outcomes and perioperative course
of esophageal cancer surgery

ESOPHAGECTOMY

Roy J.J. Verhage

ESOPHAGECTOMY outcomes and perioperative course of esophageal cancer surgery

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ESOPHAGECTOMY

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van chirurgie voor slokdarmkanker

(met een samenvatting in het Nederlands)

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geboren op 9 augustus 1981 te Leiderdorp

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CHAPTER 1

General introduction and thesis outline

GENERAL INTRODUCTION

The incidence of esophageal carcinoma is rapidly increasing. In the Netherlands alone, almost 2000 patients are diagnosed with esophageal cancer every year.[1] Esophageal adenocarcinoma (EAC) and squamous cell carcinoma (ESCC) constitute the two most common histologic subtypes. The incidence of ESCC is highest worldwide.[2] However, in the Western World, EAC rates have increased rapidly over the past thirty years.[3-6] In the Netherlands, EAC has doubled in incidence since the year 2000 and now constitutes roughly 60% of new esophageal cancer diagnoses (figure 1).[1]

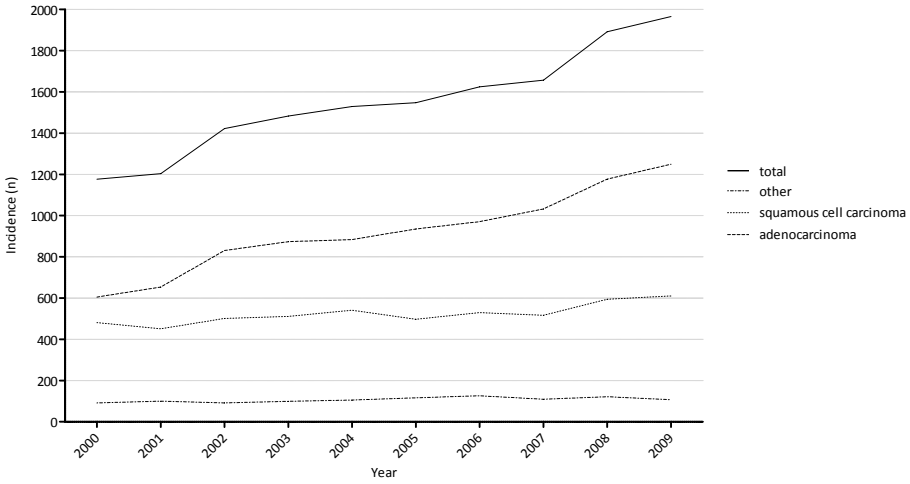

Figure 1 Incidence of esophageal cancer in the Netherlands from 2000 to 2009 by Histological subtype. (Source: Netherlands Cancer Registry, 2011)

Although treatment options have improved more people die from esophageal cancer each year due to the continuously rising incidence. Worldwide, esophageal cancer is the 7th leading cause of cancer death.[7,8] The overall 5 year survival rate of patients diagnosed with esophageal carcinoma is estimated at approximately 15%.[1] The disease typically disseminates early via lymphatic and hematogenous routes. With its aggressive and invasive nature, esophageal cancer has a strong tendency to grow into vital tissues which are in close proximity to the esophagus, such as the pleura, trachea and pericardium. Consequently, the tumor is often diagnosed at an already advanced disease stage with local or systemic spreading of cancer cells. Therapeutic options are then often limited to palliative treatment which causes the unfavorable overall survival rate of esophageal cancer (figure 2).

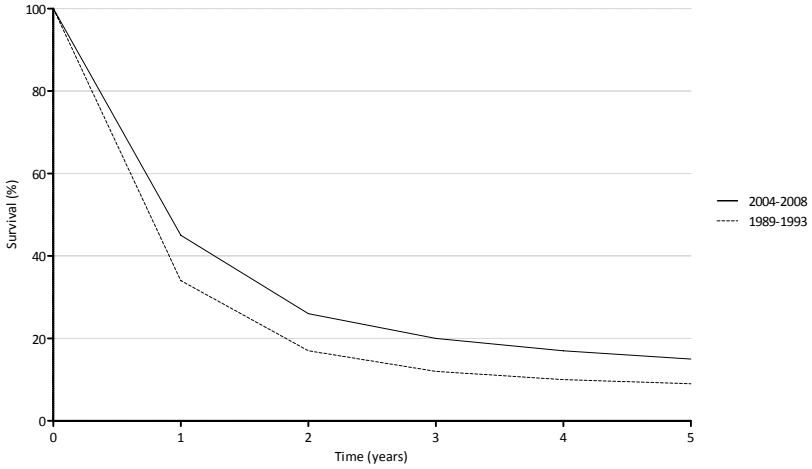

Figure 2 Survival percentage of patients diagnosed with esophageal cancer in the Netherlands between 1989-1993 and 2004-2008. (Source: Netherlands Cancer Registry, 2011)

However, advancements in diagnostics, surgical techniques and the application of neoadjuvant therapies have improved survival rates. Approximately half of the patients with esophageal cancer are eligible for surgery which is, optionally complemented with neoadjuvant chemotherapy, considered the best curative treatment option.[9-11] For patients with surgically treatable disease, studies report 5 year survival rates of up to 40%.[12-14] Neoadjuvant therapies which are currently applied or under investigation include perioperative chemotherapy or preoperative chemoradiotherapy.[12,15-18]

NEOADJUVANT THERAPY

Evidence for the beneficial influence of neoadjuvant chemo(radio)therapy on long term survival is growing.[17] However, the proportion of patients that adequately responds to chemotherapy remains relatively low. Reported response rates are in the range of 40-60%.[12,19] And complete pathologic response is seen in only 10% of patients.[20] Furthermore, chemotherapy is accompanied with risks of serious toxicity. Commonly known side effects of temporary nature include nausea, hair loss and neuropathic pain. More severe chemotherapy related toxicity is less apparent and can develop subclinically, but can cause serious morbidity. This includes cardiac, renal and hematologic toxicity of possibly permanent nature. Chemotherapeutic treatment is not as harmful as it used to be in the past, but it is still not possible to accurately predict which patients will respond to neoadjuvant therapy.

Patients who do not respond to neoadjuvant therapy are exposed to unnecessary toxicity. And whilst treatment is ineffective, surgery is delayed, which decreases the chance of complete oncological resection. Non-responding patients after chemoradiotherapy have

no survival benefit compared to patients who have been treated with surgery alone (i.e. primary esophagectomy without neoadjuvant therapy). Some patients even show worsened survival outcomes.[21] In addition, therapy related toxicity can potentially influence surgical outcomes. A reduced physical condition of already frail patients affects surgical outcome and potentially poses patients at a higher risk of developing postoperative complications. So careful evaluation of (underlying) toxicity and tailored neoadjuvant approaches for the individual patient are necessary when preparing patients for surgery.

SURGERY

Patients with adequate physical condition and potentially curable disease undergo esophagectomy. During an extensive procedure, the esophagus and surrounding lymph nodes are resected (*esophagolymphadenectomy* or *esophagectomy*) together with the proximal part of the stomach and perigastric lymph nodes. To restore continuity of the upper digestive tract, a gastric conduit or neo-esophagus is formed with the remaining part of the stomach. The gastric conduit is pulled up to the level of the neck where an anastomosis is formed with the cervical esophagus.

Esophagolymphadenectomy can be performed through a transhiatal (abdominal) or transthoracic (thoracoabdominal) approach. Both approaches have their pros and cons. Transhiatal esophagectomy (THE) is accompanied by less operative morbidity than transthoracic esophagectomy (TTE).[22] However, THE is limited to dissection of the gastric, truncal and lower mediastinal lymph nodes. Instead, TTE facilitates an en bloc resection which also includes the lymph nodes from the middle and upper mediastinum. This is considered to offer a better chance of oncological clearance.[14] It is argued that TTE is indicated only for tumors of the middle and upper esophagus. However, tumors of the lower esophagus also disseminate to lymph nodes in the upper mediastinum.[23,24] Even early stage tumors (i.e. tumors not yet invading the muscular layer) have a tendency to spread to lymph node stations in the upper mediastinum through the submucosal longitudinal lymphatic drainage system.[24-26] These findings support the use of an extended (transthoracic) resection also for lower esophageal tumors and early stage submucosal tumors.

