non-metastatic rectal cancer by univariate analysis. Moreover, a favorable OS in both low expression of VEGFR1 and TUBB3 was noted as compared to others. In addition, the association between VEGFR1 expression and lymph node metastasis was also assessed. The combination of lymph node metastasis and VEGFR1 expression might also provide a promising tool for the prognosis of non-metastatic rectal cancer.

Reportedly, VEGFR correlates with poor prognosis, metastasis, and recurrence in various tumor types, including breast and lung cancers[14,15]. Moreover, previous studies demonstrated that VEGF plays a crucial role as a potent angiogenic factor in both experimental and human studies with respect to colorectal cancer progression and metastasis[16-18]. The co-expression of VEGF and VEGFR1/2 in the nucleus stimulates the proliferation and migration of endothelial cells, thereby providing nutrition for growing tumors and establishing a continuity between tumor cells and host vasculature[19].

VEGFR1 is primarily localized in the nucleus of endothelial cells; As the predominant receptor of the tumor microenvironment, it is essential for the survival of endothelial cells[20]. Tsai *et al*[21] reported that the overexpression of VEGF is a significant positive predictor for early postoperative relapse in stage Ⅰ-Ⅲ colorectal cancer patients, leading to poor OS ($P = 0.002$). Similarly, Nriagu *et al*[22] reported that the overexpression of VEGF mRNA was an independent factor affecting OS as assessed by multivariate analysis (HR = 1.94, $P = 0.005$). Herein, we found that the low expression of VEGFR1 might positively affect OS with a 5-year OS of 69.8% for low

Table 3 Cox regression analysis for overall survival

Variables	Univariate			Multivariate		
	HR	95%CI	*P*	HR	95%CI	*P*
Gender						
Female/male	1.018	0.519-1.997	0.958			
Age						
≤ 60/> 60	1.175	0.548-2.518	0.679			
Pre-CEA						
≤ 5/> 5	1.067	0.496-2.298	0.868			
Pre-Hb						
≤ 120/> 20	0.651	0.328-1.290	0.219			
Distance to verge (cm)						
≤ 5/> 5	1.265	0.642-2.491	0.497			
T stage						
T1 + T2/T3 + T4	4.221	1.011-17.632	0.048	4.05	0.968-116.93	0.055
Lymph node metastasis						
Negative/positive	6.247	1.905-20.491	0.003	3.042	1.137-8.142	0.027
Tumor thrombus						
Negative/positive	1.303	0.458-3.705	0.62			
Tumor differentiation						
Poorly/moderately-well	0.503	0.251-1.006	0.052	-	-	0.18
Chemotherapy						
No/yes	1.407	0.430-4.605	0.572			
TUBB3 expression						
Low/high	2.407	1.188-4.877	0.015	-	-	0.1
VEGFR1 expression						
Low/high	2.817	1.424-5.570	0.003	2.151	1.062-4.355	0.033

Pre-CEA: Preoperative carcino-embryonic antigen; Pre-Hb: Preoperative hemoglobin.

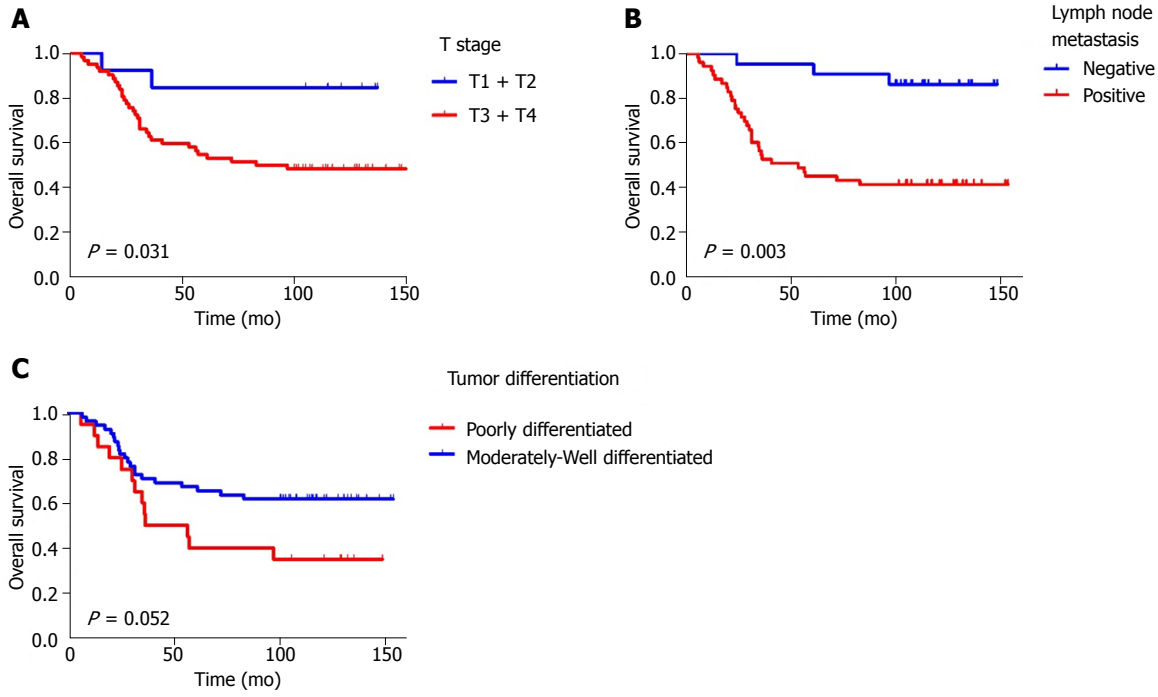


Figure 2 Kaplan-Meier survival curves of overall survival. A: T stage (T1 + T2 *vs* T3 + T4, *P* = 0.031); B: Lymph node metastasis (negative *vs* positive, *P* = 0.003); C: Tumor differentiation (poorly differentiated *vs* moderately-well differentiated, *P* = 0.052).

vs 36.4% for the high-expression group (HR = 2.151, *P* = 0.033). These results indicated that VEGFR1 functions as a positive regulator of angiogenesis[23], which might lead to poor survival in cancer patients.

A previous study evaluated VEGF expression in 117 colorectal adenocarcinoma patients, and confirmed that lymph node metastasis (positive *vs* negative, *P* < 0.001) and TNM stage (stage Ⅲ *vs* Ⅰ/Ⅱ, *P* < 0.001) were related to increased VEGF expression. Moreover, the mean number of metastatic nodes was significantly associated with VEGF expression (1.06 ± 2.84 for low expression *vs* 2.45 ± 4.03 for high expression, *P* =