POSTOPERATIVE MORBIDITY

Besides the choice of surgical approach, operative morbidity forms an major concern in esophageal cancer treatment. Although in-hospital mortality rates have reduced significantly to the range of 1-5% due to centralization of esophageal surgery and improvements in perioperative care, surgery is still accompanied by serious morbidity.[27-31] Morbidity not only affects short term surgical outcome. A correlation has also been found between postoperative morbidity and the interval between surgery and death due to recurrence of disease.[32-34]

To reduce the risk of postoperative complications, a multitude of strategies have been explored. Some are now considered standard of care, many are still under investigation and many more will be developed. Much knowledge has been gained by assessing which patients are at increased risk of developing complications.[35] This enables adequate preoperative patient selection and provides etiological insights. These insights can direct research into developing new techniques that will aid in future prevention of complications. In addition, the paradigm of minimally invasive surgery has also influenced esophageal cancer surgery. The traditional, but highly invasive, concept of esophagectomy is under debate and minimally invasive techniques for esophagectomy (MIE) are evolving rapidly. Evidence that confirms the supposed advantages of MIE over traditionally open esophagectomy, with respect to the reduction of postoperative complications and hospital stay, is gradually growing. However, conclusions are based on available case series or cohort studies.[36-38] Well designed trials which carefully evaluate MIE and open esophagectomy are being performed, but have not been published to date.[39-41]
Regardless of surgical approach or technique, esophagectomy will inevitably remain one the most extreme types of surgery one can undergo. The procedure is associated with high complication rates when compared to other types of surgery. Hence, studies which focus on the prevention of the most prominent complications after esophagectomy are warranted. Frequently encountered and feared complications are anastomotic leakage and pneumonia. Anastomotic leakage occurs in approximately 15% of patients and is associated with long term morbidity and stricture.[42-45] The incidence of pneumonia after esophagectomy is reported in the range of 20-60% and is associated with a significantly increased risk of postoperative mortality.[46,47]

PROGNOSIS

After surgical treatment, it is important to accurately predict long term outcome. Patients request a pragmatic indication of their prognosis tailored to their individual case.[48,49] But customized prognostication in esophageal cancer is a challenge. Many factors play a role in determining the chances of long term survival. The most important and commonly used prognostic indicators are found at histopathological examination of the resected specimen. Tumor specific characteristics, such as tumor cell differentiation and depth of infiltration, are important variables for estimating long term survival.[50] Furthermore, oncological clearance can be determined by carefully assessing whether or not the tumor has been completely resected and whether or not lymphatic tissue is free of tumor cells.[51,52] Focus is now also turning towards more recently discovered molecular factors. Genetic alterations in tumor cells can cause increased expression of certain molecular biomarkers which a play a key role in carcinogenesis.[53] These biomarkers have shown to be correlated with long term survival.[54] Insight into the expression profiles of these markers can aid cancer specialists discussing prognosis more accurately with their patients.

Moreover, these biomarkers are thought to be suitable targets for customized treatment. A disadvantage of traditional chemotherapy is that chemotherapeutic agents are harmful for all proliferating cells, both cancerous and healthy. Patients are in need of therapies that selectively act on tumor cells by aiming at molecular characteristics specific to the tumor. Therefore, research is currently moving towards biomarkers and their therapeutic significance for so-called targeted therapies.[53,55-57] Targeted therapy has the potential to increase the proportion of patients that respond to neoadjuvant treatment. To adequately direct research into targeted therapy it is important to gain insight into the biomarker expression patterns of esophageal cancer. The presence of biomarkers can be assessed by immunohistochemical analysis of resected tumor specimens. This can be achieved with tissue microarray (TMA) technology which facilitates high throughput immunohistochemical evaluation of biomarker expression.[58]

THESIS OUTLINE

The studies presented in this thesis focus on various features of esophageal cancer treatment. These include perioperative morbidity (short term outcomes) and factors that influence prognostication of survival (long term outcomes). The thesis is subdivided into three parts. **PART ONE - Perioperative Morbidity**, investigates strategies to identify and reduce perioperative complications associated with neoadjuvant and surgical treatment of esophageal cancer. **PART TWO - Prognosis**, concentrates on pathological aspects affecting survival in patients with esophageal adenocarcinoma who underwent esophagectomy. **PART THREE - Summary**, summarizes the thesis, provides a general discussion and discusses future perspectives in esophageal cancer research.

PART ONE | PERIOPERATIVE MORBIDITY

Therapy associated morbidity is common in esophageal cancer patients. Morbidity can occur preoperatively during neoadjuvant therapy and postoperatively after esophagectomy. **Chapter 2** reviews differences in postoperative short term outcomes of MIE and open esophagectomy. Prior to surgery patients are often treated with neoadjuvant chemotherapy which is aimed at downstaging of the tumor and improvement of long term survival after surgery. Patients are exposed to toxicity of chemotherapy which can result in serious morbidity and could possibly affect surgical outcome. **Chapter 3** evaluates the incidence of thromboembolic complications attributed to a widely used neoadjuvant chemotherapy regimen as well as their effect on perioperative course.

A feared complication after surgery is anastomotic leakage, which is associated with significant morbidity and mortality. Leakage generally presents within the first week of postoperative recovery. It has been suggested that early anastomotic strength can be

increased with sealants to potentially prevent anastomotic leakage. **Chapter 4** describes an experimental study which evaluates the effect of a fibrin-thrombin coated patch on anastomotic strength of esophagogastric anastomoses in a rat model.

Another major cause of significant morbidity and increased risk of mortality, particularly after thoracic esophagectomy, is postoperative pneumonia. Prolonged single lung ventilation during the thoracic phase of the operation and manipulation of the collapsed lung causes pulmonary damage and is held responsible for an increased risk of developing pneumonia. The response of the innate immunological system functions a marker for the surgical and pulmonary trauma during esophagectomy. The trial presented in **Chapter 5** hypothesizes that the application of continuous positive air pressure on the collapsed lung during single lung ventilation reduces pulmonary trauma and consequently reduces local and systemic immunological response.

For diagnosing pneumonia in mechanically ventilated patients, diagnostic criteria and scoring systems are available. However, after esophagectomy, the condition is frequently diagnosed during postoperative recovery at the hospital ward. The objective of **Chapter 6** is to provide insight into the value of the diagnostic determinants for pneumonia and to develop a straightforward clinical scoring system.

PART TWO | PROGNOSIS

An important predictor of long term survival after esophagectomy is the surgical resection margin. Two different guidelines for defining the circumferential resection margin are used in the literature. Both definitions are evaluated in **Chapter 7** to asses which definition offers the best predictive value for estimating chances of long term survival.

The prognostic significance of molecular tumor characteristics has led to an increasing interest in the therapeutic value of biomarkers that are known to play a key role in carcinogenesis and disease progression. Markers which can be selectively blocked with pharmaceuticals deserve particular attention. **Chapter 8** studies the expression and prognostic significance of two of these biomarkers in EAC, cyclooxygenase-2 (COX-2) and vascular endothelial growth factor (VEGF).

PART THREE | SUMMARY

Chapters 9 and **10** summarize and discuss the studies presented in parts I and II of this thesis.

REFERENCES

1. Comprehensive Cancer Centers: Netherlands Cancer Registry. 2011.
2. 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(14)2137-2150.
3. Bollschweiler E, Wolfgarten E, Gutschow C, Holscher AH. Demographic variations in the rising incidence of esophageal adenocarcinoma in white males. Cancer. 2001;92(3)549-555.
4. Bosetti C, Levi F, Ferlay J, Garavello W, Lucchini F, Bertuccio P, Negri E, La VC. Trends in oesophageal cancer incidence and mortality in Europe. Int J Cancer. 2008;122(5)1118-1129.
5. Botterweck AA, Schouten LJ, Volovics A, Dorant E, van den Brandt PA. Trends in incidence of adenocarcinoma of the oesophagus and gastric cardia in ten European countries. Int J Epidemiol. 2000;29(4)645-654.
6. Cook MB, Chow WH, Devesa SS. Oesophageal cancer incidence in the United States by race, sex, and histologic type, 1977-2005. Br J Cancer. 2009;101(5)855-859.
7. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin. 2011
8. Jemal A, Siegel R, Xu J, Ward E. Cancer statistics, 2010. CA Cancer J Clin. 2010;60(5)277-300.
9. Enzinger PC, Mayer RJ. Esophageal cancer. N Engl J Med. 2003;349(23)2241-2252.
10. Koshy M, Esiashvilli N, Landry JC, Thomas CR, Matthews RH. Multiple management modalities in esophageal cancer: combined modality management approaches. Oncologist. 2004;9(2)147-159.
11. Graham AJ, Shrive FM, Ghali WA, Manns BJ, Grondin SC, Finley RJ, Clifton J. Defining the optimal treatment of locally advanced esophageal cancer: a systematic review and decision analysis. Ann Thorac Surg. 2007;83(4)1257-1264.
12. Cunningham D, Allum WH, Stenning SP, Thompson JN, van de Velde CJ, Nicolson M, Scarffe JH, Lofts FJ, Falk SJ, Iveson TJ, Smith DB, Langley RE, Verma M, Weeden S, Chua YJ, MAGIC Trial Participants. Perioperative chemotherapy versus surgery alone for resectable gastroesophageal cancer. N Engl J Med. 2006;355(1)11-20.
13. van Meerten E, Muller K, Tilanus HW, Siersema PD, Eijkenboom WM, van Dekken H, Tran TC, van der Gaast A. Neoadjuvant concurrent chemoradiation with weekly paclitaxel and carboplatin for patients with oesophageal cancer: a phase II study. Br J Cancer. 2006;94(10)1389-1394.
14. Omloo JM, Lagarde SM, Hulscher JB, Reitsma JB, Fockens P, van Dekken H, ten Kate FJ, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the mid/distal esophagus: five-year survival of a randomized clinical trial. Ann Surg. 2007;246(6)992-1000.
15. Wijnhoven BP, van Lanschot JJ, Tilanus HW, Steyerberg EW, Van der GA. Neoadjuvant chemoradiotherapy for esophageal cancer: a review of meta-analyses. World J Surg. 2009;33(12)2606-2614.
16. van Heijl M, van Lanschot JJ, Koppert LB, van Berge Henegouwen MI, Muller K, Steyerberg EW, van Dekken H, Wijnhoven BP, Tilanus HW, Richel DJ, Busch OR, Bartelsman JF, Koning CC, Offerhaus GJ, van der Gaast A. Neoadjuvant chemoradiation followed by surgery versus surgery alone for patients with adenocarcinoma or squamous cell carcinoma of the esophagus (CROSS). BMC Surg. 2008;8:21.
17. Gebski V, Burmeister B, Smithers BM, Foo K, Zalcberg J, Simes J. Survival benefits from neoadjuvant chemoradiotherapy or chemotherapy in oesophageal carcinoma: a meta-analysis. Lancet Oncol. 2007;8(3)226-234.
18. Sjoquist KM, Burmeister BH, Smithers BM, Zalcberg JR, Simes RJ, Barbour A, Gebski V. Survival after neoadjuvant chemotherapy or chemoradiotherapy for resectable oesophageal carcinoma: an updated meta-analysis. Lancet Oncol. 2011;12(7)681-692.
19. Ngamruengphong S, Sharma VK, Nguyen B, Das A. Assessment of response to neoadjuvant therapy in esophageal cancer: an updated systematic review of diagnostic accuracy of endoscopic ultrasonography and fluorodeoxyglucose positron emission tomography. Dis Esophagus. 2010;23(3)216-231.

20. Merkow RP, Bilimoria KY, McCarter MD, Chow WB, Ko CY, Bentrem DJ. Use of Multimodality Neoadjuvant Therapy for Esophageal Cancer in the United States: Assessment of 987 Hospitals. Ann Surg Oncol. 2011[epub ahead of print]

21. Dittrick GW, Weber JM, Shridhar R, Hoffe S, Melis M, Almhanna K, Barthel J, McLoughlin J, Karl RC, Meredith KL. Pathologic Nonresponders after Neoadjuvant Chemoradiation for Esophageal Cancer Demonstrate no Survival Benefit Compared with Patients Treated with Primary Esophagectomy. Ann Surg Oncol. 2011[epub ahead of print]

22. Hulscher JB, van Sandick JW, de Boer AG, Wijnhoven BP, Tijssen JG, Fockens P, Stalmeier PF, ten Kate FJ, van Dekken H, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the esophagus. N Engl J Med. 2002;347(21)1662-1669.

23. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8)878-886.

24. Tachimori Y, Nagai Y, Kanamori N, Hokamura N, Igaki H. Pattern of lymph node metastases of esophageal squamous cell carcinoma based on the anatomical lymphatic drainage system. Dis Esophagus. 2011;24(1)33-38.

25. Barbour AP, Jones M, Brown I, Gotley DC, Martin I, Thomas J, Clouston A, Smithers BM. Risk stratification for early esophageal adenocarcinoma: analysis of lymphatic spread and prognostic factors. Ann Surg Oncol. 2010;17(9)2494-2502.

26. Grotenhuis BA, van HM, Wijnhoven BP, Henegouwen MI, Biermann K, ten Kate FJ, Busch OR, Dinjens WN, Tilanus HW, van Lanschot JJ. Lymphatic micrometastases in patients with early esophageal adenocarcinoma. J Surg Oncol. 2010;102(7)863-867.

27. Swisher SG, Deford L, Merriman KW, Walsh GL, Smythe R, Vaporicyan A, Ajani JA, Brown T, Komaki R, Roth JA, Putnam JB. Effect of operative volume on morbidity, mortality, and hospital use after esophagectomy for cancer. J Thorac Cardiovasc Surg. 2000;119(6)1126-1132.

28. Whooley BP, Law S, Murthy SC, Alexandrou A, Wong J. Analysis of reduced death and complication rates after esophageal resection. Ann Surg. 2001;233(3)338-344.

29. Wu PC, Posner MC. The role of surgery in the management of oesophageal cancer. Lancet Oncol. 2003;4(8)481-488.

30. Stein HJ, Siewert JR. Improved prognosis of resected esophageal cancer. World J Surg. 2004;28(6)520-525.

31. Wouters MW, Karim-Kos HE, le CS, Wijnhoven BP, Stassen LP, Steup WH, Tilanus HW, Tollenaar RA. Centralization of esophageal cancer surgery: does it improve clinical outcome? Ann Surg Oncol. 2009;16(7)1789-1798.

32. Lagarde SM, de Boer JD, ten Kate FJ, Busch OR, Obertop H, van Lanschot JJ. Postoperative complications after esophagectomy for adenocarcinoma of the esophagus are related to timing of death due to recurrence. Ann Surg. 2008;247(1)71-76.

33. Takeuchi H, Saikawa Y, Oyama T, Ozawa S, Suda K, Wada N, Takahashi T, Nakamura R, Shigematsu N, Ando N, Kitajima M, Kitagawa Y. Factors influencing the long-term survival in patients with esophageal cancer who underwent esophagectomy after chemoradiotherapy. World J Surg. 2010;34(2)277-284.

34. Lerut T, Moons J, Coosemans W, Van RD, De LP, Decaluwe H, Decker G, Nafteux P. Postoperative complications after transthoracic esophagectomy for cancer of the esophagus and gastroesophageal junction are correlated with early cancer recurrence: role of systematic grading of complications using the modified Clavien classification. Ann Surg. 2009;250(5)798-807.

35. Grotenhuis BA, Wijnhoven BP, Grune F, van BJ, Tilanus HW, van Lanschot JJ. Preoperative risk assessment and prevention of complications in patients with esophageal cancer. J Surg Oncol. 2010;101(3)270-278.

36. Verhage RJ, Hazebroek EJ, Boone J, van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64(2)135-146.

37. Biere SS, Cuesta MA, van der Peet DL. Minimally invasive versus open esophagectomy for cancer: a systematic review and meta-analysis. Minerva Chir. 2009;64(2)121-133.

38. Decker G, Coosemans W, De LP, Decaluwe H, Nafteux P, Van RD, Lerut T. Minimally invasive esophagectomy for cancer. Eur J Cardiothorac Surg. 2009;35(1)13-20.

39. Biere SS, Maas KW, Bonavina L, Garcia JR, van Berge Henegouwen MI, Rosman C, Sosef MN, de Lange ES, Bonjer HJ, Cuesta MA, van der Peet DL. Traditional invasive vs. minimally invasive esophagectomy: a multi-center, randomized trial (TIME-trial). BMC Surg. 2011;11:2.

40. van der Sluis PC, Ruurda JP, van der Horst S, Besselink MG, Verhage RJ, Joore JCA, Kroese AC, van Leeuwen MS, Lolkema MP, Reerink O, Steenhagen E, Vleggaar FP, Schipper ME, Siersema PD, ten Kate FJ, Koffijberg H, Borel Rinkes IB, van Hillegersberg R. ROBOT trial: Robot-assisted minimally invasive thoraco-laparoscopic esophagectomy vs. transthoracic esophagectomy. unpublished trial protocol. 2011.