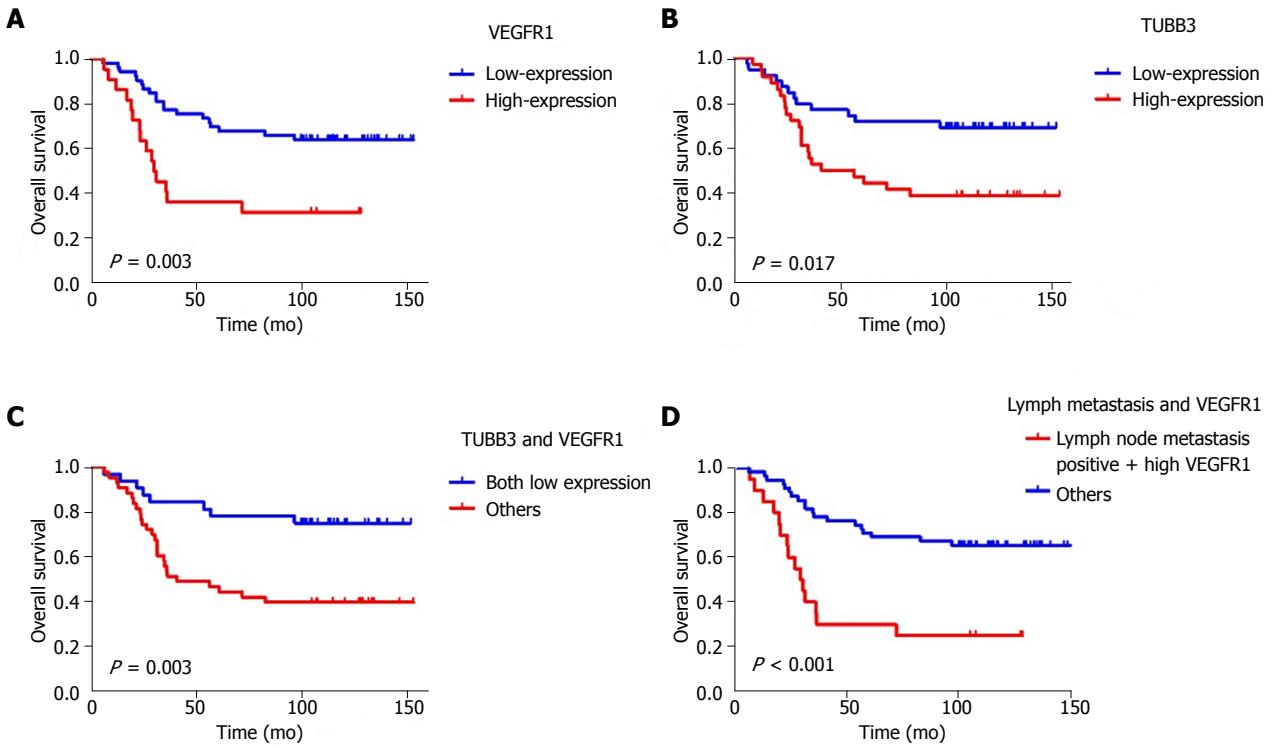


Figure 3 Kaplan-Meier survival curves of Overall Survival. A: Vascular endothelial growth factor receptor 1 (VEGFR1) expression (low *vs* high, $P = 0.003$); B: Class Ⅲ β-tubulin (TUBB3) expression (low *vs* high, $P = 0.017$); C: TUBB3 and VEGFR1 (both low expression *vs* others, $P = 0.003$); D: TNM stage and VEGFR1 (stage Ⅲ + high VEGFR1 expression vs others, $P < 0.001$).

0.031)[24]. Similarly, our study implied that VEGFR1 expression was related to lymph node metastases ($P = 0.013$). However, whether the function of VEGF/VEGFR1 affects lymph node metastasis is yet unclear. Nagy *et al*[25] hypothesized that tumor cells in the circulation directly reached the regional lymph nodes through the supply vessels or blood vessel-lymph vessel junctions.

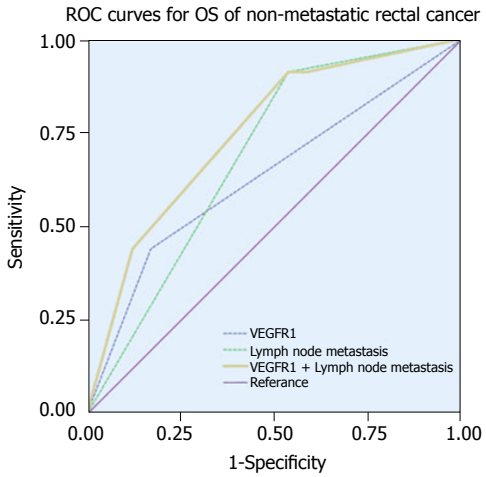
A retrospective study reported that VEGF expression could identify an unfavorable subgroup of patients with stage Ⅱ colon cancer for optimal treatment strategy (the recurrence rate was 50% for VEGF-positive *vs* 11.7% for VEGF-negative, $P = 0.001$)[26]. As shown by ROC curves in our analysis, though low sensitivity of VEGFR1 (44.1%), the specificity was high with 82.9%, which exerted a similar effect on prognosis as lymph node metastasis. Moreover, the sensitivity increased when combined with lymph node status, and a superior prognostic value was noted for the combination. Further identification of a group of lymph node metastasis-positive with high VEGFR1 expression allows for selective treatment with adjuvant chemotherapy using antiangiogenic therapy, including VEGFR1 antisense and monoclonal antibodies, as well as postoperative follow-up.

Several clinical studies demonstrated that the increased expression of TUBB3 in various human malignancies was related to low response rate and poor survival in patients treated with taxane-based chemotherapies[27-30]. However, studies focusing on the relationship between TUBB3 and non-metastatic rectal cancer are limited. The current study showed that the low expression of TUBB3 had better OS in non-metastatic rectal cancer patients as assessed by univariate analysis (5-year OS, 71.8% *vs* 47.2%), although no significant difference was observed by multivariate analysis.

Furthermore, Makarchenko *et al*[31] and Widow *et al*[32] reported that VEGFR1 regulated the chemo-resistant genes such as TUBB3, which might result in the poor prognosis of lung and gastroesophageal cancers. The current study established a positive correlation between VEGFR1 and TUBB3 ($r = 0.315$, $P = 0.006$), and a favorable OS was observed in both low expression groups ($P = 0.003$). Paradiso *et al*[33] had investigated the combination of TUBB3 and VEGFR1 in advanced breast cancer. Hypoxia in the tumor microenvironment promotes angiogenesis, and VEGFR1 is known to be related to angiogenesis[23]. TUBB3 was found to be involved in an adaptive response to low oxygen levels and poor nutrient supply in solid tumors[34,35]. Therefore, we speculate that the underlying mechanism of the two correlations might be related to anoxic environments.

Notably, this study was limited to a small-sample retrospective analysis. Thus, additional mRNA expression data might help to establish a superior predictor. Finally, prospective data and large sample size are essential for further substantiation of the results.

We confirmed that the increased expression of VEGFR1 and TUBB3 might be negatively correlated with long-term prognosis of non-metastatic rectal cancer.



	AUC	95%CI	*P*
VEGFR1	0.635	0.507-0.764	0.045
Lymph node metastasis	0.688	0.567-0.808	0.005
VEGFR1 + lymph node metastasis	0.748	0.637-0.859	< 0.001

Figure 4 Receiver operating characteristic analyses in non-metastatic rectal cancer patients. *P*-values show the area under the receiver operating characteristic (ROC) curves in the three models. ROC analyses of the prediction of overall survival by vascular endothelial growth factor receptor 1 (VEGFR1) expression model, lymph node metastasis, and the combined VEGFR1 expression-lymph node metastasis model.

Furthermore, VEGFR1 expression and lymph node metastasis affected the survival independently as well as synergistically. These results might provide additional prognostic information compared to the conventional tumor histopathological factors.

ARTICLE HIGHLIGHTS

Research background

Rectal cancer is one of the most common form of cancer in both men and women. Gene expression profiling for predicting the response and long-term prognosis of malignancies has been reported in recent decades. Vascular endothelial growth factor (VEGF) and class Ⅲ β-tubulin (TUBB3) have been reported to play a vital role in cancer progression. However, few studies focused on their role in rectal cancer.

Research motivation

We try to explore the potential prognostic value of VEGFR1 and TUBB3 for long-term survival in non-metastatic rectal cancer.

Research objectives

A total of 75 patients diagnosed with primary rectal adenocarcinoma without metastases were retrospectively analyzed.

Research methods

Multiplex branched DNA liquidchip technology was applied to detected mRNA expressions of VEGFR1 and TUBB3. The cutoff point of mRNA expression was determined by Cutoff Founder.

Research results

VEGFR1 expression was positively correlated to TUBB3. Patients with both low expression of TUBB3 and VEGFR1 presented a better overall survival (OS). In addition, VEGFR1 and lymph node metastasis had potential as prognostic factors for OS in non-metastatic rectal cancer patients, and the combination of them showed a favorable prognostic value.

Research conclusions

We confirmed that the increased expression of VEGFR1 and TUBB3 might be negatively correlated with long-term prognosis of non-metastatic rectal cancer. Furthermore, VEGFR1 expression and lymph node metastasis affected the survival independently, as well as synergistically. These results might provide additional prognostic information compared to the conventional tumor histopathological factors.