41. Briez N, Piessen G, Bonnetain F, Brigand C, Carrere N, Collet D, Doddoli C, Flamein R, Mabrut JY, Meunier B, Msika S, Perniceni T, Peschaud F, Prudhomme M, Triboulet JP, Mariette C. Open versus laparoscopically-assisted oesophagectomy for cancer: a multicentre randomised controlled phase III trial - the MIRO trial. BMC Cancer. 2011;11:310.

42. Honkoop P, Siersema PD, Tilanus HW, Stassen LP, Hop WC, van BM. Benign anastomotic strictures after transhiatal esophagectomy and cervical esophagogastrostomy: risk factors and management. J Thorac Cardiovasc Surg. 1996;111(6)1141-1146.

43. Sutcliffe RP, Forshaw MJ, Tandon R, Rohatgi A, Strauss DC, Botha AJ, Mason RC. Anastomotic strictures and delayed gastric emptying after esophagectomy: incidence, risk factors and management. Dis Esophagus. 2008;21(8)712-717.

44. van Heijl M, van Wijngaarden AK, Lagarde SM, Busch OR, van Lanschot JJ, van Berge Henegouwen MI. Intrathoracic manifestations of cervical anastomotic leaks after transhiatal and transthoracic oesophagectomy. Br J Surg. 2010;97(5)726-731.

45. van Heijl M, Gooszen JA, Fockens P, Busch OR, van Lanschot JJ, van Berge Henegouwen MI. Risk factors for development of benign cervical strictures after esophagectomy. Ann Surg. 2010;251(6)1064-1069.

46. Atkins BZ, Shah AS, Hutcheson KA, Mangum JH, Pappas TN, Harpole DH, D'Amico TA. Reducing hospital morbidity and mortality following esophagectomy. Ann Thorac Surg. 2004;78(4)1170-1176.

47. Atkins BZ, D'Amico TA. Respiratory complications after esophagectomy. Thorac Surg Clin. 2006;16(1)35-48.

48. Hagerty RG, Butow PN, Ellis PM, Dimitry S, Tattersall MH. Communicating prognosis in cancer care: a systematic review of the literature. Ann Oncol. 2005;16(7)1005-1053.

49. Lagarde SM, Reitsma JB, ten Kate FJ, Busch OR, Obertop H, Zwinderman AH, Moons J, van Lanschot JJ, Lerut T. Predicting individual survival after potentially curative esophagectomy for adenocarcinoma of the esophagus or gastroesophageal junction. Ann Surg. 2008;248(6)1006-1013.

50. Wijnhoven BP, Tran KT, Esterman A, Watson DI, Tilanus HW. An evaluation of prognostic factors and tumor staging of resected carcinoma of the esophagus. Ann Surg. 2007;245(5)717-725.

51. Deeter M, Dorer R, Kuppusamy MK, Koehler RP, Low DE. Assessment of criteria and clinical significance of circumferential resection margins in esophageal cancer. Arch Surg. 2009;144(7)618-624.

52. Greenstein AJ, Litle VR, Swanson SJ, Divino CM, Packer S, Wisnivesky JP. Prognostic significance of the number of lymph node metastases in esophageal cancer. J Am Coll Surg. 2008;206(2)239-246.

53. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell. 2011;144(5)646-674.

54. Lagarde SM, ten Kate FJ, Richel DJ, Offerhaus GJ, van Lanschot JJ. Molecular prognostic factors in adenocarcinoma of the esophagus and gastroesophageal junction. Ann Surg Oncol. 2007;14(2)977-991.

55. Tew WP, Kelsen DP, Ilson DH. Targeted therapies for esophageal cancer. Oncologist. 2005;10(8)590-601.

56. Aklilu M, Ilson DH. Targeted agents and esophageal cancer--the next step? Semin Radiat Oncol. 2007;17(1)62-69.

57. Okines AF, Cunningham D. Multimodality treatment for localized gastro-oesophageal cancer. Ann Oncol. 2010;21(Suppl 7)vii286-vii293.

58. Boone J, van Hillegersberg R, Offerhaus GJ, van Diest PJ, Borel Rinkes IH, ten Kate FJ. Targets for molecular therapy in esophageal squamous cell carcinoma: an immunohistochemical analysis. Dis Esophagus. 2009;22(6)496-504.

PART ONE

PERIOPERATIVE MORBIDITY

CHAPTER 2

Minimally invasive surgery compared to open procedures in esophagectomy for cancer; a systematic review of the literature

Minerva Chirurgica 2009;64(2):135-46

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ABSTRACT

The impact of minimally invasive esophagectomy (MIE) on short term perioperative outcomes as opposed to open transhiatal esophagectomy (THE) and transthoracic esophagectomy (TTE) is still under debate. This systematic review attempts to answer whether minimally invasive surgery has improved short term outcomes compared to conventional open surgery in the management of esophageal cancer. A systematic literature search was performed using synonyms for esophagectomy, cancer and minimally invasive surgery. Ten case-controlled studies were retrieved and one systematic review. Data collection was grouped by surgical approach. Overall MIE data shows decreased blood loss (577 mL for conventional open surgery versus 312 mL for MIE) and reduction of hospital and ICU stay (open 19.6 days versus MIE 14.9 days, and open 7.6 days versus MIE 4.5 days respectively). Total complication rates are 60.4% for open esophagectomy and 43.8% for MIE. Pulmonary complications occur in 22.9% and 15.1% of respective procedures. Mean lymph node retrieval was higher in MIE (open 20.2 versus MIE 23.8). This review confirms the feasibility and safeness of minimally invasive surgery for esophageal cancer. Moreover, the retrieved studies collectively point towards improved short term outcomes after MIE. These results should be confirmed in randomized controlled trials.

INTRODUCTION

Surgical resection remains the only available curative option for patients with esophageal cancer. Surgical treatment has long been confined to open procedures using transthoracic (TTE) or transhiatal (THE) approaches. These types of esophagectomy are accompanied by relatively high incidences of post-operative morbidity and mortality. Five year survival following curative resection varies between 20-40%.[1-5]

Particularly transthoracic surgery is associated with high complication rates. Prolonged single lung ventilation, manipulation of the desufflated lung and extensive dissection of the mediastinum are thought to be responsible for a high incidence of respiratory complications after TTE. In turn, pneumonia and respiratory failure after esophagectomy are associated with an increased risk for mortality of up to 20%.[6]

In the challenge to improve postoperative outcomes, minimally invasive techniques for esophagectomy have been developed, making an effort to decrease invasiveness without compromising the extent of dissection and consequent survival. The first series of 12 patients who underwent thoracoscopic mobilization of the esophagus was published in 1993 by Collard *et al.*[7] In 1995 the Brazilian group of DePaula reported 48 cases of transhiatal laparoscopic esophagectomy (LSE).[8] Since that time thoracoscopic and laparoscopic surgery for the management of esophageal cancer has been refined by surgeons over the world of which results have been published in many case series. The largest of these series up to date has been reported by Luketich, who described 222 patients submitted to thoracolaparoscopic esophagectomy (TLSE).[9]

Although minimally invasive esophagectomy (MIE) has been under investigation for over a decade and experience with different techniques has significantly increased, the surgical techniques remain under debate. The safety and efficacy of minimally invasive resection are questioned and there is no consensus regarding neither techniques nor approaches. This systematic review attempts to answer whether minimally invasive surgery has improved short term outcomes compared to conventional open surgery in the management of esophageal cancer. It will provide an overview of the current relevant literature assessing validity and reliability with a specific focus on perioperative data, overall morbidity, pulmonary complications, mortality and lymph node harvest.

METHODS

SEARCH STRATEGY AND SELECTION

A systematic literature search was conducted using Pubmed, Embase and the Cochrane library using synonyms for esophagectomy, cancer and minimally invasive surgery (table 1).[10-12] Doubles were retrieved using the online reference software RefWorks.[13] Articles published in 2000 or later were selected and screened using titles and abstracts.

Table 1 Systematic search strategy

Domain	AND	Determinant
("esophagectomy" OR "oesophagectomy" OR "esophagus resection" OR "oesophagus resection" OR "esophageal resection" OR "oesophageal resection" OR "esophagogastrectomy" OR "oesophagogastrectomy") AND (cancer OR carcinoma OR Malignancy)		(laparoscop* OR thoracoscop* OR "minimally invasive" OR "minimally-invasive" OR "robot assisted" OR "robot-assisted" OR "robotically assisted" OR "robotically-assisted" OR "robotic assisted" OR "robotic-assisted" OR "robot surgery")
Pubmed (239)	Embase (300)	Cochrane (6)
• No limits • Title / Abstract	• Explosion search • 1995-.. • articles with abstract • Embase only	• Title / Abstract / Keywords • All of the Cochrane Library • 1995-..
www.pubmed.com	www.embase.com	www.thecochranelibrary.com

The following exclusion criteria were used: articles reporting on esophagectomy for benign lesions, leiomyoma or as palliative treatment. Also excluded were reports on endoscopic therapy, quality of life studies, papers or overview articles, technique descriptions, staging studies, case series, case reports and non-systematic reviews.

Furthermore, studies describing delayed surgery (i.e. gastric conditioning followed by esophagectomy several days later) and reports solely on gastric conditioning were excluded. Only articles in English were selected. When authors described sequential or overlapping results using (partly) the same population in different reports, only the most recent publication was included to avoid repeated analysis of cases.

The retrieved articles were assessed for the types of interventions, number of patients, study design, retrospective or prospective nature, baseline comparability of patients, comparability of American Society of Anesthesiologists (ASA) classification and the reported perioperative data. The following outcomes were indexed: operating time, estimated operative blood loss, hospital and Intensive Care Unit (ICU) stay, lymph node harvest, pulmonary complication rates, total complication rates and perioperative mortality. The latter was defined as death within 30 days or in-hospital death following surgery. Levels of evidence were assigned according to Harbour and Miller.[14] Data on the reported outcomes were collected and presented in tables. Where possible, weighted means were calculated using the statistical software SPSS version 15.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

After removing doubles the search identified 377 articles (see figure 1 for flowchart). Selection through screening of titles and abstracts deemed 19 articles eligible. An

additional search for related articles yielded two more studies, of which one is yet to be published.[15,16] After the search was further restricted to articles in English, 15 studies remained for full-text evaluation. Four additional articles, reporting results from overlapping data with three other studies, were excluded to avoid double analysis.[17-20]

Between 2000 and 2009 one article systematically reviewed minimally invasive surgery for esophagectomy and gastrectomy in general without a specific focus on the comparison with open surgery.[21] Ten selected articles compared open esophagectomy to some form of MIE. These were all case-controlled studies with varying levels of evidence (table 2).[15,16,22-29] No randomized controlled trials were found. Four studies prospectively collected their data on MIE, and compared it to retrospectively gathered data from open series or matched controls. The remaining six reports were either of retrospective nature or did not provide insight in the type of data collection. Total patient numbers varied from 22 to 446 (mean 111, median 58), and the total number of patients who underwent some form of MIE ranged from 9 to 33 (mean 51, median 20). Overall, 494 patients underwent open transhiatal or transthoracic surgery, versus 616 patients receiving some type of MIE. The studies were evaluated through critical appraisal and are accordingly presented in table 2. Since the authors used different types of minimally invasive approaches for esophagectomy, the studies are sorted by techniques for interstudy analysis. Two studies compared THE with LSE.[16,22] Four studies analyzed combined transthoracic and transhiatal surgery to compare with TLSE.[15,23-25] The remaining four studies used varying approaches which could not be analysed within a specific subgroup and are therefore only used for an overall analysis of open versus minimally invasive surgery.[26-29]

Figure 1 Flowchart of literature search

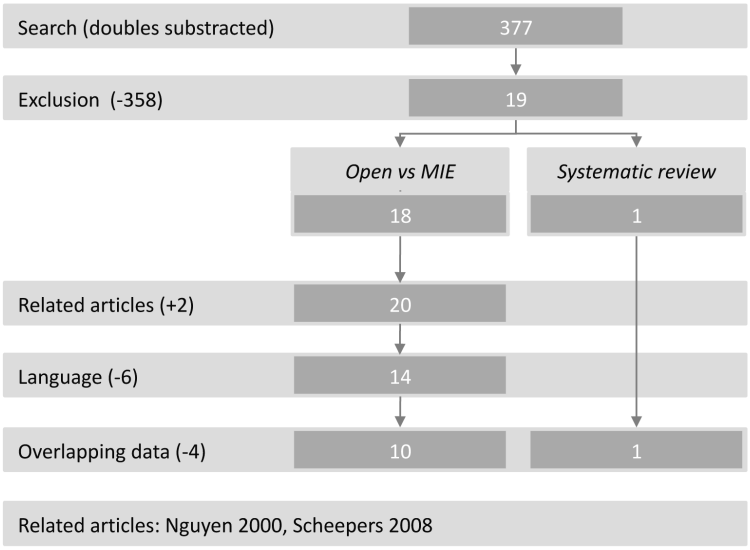

Table 2 Characteristics of retrieved articles

							outcomes								
	interventions	patients (n)	design	perspective	baseline comparability	ASA comparability	operating time	EBL	ICU stay	hospital stay	lymfnode retrieval	pulmonary complications	complications	peri-operative mortality	LOE[14]
Scheepers 2009[16]	THE LSE	50 50	case control	R&P	+	+	+	+	+	+	+	+	+	+	2+
Bernabe 2005[22]	THE LSE	14 17	case control	R	+	+	+	+	-	+	+	-	-	-	2+
Fabian 2008[23]	TTE or THE TLSE	43 22	case control	R&P	+	na	+	+	-	+	+	+	+	+	2+
Smithers 2007[24]	TTE TSE TLSE	114 309 23	case control	P	+/-	+	+	+	+	+	+	+	+	+	2-
Kunisaki 2004[25]	TTE TLSE	30 15	case control	na	+	na	+	+	+	+	+	+	+	+	2+
Nguyen 2000[15]	THE TTE TLSE	20 16 18	case control	R&P	+/-	+	+	+	+	+	+	+	+	+	2-
Benzoni 2008[29]	LSE TTE+LSE	9 13	case control	na	+/-	+/-	+	-	+	+	+	+	+	+	2--
Kitagawa 2008[26]	TTE TTE+LSE	20 16	case control	na	+/-	na	+	+	+	-	-	-	+	+	2--
Braghetto 2006[27]	TTE or THE TSE or LSE	119 47	case control	na	-	na	-	-	-	-	-	+	+	+	2-
Osugi 2003[28]	TTE TSE	72 77	case control	R	+	na	+	+	-	-	+	+	+	+	2+
Gemmill 2007[21]	MIE	1398	syst. review				+	+	-	+	+	+	+	+	1-

TTE - transthoracic esophagectomy; THE - transhiatal esophagectomy; LSE - laparoscopic esophagectomy; TLSE - thoraco-laparoscopic esophagectomy; TSE - thoracoscopic esophagectomy; MIE - minimally invasive esophagectomy; R - retrospective; P - prospective; LOE - level of evidence; n - number; na - not available.

OPEN TRANSHIATAL VERSUS LAPAROSCOPIC ESOPHAGECTOMY

Laparoscopic procedures were described in four case-controlled studies. Scheepers *et al*. and Bernabe *et al*. compared a fully laparoscopic procedure with the open transhiatal technique.[16,22] One study compared LSE with a hybrid procedure (i.e. combined open transthoracic followed by laparoscopy).[29] The fourth study, conducted by Kitagawa *et al*., compared an open transthoracic group with an open transthoracic-laparoscopic group.[26] The latter two studies are difficult to compare, because of the surgically different strategies. Therefore, these were only included in the overall open versus MIE analysis. It must be noted that Scheepers *et al*. only provided medians for the outcomes operating time, blood loss, hospital and ICU stay, and the number of lymph nodes. These medians could not be included in the overall analysis of LSE versus THE, for which calculation of the weighted means is necessary. Data are presented in table 3.

Table 3 Average short term post-operative outcomes for open transhiatal esophagectomy versus laparoscopic esophagectomy

		Scheepers 2009[16]			Bernabe 2005[22]		
		THE	LSE	*p*	THE	LSE	*p*
	n	50	50		14	17	
Operating time	(min)	280*	300*	*0.110*	388	336	*0.099*
EBL	(ml)	900*	500*	*0.000*	543	331	*0.011*
ICU stay	(days)	3*	1*	*0.000*			
Hospital stay	(days)	16*	13*	*0.001*	11.6	9.1	*0.037*
Lymph nodes	(n)	11*	14*	*0.754*	9.8	8.7	*na*
Pulmonary complications	(%)	26	18	*na*			
Complications	(%)	66	42	*na*			
Peri-operative mortality	(%)	2	0	*na*	0	0	*ns*

* medians. EBL - estimated blood loss; ICU - intensive care unit; na - not available; ns - not significant; THE - transhiatal esophagectomy; LSE - laparoscopic esophagectomy; n - number; ml - millilitres; min - minutes.