Research perspectives

VEGFR1 has the potential to contribute to decision making regarding individual treatment in rectal cancer. A larger sample size and additional mRNA expression data are warranted to establish a superior prognosis model.

REFERENCES

1 **Siegel RL**, Miller KD, Jemal A. Cancer Statistics, 2017. *CA Cancer J Clin* 2017; **67**: 7-30 [PMID: 28055103 DOI: 10.3322/caac.21387]
2 **Barr RD**. A two-year prospective analysis of emergency admissions to an adult medical unit at the Kenyatta National Hospital, Nairobi. *East Afr Med J* 1972; **49**: 772-782 [PMID: 4666003 DOI: 10.1001/jamasurg.2014.1756]
3 **Wang H**, Yang B, Geng T, Li B, Dai P, Chen C. Tissue-specific selection of optimal reference genes for expression analysis of anticancer drug-related genes in tumor samples using quantitative real-time RT-PCR. *Exp Mol Pathol* 2015; **98**: 375-381 [PMID: 25445497 DOI: 10.1016/j.yexmp.2014.10.014]
4 **Banerjee CM**. Dr. K.C. Chaudhuri--reminiscences. *Indian J Pediatr* 1974; **41**: 89-90 [PMID: 4609421 DOI: 10.1155/2015/921435]
5 **Folkman J**. Tumor angiogenesis: therapeutic implications. *N Engl J Med* 1971; **285**: 1182-1186 [PMID: 4938153 DOI: 10.1056/NEJM197111182852108]
6 **Goto Y**. Experimental animal models of diabetes mellitus. *Nihon Rinsho* 1991; **49** Suppl: 789-795 [PMID: 1851902 DOI: 10.1016/j.ygyno.2006.10.062]
7 **Grimminger PP**, Danenberg P, Dellas K, Arnold D, Rödel C, Machiels JP, Haustermans K, Debucquoy A, Velenik V, Sempoux C, Bracko M, Hölscher AH, Semrau R, Yang D, Danenberg K, Lenz HJ, Vallböhmer D. Biomarkers for cetuximab-based neoadjuvant radiochemotherapy in locally advanced rectal cancer. *Clin Cancer Res* 2011; **17**: 3469-3477 [PMID: 21558395 DOI: 10.1158/1078-0432.CCR-10-2273]
8 **Katsetos CD**, Herman MM, Mörk SJ. Class III beta-tubulin in human development and cancer. *Cell Motil Cytoskeleton* 2003; **55**: 77-96 [PMID: 12740870 DOI: 10.1002/cm.10116]
9 **Leandro-García LJ**, Leskelä S, Landa I, Montero-Conde C, López-Jiménez E, Letón R, Cascón A, Robledo M, Rodríguez-Antona C. Tumoral and tissue-specific expression of the major human beta-tubulin isotypes. *Cytoskeleton* (Hoboken) 2010; **67**: 214-223 [PMID: 20191564 DOI: 10.1002/cm.20436]
10 **Tsourlakis MC**, Weigand P, Grupp K, Kluth M, Steurer S, Schlomm T, Graefen M, Huland H, Salomon G, Steuber T, Wilczak W, Sirma H, Simon R, Sauter G, Minner S, Quaas A. βIII-tubulin overexpression is an independent predictor of prostate cancer progression tightly linked to ERG fusion status and PTEN deletion. *Am J Pathol* 2014; **184**: 609-617 [PMID: 24378408 DOI: 10.1016/j.ajpath.2013.11.007]
11 **Yao H**, Wu H, Liu Y. [Improvement of prognostic and predictive network of colorectal cancer based upon the 8th edition of AJCC colorectal cancer staging system]. *Zhonghua Wei Chang Wai Ke Za Zhi* 2017; **20**: 24-27 [PMID: 28105615]
12 **Meneilly GS**, Elahi D, Minaker KL, Rowe JW. The dawn phenomenon does not occur in normal elderly subjects. *J Clin Endocrinol Metab* 1986; **63**: 292-296 [PMID: 3522617 DOI: 10.1371/journal.pone.0051862]
13 **DeLong ER**, DeLong DM, Clarke-Pearson DL. Comparing the areas under two or more correlated receiver operating characteristic curves: a nonparametric approach. *Biometrics* 1988; **44**: 837-845 [PMID: 3203132 DOI: 10.2307/2531595]

14 **Brancatisano A**, Amis TC, Tully A, Engel LA. Blood flow distribution within the rib cage muscles. *J Appl Physiol* (1985) 1991; **70**: 2559-2565 [PMID: 1885450 DOI: 10.1371/journal.pmed.0040186]

15 **Fontanini G**, Lucchi M, Vignati S, Mussi A, Ciardiello F, De Laurentiis M, De Placido S, Basolo F, Angeletti CA, Bevilacqua G. Angiogenesis as a prognostic indicator of survival in non-small-cell lung carcinoma: a prospective study. *J Natl Cancer Inst* 1997; **89**: 881-886 [PMID: 9196255 DOI: 10.1093/jnci/89.12.881]

16 **Sky-Peck HH**. Distribution of trace elements in human hair. *Clin Physiol Biochem* 1990; **8**: 70-80 [PMID: 2361355 DOI: 10.1038/sj.bjc.6603176]

17 **Chin KF**, Greenman J, Gardiner E, Kumar H, Topping K, Monson J. Pre-operative serum vascular endothelial growth factor can select patients for adjuvant treatment after curative resection in colorectal cancer. *Br J Cancer* 2000; **83**: 1425-1431 [PMID: 11076648 DOI: 10.1054/bjoc.2000.1508]

18 **Tamura M**, Oda M, Tsunezuka Y, Matsumoto I, Kawakami K, Watanabe G. Vascular endothelial growth factor expression in metastatic pulmonary tumor from colorectal carcinoma: utility as a prognostic factor. *J Thorac Cardiovasc Surg* 2004; **128**: 517-522 [PMID: 15457151 DOI: 10.1016/j.jtcvs.2004.03.056]

19 **Grau U**. Chemical stability of insulin in a delivery system environment. *Diabetologia* 1985; **28**: 458-463 [PMID: 3899829 DOI: 10.1053/j.gastro.2013.10.011]

20 **Gandolfi SA**, Maier JA, Petronini PG, Wheeler KP, Borghetti AF. Multicomponent analysis of amino acid transport System L in normal and virus-transformed fibroblasts. *Biochim Biophys Acta* 1987; **904**: 29-35 [PMID: 2822115 DOI: 10.1038/cdd.2009.152]

21 **Tsai HL**, Yang IP, Lin CH, Chai CY, Huang YH, Chen CF, Hou MF, Kuo CH, Juo SH, Wang JY. Predictive value of vascular endothelial growth factor overexpression in early relapse of colorectal cancer patients after curative resection. *Int J Colorectal Dis* 2013; **28**: 415-424 [PMID: 22961433 DOI: 10.1007/s00384-012-1570-z]

22 **Nriagu JO**. Eric I. Hamilton. *Sci Total Environ* 1991; **100**: viii-vxvi [PMID: 2063176 DOI: 10.1016/0048-9697(91)90367-N]

23 **Veikkola T**, Karkkainen M, Claesson-Welsh L, Alitalo K. Regulation of angiogenesis via vascular endothelial growth factor receptors. *Cancer Res* 2000; **60**: 203-212 [PMID: 10667560]

24 **Zafirellis K**, Agrogiannis G, Zachaki A, Gravani K, Karameris A, Kombouras C. Prognostic significance of VEGF expression evaluated by quantitative immunohistochemical analysis in colorectal cancer. *J Surg Res* 2008; **147**: 99-107 [PMID: 17655863 DOI: 10.1016/j.jss.2007.05.041]