Operating time

Scheepers *et al*. found no significant difference in operating time between THE and LSE; 280 minutes and 300 minutes respectively (P=0.11). Bernabe found an average decrease of operative time of 52 minutes without statistical significance (P=0.099).

However, comparison of their last 6 laparoscopic patients with the open control group, did show a nearly significant difference in operating time (THE 388, LSE 311; P=0.07).

Estimated blood loss

Both studies showed significant differences in blood loss between the open and laparoscopic procedure. Bernabe reported an average decrease of 212 mL (P=0.011). The median difference described by Scheepers was 400 mL (P=0.000).

Length of stay

Hospital stay was reported by both studies, whereas ICU stay was only provided by Scheepers *et al*. Length of stay (LOS) was lower in the laparoscopic groups of Scheepers and Bernabe (16 versus 13; P=0.001, and 11.6 versus 9.1; P=0.037 respectively). Scheepers also found that their patients spent significantly less days in the ICU ward after LSE than THE (3 versus 1 day; P=0.000).

Morbidity

Perioperative complication rates were not reported by Bernabe *et al*. Scheepers did not statistically evaluate their observed differences. Therefore, no *P values* can be provided. Furthermore, only Scheepers *et al*. showed the total complication rates. They found less complications after LSE; 42% versus 62% in the open group. Considering pulmonary complications, the open group showed higher rates than the laparoscopic group; 26% versus 18% respectively.

Mortality

Scheepers *et al*. had no perioperative mortality in the LSE group and 2% mortality in the open group. Bernabe experienced no perioperative death in both study groups.

Lymph node retrieval

For the harvest of lymph nodes no differences were found between open and laparoscopic surgery. Bernabe *et al*. presented an average of 9.8 lymph nodes for transhiatal surgery and 8.7 nodes in the minimally invasive procedure. Scheepers' median number was slightly higher in the scopic group (14) when compared to the open group (11), but no statistical significance was found (P=0.754).

OPEN VERSUS THORACO-LAPAROSCOPIC ESOPHAGECTOMY

Four articles compared a fully MIE, the thoracolaparoscopic approach, with open surgery. In 2000 Nguyen compared 18 TLSE cases with a historical retrospective cohort of 20 THEs and 16 TTEs.[15] Kunisaki *et al*. presented results from 15 patients operated on with hand assisted laparoscopy (HALS), aided by the robotic camera-system AESOP, followed by conventional thoracoscopy for mediastinal dissection of the esophagus.[25] A partly historical group of 30 patients who underwent conventional open TTE formed the control group. Smithers *et al*. studied a total of 446 prospectively collected esophagectomies.[24] A series of 114 patients underwent an open TTE and formed the control group. 309 patients received thoracoscopic-assisted therapy (i.e. thoracoscopic mobilization followed by laparotomy) and 23 patients had a total MIE through thoracolaparoscopy. The latter group was only implemented between 1998 and 2000, after which the procedure was deemed little beneficial over the thoracoscopic-assisted approach based on short- and medium-term follow-up data. A recent publication by

Fabian *et al*. presents a first year experience of MIE and a comparison with a retrospective control group.[23] All open esophagectomies, being transthoracic or transhiatal, were bundled into one control group consisting of 65 retrospective cases and compared with 22 patients who underwent thoracoscopic and/or laparoscopic esophagectomy. It should be regarded that Smithers *et al*. presented medians for the outcomes operating time, blood loss, hospital and ICU stay, and the number of lymph nodes. These medians could not be included in the overall analysis of TTE versus TLSE for which calculation of the weighted means is necessary. The collected outcome variables are presented in table 4.

Operating time

Kunisaki, Smithers and Fabian found the total time of surgery to be significantly increased when using the thoraco-laparoscopic approach. On the contrary, Nguyen reported a significant decrease of operating time when compared to open transthoracic surgery (364 minutes versus 437 minutes; $P<0.05$). Combined data from three studies yield an average operating time of 373 minutes for TTE, compared to 401 minutes for TLSE.

Estimated blood loss

Significant consensus is reached in all four studies considering operative blood loss. Values vary from 356 to 1046 mL for TTE and 178 to 448 mL for thoracolaparoscopic surgery. Combined weighted data over a total of 144 patients (excluding data from Smithers) give an average difference of 291 mL between open and fully scopic surgery (TTE 588 mL, TLSE 291 mL).

Length of stay

Hospital stay tended to be reduced in all studies when using the thoracolaparoscopic approach. Nguyen and Smithers found a significant decrease of LOS and Kunisaki and Fabian report a trend towards shorter admission times. Weighted average LOS for TTE is 20.5 days. After TLSE the average LOS is 15.6 days, reducing hospital stay by almost 5 days. Time spent in the ICU was significantly shorter in Nguyen's study (9.9 days for TTE and 6.1 days for TLSE; $P<0.05$). Smithers also presents shorter ICU stays (0.79 days versus 0.96 days; $P=0.030$). However, it is unclear whether these data only include post-operative stays, omitting readmittances to the ICU ward for delayed complications such as respiratory insufficiency. Kunisaki *et al*. did not find a contrast in ICU stay, reporting 6.9 days for TTE and 6.8 days for thoracolaparoscopic surgery. Fabian *et al*. did not provide insight into data concerning their patients' ICU admittance. Joined results from Nguyen and Kunisaki give a mean ICU stay of 7.9 days after open surgery and 6.4 days after TLSE.

Table 4 Average short term post-operative outcomes for open transthoracic esophagectomy versus thoraco-laparoscopic esophagectomy

		Nguyen 2000[15]			Kunisaki 2004[25]			Smithers 2007[24]			Fabian 2008[23]			weighted means	
		TTE	TLSE	*p*	TTE	TLSE	*p*	TTE	TLSE	*p*	TTE or THE	TLSE	*p*	TTE	TLSE
		16	18		30	15		114	23		43	22		89	55
Operating time total	(min)	437	364	<0.05	487.8	544.4	0.0044	300*	330*	0.01	270	333	0.01	373†	401†
Operating time thorax	(min)				257.9	301.9	0.0001	120*	90*	0.01					
EBL	(ml)	1046	297	<0.05	674.7	447.9	0.042	600*	300*	0.017	356	178	<0.0001	588†	291†
ICU stay	(days)	9.9	6.1	<0.05	6.9	6.8	0.989	0.96*	0.79*	0.030				7.9†	6.4†
Hospital stay	(days)	23	11.3	<0.05	32.7	29.6	0.530	14*	11*	0.030	11	9.5	0.3	20.5†	15.6†
Lymph nodes	(n)	6.3	10.8	ns	26.6	24.5	0.531	16*	17*	ns	8	15	na	14.0†	16.2†
Pulmonary complications	(%)	19	11	na	3.3	0	0.475	27.8	30	ns	42	14	na	26.5	15.3
Complications	(%)	50	39	na	72	70	ns	66.7	61	ns	72	59	0.78	67.3	57.1
Peri-operative mortality	(%)	0	0	1.000	0	0	1.000	2.6	0	ns	9.8	4.5	0.45	3.8	1.3

* medians

† weighted means are calculated without data from Smithers et al. because the means were not reported by the authors

EBL - estimated blood loss; ICU - intensive care unit; na - not available; ns - not significant; TTE - transthoracic esophagectomy; THE - transhiatal esophagectomy; TLSE - thoraco-laparoscopic esophagectomy; n - number; ml - millilitres; min - minutes.

Morbidity

As is to be anticipated with combined thoracic and abdominal surgery, complication rates vary from 50% to 72% for open surgery and 39% to 70% for MIE. Kunisaki, Smithers and Fabian did not find statistical differences between the minimally invasive and control groups. Nguyen experienced fewer complications after MIE (39% versus 50%) but did not report statistical analysis of these figures. Overall analysis of these four studies shows a 67.3% complication rate for TTE and a 57.1% rate for TLSE.

Pulmonary complications are renown after long transthoracic procedures as is the case with esophagectomy. The overall average of these four studies is 26.5% for TTE and 15.3% for TLSE.

Mortality

Postoperative mortality rates are provided by all four authors. Both Nguyen and Kunisaki experienced no deaths within 30 days of surgery in both their study groups. Smithers reported 3 deaths (2.6%) in the open group, compared to 0 deaths in the scopic group. Fabian *et al*. recorded 5 cases of postoperative mortality (TTE 9.8%, TLE 4.5%; P=0.45). Combined data accounts for a 3.8% mortality rate after open transthoracic surgery and a 1.3% mortality rate for thoracolaparoscopic surgery.

Lymph node retrieval

None of the authors reported significant improvements in the harvest of lymph nodes when using the thoracolaparoscopic approach. Lymph node retrieval varies greatly among studies, ranging from 8 to 26.6 lymph nodes for open surgery compared to 10.8 to 24.5 lymph nodes for TLSE. Averaged results give a weighted mean of 14 lymph nodes in open surgery versus 16.2 lymph nodes for TLSE.

OVERALL COMPARISON OF OPEN VERSUS MIE

For an overall analysis of open versus MIE data from 10 studies are combined and weighted means have been calculated based on the number of cases in the different study groups and are presented in table 5. Open surgery, including both transhiatal and transthoracic surgery, was performed in 494 patients. MIE, including all minimally invasive strategies used in the different studies, was carried out in a total of 616 cases. Weighted means could only be calculated where means were provided by the authors. Therefore, data from Scheepers and Smithers were excluded because of the use of medians for the following variables; operating time, EBL, ICU stay, hospital stay and lymph node retrieval. The calculated means show a slight increase of operating time when comparing open and minimally invasive surgery, 324 and 334 minutes respectively. A noticeable decrease of blood loss is achieved when minimally invasive procedures are applied (open 577 mL, MIE 312 mL). Both ICU stay and hospital stay are also lower in the MIE groups. The average harvest of lymph nodes appears to be slightly different between the two groups

(open 20.2, MIE 23.8). As morbidity is concerned, a trend can be seen towards lower complication rates for MIE. The mean complication rate for open surgery is 60.4% compared to 43.8% for MIE. Pulmonary complication rates were calculated to be 22.9% and 15.1% respectively. Postoperative mortality also seems to be lower after minimally invasive approaches (1.9%) when compared to open surgery (4.5%). In a systematic review published in 2007, Gemmill *et al.* calculated weighted means on 1398 patients from 21 case series and 2 case-matched studies on MIE.[21] These data are included in table 5.

Table 5 Average short term post-operative outcomes for open esophagectomy versus minimally invasive esophagectomy irrespective of applied technique

		Gemmill 200721 weighted means	Combined data from retrieved articles weighted means	
		MIE (n=1398)	open (n=494)	MIE (n=616)
Operating time*	(min)	281	324	334
EBL*	(ml)	316	577	312
ICU stay*	(days)		7.6	4.5
Hospital stay*	(days)	11.0	19.6	14.9
Lymph nodes*	(n)	17.6	20.2	23.8
Pulmonary complications	(%)	13.2	22.9	15.1
Complications	(%)	46.2	60.4	43.8
Peri-operative mortality	(%)	2.3	4.5	1.9

* weighted means are calculated without data from Smithers et al. and Scheepers et al. because the means were not reported by the authors
EBL - estimated blood loss; ICU - intensive care unit; open - transthoracic and transhiatal esophagectomy; MIE - minimally invasive esophagectomy; n - number; ml - millilitres; min - minutes.

DISCUSSION

This review collectively compares the available data on minimally invasive to open esophagectomy concerning the risks associated with these procedures. Up to date many case series have been published and authors have already concluded that MIE is feasible and safe.[9,30] However, sound comparative studies are scarce and no randomized controlled studies have been published so far. The systematic search strategy retrieved a large number of results, but eligible studies are all case-controlled and consequently of various levels of evidence. For a reliable comparison of combined data a presentation of weighted means is necessary. Statistical analysis of these means is however not deemed appropriate, because of the case-controlled designs and a limited insight in the exact population characteristics. Nonetheless, this review presents the best available evidence in the present literature regarding minimally invasive surgery for esophageal cancer.

The retrieved studies collectively point towards improved short term outcomes after MIE. Most studies show an increase in operative time when performing a minimally invasive procedure. However, all studies show that MIE is associated with a decrease of operative blood loss compared to open esophagectomy regardless of the preferred technique.
Overall, most studies show a shorter hospital stay in their minimally invasive groups. The weighted mean of LOS is decreased by approximately 4 days. As far as ICU stay is concerned, an evident decrease of 3 days is observed. The reported data on ICU stay, however, should be treated with caution. These data can easily be biased due to changing hospital policies towards ICU admittance and timing of extubation. On the other hand it should not go unnoticed that Scheepers, Bernabe, Nguyen and Smithers all reported significant decreases of both hospital and ICU stay.
The reports on morbidity and mortality are in favor of minimally invasive techniques. Total complication rates are lower in most studies and comparable in some. Though, it must be noted that studies are inconsistent in providing insight in the composition of their complication rates. The definitions of "pulmonary complications" vary among studies, including pneumonia, but also atelectasis, pneumothorax and pulmonary embolisms. Still, calculation of means resulted in an 11% difference between TTE and TLSE. Particularly in transthoracic surgery, the objective of minimally invasive procedures is to reduce the number of adverse pulmonary events. Although no significant reduction in postoperative death was reported, it is plausible that a reduced pulmonary complication rate has had a noticeable effect on the observed mortality rates (3.8% for TTE versus 1.3% for TLSE).
In the transthoracic studies the harvest rates vary from 6 to 27 lymph nodes in open surgery and 11 to 25 lymph nodes in TLSE. Weighted means show a difference of 2 lymph nodes in favor of the thoracolaparoscopic procedure. However, Nguyen and Fabian are not clear on the extent of their lymphadenectomy. Although both advocate better views with scopic surgery and therefore a higher node harvest, their lymph node retrieval is relatively low suggesting limited mediastinal lymphadenectomy. Smithers' two-field procedure yielded a higher number of lymph nodes. The Japanese group of Kunisaki chose to perform a three-field lymphadenectomy harvesting the upper mediastinal and cervical lymph nodes. Such extensive dissection resulted in higher collections of approximately 25 nodes for both open and MIE. This strategy might be responsible for their longer operative times.
The extent of lymphadenectomy remains a controversial issue. It is argued that extended lymphadenectomy reaching in the upper mediastinum results in lower recurrence rates and benefits pathological staging. No studies have yet reported significant improvement of survival for three-field lymphadenectomy, but the largest randomized trial comparing limited transhiatal resection versus extended transthoracic resection by Hulscher *et al.* showed a trend towards better 5-year survival for TTE suggesting a beneficial effect of extensive mediastinal dissection.[31] This benefit was specifically noticed in a subgroup of patients with a limited number (1-8) of positive lymph nodes.[5] In 1998 Nishihira conducted

a randomized trial comparing two-field with three-field lymphadenectomy in TTE, also reporting improved 5-year survival; 66.2% and 48.0% respectively.[32] Pulmonary complication rates were similar between groups (19% and 17%). Yet, the extended lymphadenectomy group experienced significantly higher rates for tracheostomy (53% versus 10%) and phrenic nerve palsy (13% versus 0%). With respect to the importance and complexity of extended mediastinal lymphadenectomy, robotic techniques have been developed in order to further improve MIE. Short and midterm results of such techniques show promising results, enabling a median lymph node harvest of 29.[33]

Although no consensus has yet been reached regarding the optimal strategy of esophageal resection, a considerable amount of surgeons selects minimally invasive surgery for esophageal cancer. An international survey showed that already 25% of surgeons routinely apply some form of MIE.[34]

This review confirms the feasibility and safeness of minimally invasive surgery for esophageal cancer. Moreover, it points out that MIE shows a tendency towards better short term outcomes when compared to conventional open surgery. As the presented data in this review is based on case-controlled studies, these results should ideally be confirmed in well conducted randomized controlled trials.