25 **Nagy JA**, Brown LF, Senger DR, Lanir N, Van de Water L, Dvorak AM, Dvorak HF. Pathogenesis of tumor stroma generation: a critical role for leaky blood vessels and fibrin deposition. *Biochim Biophys Acta* 1989; **948**: 305-326 [PMID: 2465781 DOI: 10.1016/0304-419X(89)90004-8]

26 **Cascinu S**, Staccioli MP, Gasparini G, Giordani P, Catalano V, Ghiselli R, Rossi C, Baldelli AM, Graziano F, Saba V, Muretto P, Catalano G. Expression of vascular endothelial growth factor can predict event-free survival in stage II colon cancer. *Clin Cancer Res* 2000; **6**: 2803-2807 [PMID: 10914727]

27 **DeCamp MM**, Demling RH. Posttraumatic multisystem organ failure. *JAMA* 1988; **260**: 530-534 [PMID: 3290526 DOI: 10.1158/0008-5472.CAN-10-1447]

28 **Galmarini CM**, Treilleux I, Cardoso F, Bernard-Marty C, Durbecq V, Gancberg D, Bissery MC, Paesmans M, Larsimont D, Piccart MJ, Di Leo A, Dumontet C. Class III beta-tubulin isotype predicts response in advanced breast cancer patients randomly treated either with single-agent doxorubicin or docetaxel. *Clin Cancer Res* 2008; **14**: 4511-4516 [PMID: 18628466 DOI: 10.1158/1078-0432.CCR-07-4741]

29 **Ferrandina G**, Zannoni GF, Martinelli E, Paglia A, Gallotta V, Mozzetti S, Scambia G, Ferlini C. Class III beta-tubulin overexpression is a marker of poor clinical outcome in advanced ovarian cancer patients. *Clin Cancer Res* 2006; **12**: 2774-2779 [PMID: 16675570 DOI: 10.1158/1078-0432.CCR-05-2715]

30 **Li WJ**, Zhong SL, Wu YJ, Xu WD, Xu JJ, Tang JH, Zhao JH. Systematic expression analysis of genes related to multidrug-resistance in isogenic docetaxel- and adriamycin-resistant breast cancer cell lines. *Mol Biol Rep* 2013; **40**: 6143-6150 [PMID: 24078162 DOI: 10.1007/s11033-013-2725-x]

31 **Makarchenko OF**. The state and prospectives of physiology in the Unkrainian SSR. *Fiziol Zh* 1972; **18**: 435-445 [PMID: 4568798]

32 **Widow W**. Treatment situation in bronchial carcinoma. *Z Erkr Atmungsorgane Folia Bronchol* 1971; **134**: 57-65 [PMID: 5209524]

33 **Paradiso A**, Mangia A, Chiriatti A, Tommasi S, Zito A, Latorre A, Schittulli F, Lorusso V. Biomarkers predictive for clinical efficacy of taxol-based chemotherapy in advanced breast cancer. *Ann Oncol* 2005; **16** Suppl 4: iv14-iv19 [PMID: 15923415 DOI: 10.1093/annonc/mdi902]

34 **Raspaglio G**, De Maria I, Filippetti F, Martinelli E, Zannoni GF, Prislei S, Ferrandina G, Shahabi S, Scambia G, Ferlini C. HuR regulates beta-tubulin isotype expression in ovarian cancer. *Cancer Res* 2010; **70**: 5891-5900 [PMID: 20587520 DOI: 10.1158/0008-5472.CAN-09-4656]

35 **Raspaglio G**, Filippetti F, Prislei S, Penci R, De Maria I, Cicchillitti L, Mozzetti S, Scambia G, Ferlini C. Hypoxia induces class III beta-tubulin gene expression by HIF-1alpha binding to its 3' flanking region. *Gene* 2008; **409**: 100-108 [PMID: 18178340 DOI: 10.1016/j.gene.2007.11.015]

P- Reviewer: Jorgensen JT, Shu X, Soh JS **S- Editor**: Cui LJ
L- Editor: Filipodia **E- Editor**: Tan WW

World Journal of Gastrointestinal Oncology

Submit a Manuscript: http://www.f6publishing.com

World J Gastrointest Oncol 2018 October 15; 10(10): 360-366

DOI: 10.4251/wjgo.v10.i10.360

ISSN 1948-5204 (online)

ORIGINAL ARTICLE

Retrospective Study

Predictive factors for lymph node metastasis and defining a subgroup treatable for laparoscopic lymph node dissection after endoscopic submucosal dissection in poorly differentiated early gastric cancer

Hua Li, Zhi-Bin Huo, Fan-Ting Kong, Qing-Qiang He, Yun-He Gao, Wen-Quan Liang, Deng-Xiang Liu

Hua Li, Zhi-Bin Huo, Fan-Ting Kong, Qing-Qiang He, Department of Surgical Oncology, Xing Tai People Hospital, Xingtai 054001, Hebei Province, China

Yun-He Gao, Wen-Quan Liang, Department of General Surgery, Chinese People's Liberation Army General Hospital, Beijing 100853, China

Deng-Xiang Liu, Institute of Cancer Control, Xing Tai People Hospital, Xingtai 054001, Hebei Province, China

Author contributions: Li H, Huo ZB and Fan-Ting Kong contributed equally to this work. Liu DX, Li H, designed the research; Li H, Huo ZB and Fan-Ting Kong analyzed the data and drafted the manuscript; He QQ revised the manuscript critically for important intellectual content and contributed to the data analysis; Gao YH and Liang WQ helped draft the manuscript; all authors read and approved the final manuscript.

ORCID number: Hua Li (0000-0003-2423-7689); Zhi-Bin Huo (0000-0003-3985-1972); Fan-Ting Kong (0000-0003-2098-0002); Qing-Qiang He (0000-0001- 7771-1618); Yun-He Gao (0000-0002-2357-0693); Wen-Quan Liang (0000-0002- 8667-0958); Deng-Xiang Liu (0000-0003-4047-1379).

Institutional review board statement: This study is a retrospective study for the data of patients collected from the Department of Surgical Oncology, Affiliated Xing Tai People's Hospital of Hebei Medical University during 1990-2015. No human body was involved in this study. In our hospital policy, this study does not require approval by the hospital.

Informed consent statement: All study participants, or their legal guardian, provided informed written consent prior to study enrollment.

Conflict-of-interest statement: No conflict of interest was declared by the authors.

Data sharing statement: No additional data are available.

Manuscript source: Unsolicited manuscript

Correspondence to: Deng-Xiang Liu, Academic Research, Chief Doctor, Surgical Oncologist, Institute of Cancer Control, Xing Tai People Hospital, No. 16 Hongxing Street, Xingtai 054001, Hebei Province, China. dengxianglfangliao@163.com
Telephone: +86-319-3286153
Fax: +86-319-3286153

Received: July 3, 2018
Peer-review started: July 3, 2018
First decision: July 11, 2018
Revised: August 24, 2018
Accepted: August 27, 2018
Article in press: August 28, 2018
Published online: October 15, 2018

Abstract

AIM

To investigate the predictive factors of lymph node metastasis (LNM) in poorly differentiated early gastric cancer (EGC); to guide the individual application of a combination of endoscopic submucosal dissection (ESD) and laparoscopic lymph node dissection (LLND) in a suitable subgroup of patients with poorly differentiated EGC.

METHODS
We retrospectively analyzed 138 patients with poorly differentiated EGC who underwent gastrectomy with lymphadenectomy between January 1990 and December 2015. The association between the clinicopathological factors and the presence of LNM was retrospectively analyzed by univariate and multivariate logistic regression analyses. Odds ratios (OR) with 95% confidence interval (95%CI) were calculated. We further examined the relationship between the positive number of the significant predictive factors and the LNM rate.

RESULTS
The tumor diameter (OR = 13.438, 95%CI: 1.773-25.673, *P* = 0.029), lymphatic vessel involvement (LVI) (OR = 38.521, 95%CI: 1.975-68.212, *P* = 0.015) and depth of invasion (OR = 14.981, 95%CI: 1.617-52.844, *P* = 0.024) were found to be independent risk factors for LNM by multivariate analysis. For the 138 patients diagnosed with poorly differentiated EGC, 21 (15.2%) had LNM. For patients with one, two and three of the risk factors, the LNM rates were 7.7%, 47.6% and 64.3%, respectively. LNM was not found in 77 patients that did not have one or more of the three risk factors.

CONCLUSION
ESD might be sufficient treatment for intramucosal poorly differentiated EGC if the tumor is less than or equal to 2 cm in size and when LVI is absent upon postoperative histological examination. ESD with LLND may lead to the elimination of unnecessary gastrectomy in poorly differentiated EGC.

Key words: Poorly differentiated cancer; Laparoscopic lymph node dissection; Lymph node metastasis; Early gastric cancer; Endoscopic submucosal dissection

Core tip: The new technique combines endoscopic submucosal dissection (ESD) with laparoscopic lymph node dissection (LLND), which may lead to the elimination of "unnecessary" gastrectomy in poorly differentiated early gastric cancer (EGC) patients that have a potential risk of lymph node metastasis (LNM). ESD followed by LLND enables the complete resection of the primary tumor and the histologic determination of the lymph node status. In this study, we determined the risk factors that were predictive of LNM in poorly differentiated EGC patients. Our results provided some suggestions to guide the application of combination of ESD and LLND for selected patients with poorly differentiated EGC.

Li H, Huo ZB, Kong FT, He QQ, Gao YH, Liang WQ, Liu DX. Predictive factors for lymph node metastasis and defining a subgroup treatable for laparoscopic lymph node dissection after endoscopic submucosal dissection in poorly differentiated early gastric cancer. *World J Gastrointest Oncol* 2018; 10(10): 360-366 Available from: URL: http://www.wjgnet.com/1948-5204/full/v10/i10/360.htm DOI: http://dx.doi.org/10.4251/wjgo.v10.i10.360

INTRODUCTION

Endoscopic submucosal dissection (ESD) has become widely accepted, as it provides *en bloc* resection and histologically complete resection and is a valuable alternative to gastrectomy for treating early gastric cancer (EGC)[1-4]. The accurate assessment of the potential presence of lymph node metastasis (LNM) is required for ESD. ESD can be used for EGC but it does not have the risk of LNM[5-7]. Because the risk of LNM is negligible (0%), ESD is often applied to well or moderately differentiated EGC confined to the mucosa without ulceration and smaller than or equal to 2 cm[8]. For undifferentiated EGC, the risk of LNM is higher so the usage of ESD has been limited. Thus, for patients with undifferentiated EGC, gastrectomy was accepted as a standard treatment. Undifferentiated carcinomas of gastric cancer consist of mucinous adenocarcinoma, primary signet ring cell carcinoma and poorly differentiated adenocarcinoma[8]. However, approximately 96.6% of poorly differentiated EGC cases with potential risk of LNM are eventually found to have no LNM after "unnecessary" gastrectomy, suggesting that it may be overtreatment for these cases[9]. The new technique combines ESD with laparoscopic lymph node dissection (LLND), which may lead to the elimination of "unnecessary" gastrectomy in EGC patients having a potential risk of LNM[10-13]. ESD followed by LLND enables the complete resection of the primary tumor and the histologic determination of the lymph node status.

In this retrospective study, we determined the risk factors that were predictive of LNM in poorly differentiated EGC patients. Our results provided some suggestions to guide the application of combination of ESD and LLND for selected patients with poorly differentiated EGC.

MATERIALS AND METHODS

Patients

EGC is considered to be a lesion confined to the mucosa or submucosa regardless of the presence or absence of LNM, according to the Japanese Classification of Gastric Carcinoma (JCGC)[8]. This retrospective study enrolled patients who had undergone radical gastrectomy due to EGC. The patients were from the Department of Surgical Oncology, Affiliated Xing Tai People's Hospital of Hebei Medical University (Xingtai, China). Time points were from January 1990 to December 2015.

For this current study, inclusion criteria included: (1) Diagnosed with poorly differentiated EGC depending on JCGC by pathological analyses through specimens and lymph nodes; (2) Lymph node dissection beyond limited (D1) dissection; (3) Over sixteen lymph nodes dissected; and (4) Available medical record from database.

Table 1 Univariate analysis of potential risk characteristics for lymph node metastasis *n* (%)

Factor	Lymph node metastasis	
	Positive	*P*-value
Age (yr)		
< 60 (*n* = 95)	16 (16.8)	0.494
⩾ 60 (*n* = 43)	5 (11.6)	
Sex		
Male (*n* = 87)	14 (16.1)	0.748
Female (*n* = 51)	7 (13.7)	
Macroscopic type		
Ⅰ (*n* = 6)	0 (0)	0.564
Ⅱ (*n* = 82)	12 (14.6)	
Ⅲ (*n* = 50)	9 (18.0)	
Family medical history		
Positive (*n* = 11)	2 (18.2)	0.809
Negative (*n* = 127)	19 (15.0)	
Location		
Upper (*n* = 29)	4 (13.8)	0.497
Middle (*n* = 8)	0 (0)	
Lower (*n* = 101)	17 (16.8)	
Number of tumors		
Single (*n* = 133)	20 (15.0)	0.799
Multitude (*n* = 5)	1 (20.0)	
Tumor size in diameter		
⩽ 2 cm (*n* = 78)	5 (6.4)	0.005
> 2 cm (*n* = 60)	16 (26.7)	
Ulceration		
Negative (*n* = 109)	18 (16.5)	0.474
Positive (*n* = 29)	3 (10.3)	
Lymphatic vessel involvement		
Negative (*n* = 122)	11 (9.0)	< 0.001
Positive (*n* = 16)	10 (62.5)	
Depth of invasion		
Mucosa (*n* = 83)	5 (6.0)	0.002
Submucosa (*n* = 55)	16 (29.1)	

During the 25 years, a total of 138 patients (87 men and 51 women) with histopathologically poorly differentiated EGC were included for analyses. The ages of the patients ranged from 29 to 81 years (mean 49).

Dissection and classification of lymph nodes

For each patient, lymph nodes were dissected from the *en bloc* specimens. The classification was performed according to the JCGC[8]. After careful review of specimens, an experienced surgeon gave the classification of the dissected lymph nodes[8]. After that, the lymph nodes were sectioned and the histopathologic and immunohistochemical features were detected by eosin and hematoxylin staining and immunohistochemistry. Pathological examination for metastasis and lymphatic vessel involvement (LVI) was detected by immunohistochemistry with D2-40. We used uniform measurement standards to guarantee uniformity of treatment among the sample over the 25 years. Histologic slides were reread in a blind manner by one pathologist. The main clinical and pathological data could be obtained from archival documents, including surgical report, conclusions of the pathologist, and the patient card.

Association between clinicopathological parameters and LNM

In this current study, we included clinicopathological parameters according to JCGC[8] for analysis. These parameters included family medical history of gastric cancer, gender (female, male), age (⩾ 60 years, < 60 years), lymphatic vessel involvement, depth of invasion (mucosa, submucosa), macroscopic type, ulceration, tumor size (maximum dimension ⩽ 2 cm, or > 2 cm), location of tumor (lower, middle, or upper stomach), number of tumors (single or multiple). As described below, the relationship between LNM and clinicopathological factors was explored.

Statistical analysis

Chi-squared test was performed to determine differences between patients with and without LNM in clinicopathological parameters. After that, multivariate stepwise logistic regression analysis was carried out to identify independent risk factors for LNM. Hazard ratio and 95% confidence interval (CI) were calculated. A P value < 0.05 was considered to have statistical significance. All statistical analyses were performed using SPSS v21.0 software (IBM Corp, Armonk, NY, United States).

RESULTS

Association between clinicopathological parameters and LNM

Table 1 showed the relationship of LNM and clinicopathological factors using a χ^2 test. Results showed that tumor diameter > 2 cm, the presence of LVI, and submucosal invasion were associated with a high LNM rate (P < 0.05). On the other hand, no significant association was observed between LNM and family medical history, macroscopic type, ulceration, location, number, age or gender.

Potential independent risk clinicopathological parameters for LNM

Univariate analysis results demonstrated that there are three significantly associated characteristics with LNM. Multivariate analysis showed that for LNM, all three characteristics were independent and significant risk factors (P < 0.05, Table 2).

LNM in poorly differentiated EGC

Twenty-one (15.2%) of 138 patients diagnosed with poorly differentiated EGC had LNM. The relationship between the three risk clinicopathological factors (tumor diameter > 2 cm, LVI, and submucosal invasion) and LNM was studied in poorly differentiated EGC. In poorly differentiated EGC, for patients with one, two or three risk factors, LNM rates were 7.7% (2/26), 47.6% (10/21) and 64.3% (9/14), respectively. For the other 77 patients without any of the risk factors, we did not find any LNM (Table 3).

DISCUSSION

Endoscopic treatments, such as EMR and ESD, are standard treatments for EGC. ESD is superior in allowing

Table 2 Multivariate analysis of potential risk factors for lymph node metastasis

Characters	Hazard ratio	95%CI	*P*-value
Tumor size			
⩽ 2 cm	13.438	1.773-25.673	0.029
> 2 cm			
Lymphatic vessel involvement			
Positive	38.521	1.975-69.212	0.015
Negative			
Depth of invasion			
Mucosa	14.981	1.617-52.844	0.024
Submucosa			

CI: Confidence interval.

Table 3 Association between the three identified risk factors and lymph node metastasis in poorly differentiated early gastric cancer

Number of positive risk factors	Lymph metastasis rate
None	0% (0/77)
One	9.1% (2/26)
Two	22.2% (10/21)
Three	57.1% (9/14)

en bloc resection at the submucosal location, leading to accurate pathologic assessment of specimens[14-16]. The dominance of ESD over surgery is less invasive, less expensive, and it better preserves physiological function[17,18]. ESD is applied to EGC without LNM, and the indication criteria for differentiated cancer. On the other hand, even though the gastric lesions can be completely removed with ESD for patients with poorly differentiated EGC, standard gastrectomy with lymph node dissection is usually performed. However, gastrectomy may be not necessary for poorly differentiated EGC patients, of which approximately 96.6% patients with surgically treatment actually do not have LNM[9]. Complications from gastrectomy are rare and not serious, including postoperative reflux esophagitis, dumping syndrome and impaired food intake[19,20]. If gastric lesions can be completely removed and lymph node status can be histologically determined before gastrectomy, unnecessary surgery could be obviated. The new technique combines ESD with LLND, and not only completely resects the primary tumor but also determines the histologic status of the lymph node.

A precise prediction of the presence of LNM plays a vital role in choosing ESD for EGC. The factors that can help to predict LNM have been verified by previous studies in EGC. However, few studies have tried to explore whether ESD can be used in poorly differentiated EGC. Thus, we would like to seek a possible way to expand ESD in poorly differentiated EGC. In this study, we retrospectively examined the poorly differentiated EGC cases to confirm whether LNM could be predicted. Our data indicated that LNM has significant predictive factors, including tumor diameter > 2 cm, presence of LVI, and submucosal invasion. This present study demonstrates that poorly differentiated EGC are in accordance with some published studies, indicating the existence of a significant correlation between the presence of LVI, submucosal invasion and large tumor size with high LNM incidence[21-29].

During the analysis of this study, numerous relevant subgroup analyses were also done to identify patients of whom the potential LNM can be excluded and then find the candidates who are potentially curable by ESD treatment. Interestingly, we found that patients whose tumor is confined to the intramucosa, and is less than or equal to 2 cm without LVI did not have LNM, indicating that for these cases, ESD could be sufficient and overtreatment may be avoided.

In addition, the association between the positive number of the three risk factors (presence of LVI, tumor diameter > 2 cm, and submucosal invasion) and LNM rate were further studied to discuss management strategies for the treatment of poorly differentiated EGC. From the results of this study, we have determined that there is a certain association between LNM rate and number of significant risk factors. When the number of factors is one, two or three, LNM rates were 7.7%, 47.6% and 64.3%, respectively. Therefore, gastrectomy with lymphadenectomy is preferable for these patients with risk factors.

Standard gastrectomy with lymphadenectomy remains of value as standard therapy for the potential presence of LNM in poorly differentiated EGC patients. However, the combination of ESD and LLND could avoid unnecessary gastrectomy. Studies have been reported that some patients with EGC received ESD, but the surgery could not meet standard or expanded resection. Salvage treatment of LLND showed overall survival benefits[30,31]. ESD has a high complete resection rate for localized primary tumor, and LLND has complementary surgical benefits, which could enable the confirmation of negative LNM[32]. Thus, this combination was a survival effective strategy compared to conventional treatment. Indeed, previous data have shown that this combination has a significantly greater effect on overall survival during the long-term follow-up period[33]. The combination of ESD and LLND has fewer complications (such as perforation, *etc*.) and can be used in any areas in the stomach. Therefore, the combination of ESD and

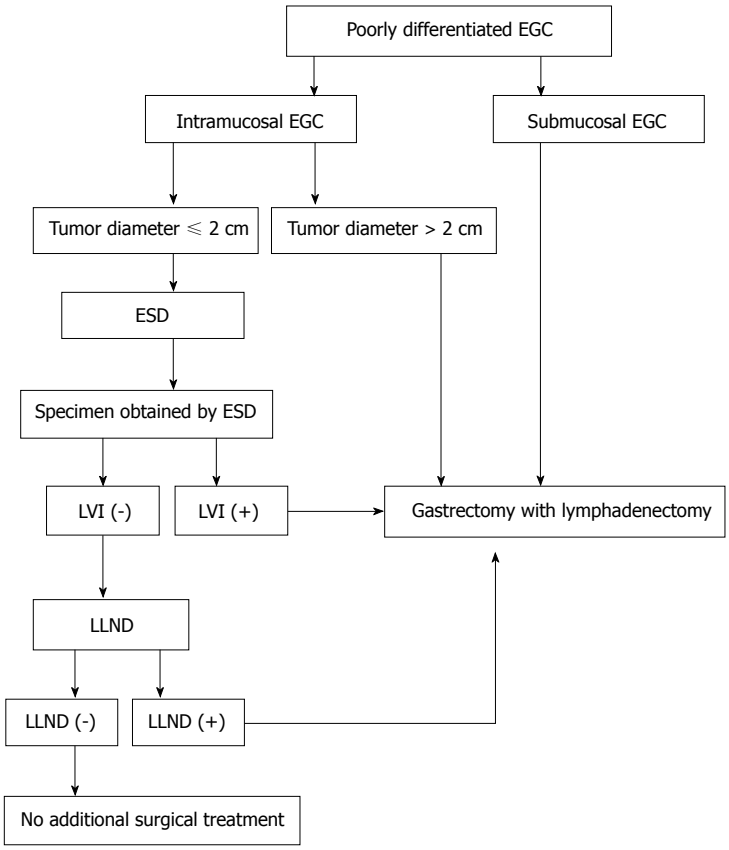


Figure 1 Flow chart of the therapeutic strategy for cases with poorly differentiated early gastric cancer. EGC: Early gastric cancer; ESD: Endoscopic submucosal dissection; LLND: Laparoscopic lymph node dissection; LVI: Lymphatic vessel involvement.

LLND may be an effective, minimally invasive treatment and beneficial for long-term quality of life in poorly differentiated EGC patients.

However, this study has several limitations. It was a single center study, and the sample size was relatively small. Moreover, our study was performed retrospectively, and the data collected were not randomized and could have been subject to associated bias. Therefore, our findings and conclusions may be not very informative to make robust conclusions. Randomized, prospective studies are needed to verify these results.

In this study, we proposed a novel treatment strategy for patients with poorly differentiated EGC (Figure 1). For patients with a tumor less than or equal to 2 cm in size or when LVI is absent upon postoperative histological examination, ESD might be sufficient treatment. The combination of ESD and LLND enables complete resection for not only the primary tumor but also the potentially metastatic lymph node. When LLND reveals LNM or specimens of ESD shows with LVI, gastrectomy with lymphadenectomy may be a better choice to achieve R0 resection. We believe that LLND may lead to the elimination of ESD in poorly differentiated EGC patients having a potential risk of LNM.

ARTICLE HIGHLIGHTS

Research background

For patients with poorly differentiated early gastric cancer (EGC), gastrectomy with lymphadenectomy is usually performed even though the gastric lesions can be completely removed with endoscopic submucosal dissection (ESD) due to the higher risk of lymph node metastasis (LNM). However, many surgical EGC cases actually do not have LNM, indicating that this surgery may not be necessary for many cases of EGC. To avoid this unnecessary surgery, the new technique combines ESD with laparoscopic lymph node dissection (LLND), which may lead to the elimination of unnecessary gastrectomy in poorly differentiated EGC patients having a potential risk of LNM.

Research motivation

We attempted to identify a subgroup of poorly differentiated EGC patients in whom the risk of LNM can be ruled out and treated them with ESD and LLND, which may serve as a breakthrough treatment for poorly differentiated EGC.

Research objectives

In this study, we intended to determine the risk factors that were predictive of LNM in poorly differentiated EGC patients and to provide some suggestions to guide the application of the combination of ESD and LLND for selected patients with poorly differentiated EGC.

Research methods

We retrospectively analyzed 138 patients with poorly differentiated EGC who underwent gastrectomy with lymphadenectomy (between January 1990 and December 2015). We also retrospectively analyzed (by univariate and multivariate logistic regression analyses) the association between the clinicopathological factors and the presence of LNM. We further examined the relationship between the positive number of the significant predictive factors and the LNM rate.

Research results

Tumor size, depth of invasion and lymphatic vessel involvement were found to be independently risk clinicopathological factors for LNM in poorly differentiated

EGC. Furthermore, we established a simple criterion to expand the possibility of using ESD and LLND for the treatment of poorly differentiated EGC.

Research conclusions

ESD might be sufficient treatment for intramucosal poorly differentiated EGC if the tumor is less than or equal to 2 cm in size, and when lymphatic vessel involvement is absent upon postoperative histological examination. We found that the ESD with LLND may lead to the elimination of unnecessary gastrectomy in poorly differentiated EGC.

Research perspectives

The minimization of therapeutic invasiveness in order to preserve quality of life is a major topic in the management of EGC. One of the critical factors in choosing minimally invasive surgery for EGC would be the precise prediction of whether the patient has LNM. Therefore, in the future, we will carry out this retrospective study to determine the clinicopathological factors that are predictive of LNM in EGC and to guide the individual application of minimally invasive surgery in a suitable subgroup of patients with EGC.

REFERENCES

1 **Koeda K**, Nishizuka S, Wakabayashi G. Minimally invasive surgery for gastric cancer: the future standard of care. *World J Surg* 2011; **35**: 1469-1477 [PMID: 21476116 DOI: 10.1007/s00268-011-1051-5]

2 **Jeon HK**, Lee SJ, Kim GH, Park DY, Lee BE, Song GA. Endoscopic submucosal dissection for undifferentiated-type early gastric cancer: short- and long-term outcomes. *Surg Endosc* 2018; **32**: 1963-1970 [PMID: 29046960 DOI: 10.1007/s00464-017-5892-3]

3 **Karttunen P**, Saano V, Paronen P, Peura P, Vidgren M. Pharmacokinetics of ibuprofen in man: a single-dose comparison of two over-the-counter, 200 mg preparations. *Int J Clin Pharmacol Ther Toxicol* 1990; **28**: 251-255 [PMID: 2376426 DOI: 10.1007/s00464-013-3030-4]

4 **Fukunaga S**, Nagami Y, Shiba M, Ominami M, Tanigawa T, Yamagami H, Tanaka H, Muguruma K, Watanabe T, Tominaga K, Fujiwara Y, Ohira M, Hirakawa K, Arakawa T. Long-term prognosis of expanded-indication differentiated-type early gastric cancer treated with endoscopic submucosal dissection or surgery using propensity score analysis. *Gastrointest Endosc* 2017; **85**: 143-152 [PMID: 27365265 DOI: 10.1016/j.gie.2016.06.049]

5 **Yoshida K**, Yamaguchi K, Okumura N, Osada S, Takahashi T, Tanaka Y, Tanabe K, Suzuki T. The roles of surgical oncologists in the new era: minimally invasive surgery for early gastric cancer and adjuvant surgery for metastatic gastric cancer. *Pathobiology* 2011; **78**: 343-352 [PMID: 22104206 DOI: 10.1159/000328197]

6 **Son T**, Kwon IG, Hyung WJ. Minimally invasive surgery for gastric cancer treatment: current status and future perspectives. *Gut Liver* 2014; **8**: 229-236 [PMID: 24827617 DOI: 10.5009/gnl.2014.8.3.229]

7 **Fatourou E**, Roukos DH. Endoscopic submucosal dissection: can it safely expand indications for a minimally invasive approach to patients with early gastric cancer? *Surg Endosc* 2010; **24**: 1793-4; author reply 1795 [PMID: 20041265 DOI: 10.1007/s00464-009-0844-1]

8 **Japanese Gastric Cancer Association**. Japanese classification of gastric carcinoma: 3rd English edition. *Gastric Cancer* 2011; **14**: 101-112 [PMID: 21573743 DOI: 10.1007/s10120-011-0041-5]

9 **Gotoda T**, Yanagisawa A, Sasako M, Ono H, Nakanishi Y, Shimoda T, Kato Y. Incidence of lymph node metastasis from early gastric cancer: estimation with a large number of cases at two large centers. *Gastric Cancer* 2000; **3**: 219-225 [PMID: 11984739 DOI: 10.1007/PL00011720]

10 **Ono H**, Kondo H, Gotoda T, Shirao K, Yamaguchi H, Saito D, Hosokawa K, Shimoda T, Yoshida S. Endoscopic mucosal resection for treatment of early gastric cancer. *Gut* 2001; **48**: 225-229 [PMID: 11156645 DOI: 10.1136/gut.48.2.225]

11 **Oda I**, Saito D, Tada M, Iishi H, Tanabe S, Oyama T, Doi T, Otani Y, Fujisaki J, Ajioka Y, Hamada T, Inoue H, Gotoda T, Yoshida S. A multicenter retrospective study of endoscopic resection for early gastric cancer. *Gastric Cancer* 2006; **9**: 262-270 [PMID: 17235627 DOI: 10.1007/s10120-006-0389-0]

12 **Gotoda T**. Endoscopic resection of early gastric cancer. *Gastric Cancer* 2007; **10**: 1-11 [PMID: 17334711 DOI: 10.1007/s10120-006-0408-1]

13 **Ryu SJ**, Kim BW, Kim BG, Kim JH, Kim JS, Kim JI, Park JM, Oh JH, Kim TH, Kim JJ, Park SM, Park CH, Song KY, Lee JH, Kim SG, Kim DJ, Kim W. Endoscopic submucosal dissection versus surgical resection for early gastric cancer: a retrospective multicenter study on immediate and long-term outcome over 5 years. *Surg Endosc* 2016; **30**: 5283-5289 [PMID: 27338583 DOI: 10.1007/s00464-016-4877-y]

14 **Ahn JY**, Park HJ, Park YS, Lee JH, Choi KS, Jeong KW, Kim DH, Choi KD, Song HJ, Lee GH, Jung HY. Endoscopic Resection for Undifferentiated-Type Early Gastric Cancer: Immediate Endoscopic Outcomes and Long-Term Survivals. *Dig Dis Sci* 2016; **61**: 1158-1164 [PMID: 26715501 DOI: 10.1007/s10620-015-3988-y]

15 **Inokuchi Y**, Kobayashi M, Kudo K, Yamada H, Inoue S, Nishimura K, Nakayama N, Motohashi O. Outcomes and precautions of endoscopic submucosal dissection for undifferentiated-type early gastric cancer. *Therap Adv Gastroenterol* 2015; **8**: 255-262 [PMID: 26327915 DOI: 10.1177/1756283X15582139]

16 **Tanabe S**, Hirabayashi S, Oda I, Ono H, Nashimoto A, Isobe Y, Miyashiro I, Tsujitani S, Seto Y, Fukagawa T, Nunobe S, Furukawa H, Kodera Y, Kaminishi M, Katai H. Gastric cancer treated by endoscopic submucosal dissection or endoscopic mucosal resection in Japan from 2004 through 2006: JGCA nationwide registry conducted in 2013. *Gastric Cancer* 2017; **20**: 834-842 [PMID: 28205058 DOI: 10.1007/s10120-017-0699-4]

17 **Fujimoto A**, Goto O, Nishizawa T, Ochiai Y, Horii J, Maehata T, Akimoto T, Kinoshita S, Sagara S, Sasaki M, Uraoka T, Yahagi N. Gastric ESD may be useful as accurate staging and decision of future therapeutic strategy. *Endosc Int Open* 2017; **5**: E90-E95 [PMID: 28210705 DOI: 10.1055/s-0042-119392]

18 **Lee IS**, Lee S, Park YS, Gong CS, Yook JH, Kim BS. Applicability of endoscopic submucosal dissection for undifferentiated early gastric cancer: Mixed histology of poorly differentiated adenocarcinoma and signet ring cell carcinoma is a worse predictive factor of nodal metastasis. *Surg Oncol* 2017; **26**: 8-12 [PMID: 28317588 DOI: 10.1016/j.suronc.2016.12.001]

19 **Fujii H**, Ishii E, Tochitani S, Nakaji S, Hirata N, Kusanagi H, Narita M. Lymph node metastasis after endoscopic submucosal dissection of a differentiated gastric cancer confined to the mucosa with an ulcer smaller than 30 mm. *Dig Endosc* 2015; **27**: 159-161 [PMID: 24684669 DOI: 10.1111/den.12261]

20 **Sung CM**, Hsu CM, Hsu JT, Yeh TS, Lin CJ, Chen TC, Su MY, Chiu CT. Predictive factors for lymph node metastasis in early gastric cancer. *World J Gastroenterol* 2010; **16**: 5252-5256 [PMID: 21049560 DOI: 10.3748/wjg.v16.i41.5252]

21 **Kim KJ**, Park SJ, Moon W, Park MI. Analysis of factors related to lymph node metastasis in undifferentiated early gastric cancer. *Turk J Gastroenterol* 2011; **22**: 139-144 [PMID: 21796549 DOI: 10.4318/tjg.2011.0182]

22 **He MJ**, Li QL, Chen WF, Zhou PH, Yao LQ, Xu MD. [Analysis of associated factors of lymph node metastasis in intramucosal early gastric cancer]. *Zhonghua Weichang Waike Zazhi* 2013; **16**: 144-146 [PMID: 23446474]

23 **Ichikawa D**, Komatsu S, Kosuga T, Konishi H, Okamoto K, Shiozaki A, Fujiwara H, Otsuji E. Clinicopathological characteristics of clinical early gastric cancer in the upper-third stomach. *World J Gastroenterol* 2015; **21**: 12851-12856 [PMID: 26668509 DOI: 10.3748/wjg.v21.i45.12851]

24 **Fang C**, Shi J, Sun Q, Gold JS, Xu GF, Liu WJ, Zou XP, Huang Q. Risk factors of lymph node metastasis in early gastric carcinomas diagnosed by WHO criteria in 379 Chinese patients. *J Dig Dis* 2016; **17**: 526-537 [PMID: 27434552 DOI: 10.1111/1751-2980.12385]

25 **Ji T**, Zhou F, Wang J, Zi L. Risk factors for lymph node metastasis of early gastric cancers in patients younger than 40. *Medicine* (Baltimore) 2017; **96**: e7874 [PMID: 28906366 DOI: 10.1097/MD.0000000000007874]

26 **Shin N**, Jeon TY, Kim GH, Park DY. Unveiling lymph node met-

astasis in early gastric cancer. *World J Gastroenterol* 2014; **20**: 5389-5395 [PMID: 24833868 DOI: 10.3748/wjg.v20.i18.5389]

27 **Goto A**, Nishikawa J, Hideura E, Ogawa R, Nagao M, Sasaki S, Kawasato R, Hashimoto S, Okamoto T, Ogihara H, Hamamoto Y, Sakaida I. Lymph node metastasis can be determined by just tumor depth and lymphovascular invasion in early gastric cancer patients after endoscopic submucosal dissection. *Eur J Gastroenterol Hepatol* 2017; **29**: 1346-1350 [PMID: 29084076 DOI: 10.1097/MEG.0000000000000987]

28 **Guo CG**, Chen YJ, Ren H, Zhou H, Shi JF, Yuan XH, Zhao P, Zhao DB, Wang GQ. A nomogram for predicting the likelihood of lymph node metastasis in early gastric signet ring cell carcinoma: A single center retrospective analysis with external validation. *Medicine* (Baltimore) 2016; **95**: e5393 [PMID: 27861374 DOI: 10.1097/MD.0000000000005393]

29 **Li H**, Huo ZB, Chen SB, Li H, Wu DC, Zhai TS, Xiao QH, Wang SX, Zhang LL. Feasibility study on expanded indication for endoscopic submucosal dissection of intramucosal poorly differentiated early gastric cancer. *World J Gastroenterol* 2016; **22**: 6736-6741 [PMID: 27547016 DOI: 10.3748/wjg.v22.i29.6736]

30 **Seto Y**, Yamaguchi H, Shimoyama S, Shimizu N, Aoki F, Kaminishi M. Results of local resection with regional lymphadenectomy for early gastric cancer. *Am J Surg* 2001; **182**: 498-501 [PMID: 11754858 DOI: 10.1016/S0002-9610(01)00747-4]

31 **Abe N**, Mori T, Takeuchi H, Ueki H, Yanagida O, Masaki T, Sugiyama M, Atomi Y. Successful treatment of early stage gastric cancer by laparoscopy-assisted endoscopic full-thickness resection with lymphadenectomy. *Gastrointest Endosc* 2008; **68**: 1220-1224 [PMID: 18547568 DOI: 10.1016/j.gie.2008.02.077]

32 **Cho WY**, Kim YJ, Cho JY, Bok GH, Jin SY, Lee TH, Kim HG, Kim JO, Lee JS. Hybrid natural orifice transluminal endoscopic surgery: endoscopic full-thickness resection of early gastric cancer and laparoscopic regional lymph node dissection--14 human cases. *Endoscopy* 2011; **43**: 134-139 [PMID: 21108175 DOI: 10.1055/s-0030-1255955]

33 **Abe N**, Takeuchi H, Ohki A, Yanagida O, Masaki T, Mori T, Sugiyama M. Long-term outcomes of combination of endoscopic submucosal dissection and laparoscopic lymph node dissection without gastrectomy for early gastric cancer patients who have a potential risk of lymph node metastasis. *Gastrointest Endosc* 2011; **74**: 792-797 [PMID: 21951475 DOI: 10.1016/j.gie.2011.06.006]

P- Reviewer: Aurello P, Aykan NF, Kaplan MA, Senchukova MA, Yarema RR **S- Editor**: Wang JL **L- Editor**: Filipodia **E- Editor**: Tan WW

Published by **Baishideng Publishing Group Inc**
7901 Stoneridge Drive, Suite 501, Pleasanton, CA 94588, USA
Telephone: +1-925-223-8242
Fax: +1-925-223-8243
E-mail: bpgoffice@wjgnet.com
Help Desk: http://www.f6publishing.com/helpdesk
http://www.wjgnet.com