REFERENCES

1. Sant M, Aareleid T, Berrino F, Bielska Lasota M, Carli PM, Faivre J, Grosclaude P, Hédelin G, Matsuda T, Møller H, Möller T, Verdecchia A, Capocaccia R, Gatta G, Micheli A, Santaquilani M, Roazzi P, Lisi D; EUROCARE Working Group. EUROCARE-3: survival of cancer patients diagnosed 1990-94--results and commentary. Ann Oncol. 2003;14(Suppl 5):v61-118.
2. Rudiger Siewert J, Feith M, Werner M, Stein HJ. Adenocarcinoma of the esophagogastric junction: results of surgical therapy based on anatomical/topographic classification in 1,002 consecutive patients. Ann Surg. 2000;232:353-61.
3. Lerut T, Nafteux P, Moons J, Coosemans W, Decker G, De Leyn P, Van Raemdonck D, Ectors N. Three-field lymphadenectomy for carcinoma of the esophagus and gastroesophageal junction in 174 R0 resections: impact on staging, diseasefree survival, and outcome: a plea for adaptation of TNM classification in upper-half esophageal carcinoma. Ann Surg. 2004;240:962-72.
4. Lagarde SM, Reitsma JB, Ten Kate FJ, Busch OR, Obertop H, Zwinderman AH, Moons J, van Lanschot JJ, Lerut T. Predicting individual survival after potentially curative esophagectomy for adenocarcinoma of the esophagus or gastroesophageal junction. Ann Surg. 2008;248:1006-13.
5. Omloo JM, Lagarde SM, Hulscher JB, Reitsma JB, Fockens P, van Dekken H, Ten Kate FJ, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the mid/distal esophagus: five-year survival of a randomized clinical trial. Ann Surg. 2007;246:992-1000.
6. Atkins BZ, Shah AS, Hutcheson KA, Mangum JH, Pappas TN, Harpole DH Jr, D'Amico TA. Reducing hospital morbidity and mortality following esophagectomy. Ann Thorac Surg. 2004;78:1170-6.
7. Collard JM, Lengele B, Otte JB, Kestens PJ. En bloc and standard esophagectomies by thoracoscopy. Ann Thorac Surg. 1993;56:675-9.
8. DePaula AL, Hashiba K, Ferreira EA, de Paula RA, Grecco E. Laparoscopic transhiatal esophagectomy with esophagogastroplasty. Surg Laparosc Endosc. 1995;5:1-5.
9. Luketich JD, Alvelo-Rivera M, Buenaventura PO, Christie NA, McCaughan JS, Litle VR, Schauer PR, Close JM, Fernando HC. Minimally invasive esophagectomy: outcomes in 222 patients. Ann Surg. 2003;238(4):486-94.
10. Pubmed. Homepage [Internet]. Available from www.pubmed.com [cited 2009, Feb 18].
11. Embase. Homepage [Internet]. Available from www.embase.com [cited 2009, Feb 18].
12. The Cochrane Library. Homepage [Internet]. www.thecochranelibrary. com [cited 2009, Feb 18].
13. RefWorks. Homepage [Internet]. www.refworks.com [cited 2009, Feb 18].
14. Harbour R, Miller J. A new system for grading recommendations in evidence based guidelines. BMJ. 2001;11;323:334-6.
15. Nguyen NT, Follette DM, Wolfe BM, Schneider PD, Roberts P, Goodnight JE Jr. Comparison of minimally invasive esophagectomy with transthoracic and transhiatal esophagectomy. Arch Surg. 2000;135:920-5.
16. Scheepers JJG, Veenhof AAFA, van der Peet DL, van Groeningen C, Mulder Ch, Meijer S et al. Laparoscopic versus open transhiatal resection for malignancies of the distal esophagus: a comparison of two cohorts. 2009; unpublished material.
17. Van den Broek WT, Makay O, Berends FJ, Yuan JZ, Houdijk AP, Meijer S, Cuesta MA. Laparoscopically assisted transhiatal resection for malignancies of the distal esophagus. Surg Endosc. 2004;18:812-7.
18. Taguchi S, Osugi H, Higashino M, Tokuhara T, Takada N, Takemura M, Lee S, Kinoshita H. Comparison of three-field esophagectomy for esophageal cancer incorporating open or thoracoscopic thoracotomy. Surg Endosc. 2003;17:1445-50.
19. Bresadola V, Terrosu G, Cojutti A, Benzoni E, Baracchini E, Bresadola F. Laparoscopic versus open gastroplasty in esophagectomy for esophageal cancer: a comparative study. Surg Laparosc Endosc Percutan Tech. 2006;16:63-7.

20 Benzoni E, Terrosu G, Bresadola V, Uzzau A, Intini S, Noce L, Cedolini C, Bresadola F, De Anna D. A comparative study of the transhiatal laparoscopic approach versus laparoscopic gastric mobilisation and right open transthoracic esophagectomy for esophageal cancer management. J Gastrointestin Liver Dis. 2007;16:395-401.

21. Gemmill EH, McCulloch P. Systematic review of minimally invasive resection for gastro-oesophageal cancer. Br J Surg. 2007;94:1461-7.

22. Bernabe KQ, Bolton JS, Richardson WS. Laparoscopic hand-assisted versus open transhiatal esophagectomy: a case-control study. Surg Endosc. 2005;19:334-7.

23. Fabian T, Martin JT, McKelvey AA, Federico JA. Minimally invasive esophagectomy: a teaching hospital's first year experience. Dis Esophagus. 2008;21:220-5.

24. Smithers BM, Gotley DC, Martin I, Thomas JM. Comparison of the outcomes between open and minimally invasive esophagectomy. Ann Surg. 2007;245:232-40.

25. Kunisaki C, Hatori S, Imada T, Akiyama H, Ono H, Otsuka Y, Matsuda G, Nomura M, Shimada H. Video-assisted thoracoscopic esophagectomy with a voice-controlled robot: The AESOP system. Surg Laparoscopy Endosc Percutaneous Tech. 2004;14:323-7.

26. Kitagawa H, Akimori T, Okabayashi T, Namikawa T, Sugimoto T, Kobayashi M, Hanazaki K. Total laparoscopic gastric mobilization for esophagectomy. Langenbecks Arch Surg. 2009;394:617-21.

27. Braghetto I, Csendes A, Cardemil G, Burdiles P, Korn O, Valladares H. Open transthoracic or transhiatal esophagectomy versus minimally invasive esophagectomy in terms of morbidity, mortality and survival. Surg Endosc. 2006;20:1681-6.

28. Osugi H, Takemura M, Higashino M, Takada N, Lee S, Kinoshita H. A comparison of video-assisted thoracoscopic oesophagectomy and radical lymph node dissection for squamous cell cancer of the oesophagus with open operation. Br J Surg. 2003;90:108-13.

29. Benzoni E, Bresadola V, Terrosu G, Uzzau A, Cedolini C, Intini S, Noce L, Bresadola F. Minimally invasive esophagectomy: a comparative study of transhiatal laparoscopic approach versus laparoscopic right transthoracic esophagectomy. Surg Laparosc Endosc Percutan Tech. 2008;18:178-87.

30. Palanivelu C, Prakash A, Senthilkumar R, Senthilnathan P, Parthasarathi R, Rajan PS, Venkatachlam S. Minimally invasive esophagectomy: thoracoscopic mobilization of the esophagus and mediastinal lymphadenectomy in prone position--experience of 130 patients. J Am Coll Surg. 2006;203:7-16.

31. Hulscher JB, van Sandick JW, de Boer AG, Wijnhoven BP, Tijssen JG, Fockens P, Stalmeier PF, ten Kate FJ, van Dekken H, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the esophagus. N Engl J Med. 2002;21;347:1662-9.

32. Nishihira T, Hirayama K, Mori S. A prospective randomized trial of extended cervical and superior mediastinal lymphadenectomy for carcinoma of the thoracic esophagus. Am J Surg. 1998;175:47-51.

33. Boone J, Schipper MEI, Moojen WA, Borel Rinkes IHM, Cromheecke GJE, van Hillegersberg R. Robotassisted thoracoscopic esophagectomy for esophageal cancer: short- and mid-term results. Br J Surg. 2009;96:878-86.

34. Boone J, Liverstro DP, Elias SG, Borel Rinkes IHM, van Hillegersberg R. International survey on esophageal cancer: Part I Surgical techniques. Dis Esophagus. 2009;22:195-202.

CHAPTER 3

Risk of thromboembolic events after perioperative chemotherapy versus surgery alone for esophageal adenocarcinoma

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ABSTRACT

Background

Major oncologic surgery is associated with a high incidence of thromboembolic events (TEE). Addition of perioperative chemotherapy in esophageal cancer surgery may increase the risk of TEE.

Materials and methods

The thromboembolic toxicity profile was analyzed in patients with esophageal adenocarcinoma (EAC). Two groups were identified: patients who underwent esophagectomy and received perioperative chemotherapy with epirubicin, cisplatin and capecitabine (ECC; n=52), and patients who were treated with surgery alone (n=35).

Results

A total of 22 TEEs was observed in 17 patients (32.7%) in the chemotherapy group and 3 patients (7.5%) in the surgery-alone group ($p<.01$). The relative risk of developing a TEE for patients receiving perioperative chemotherapy during the whole treatment period was 3.8 (95% CI 1.2 to 12.0). A preoperatively occurring TEE did not increase the risk of postoperative TEE, nor did it increase postoperative hospital stay ($p=.325$). Median postoperative hospital stay was 23 days (range 14-78) for patients with a postoperative TEE and 15 days (range 10-105) for patients without TEE ($p=.126$). Perioperative chemotherapy with the epirubicin, cisplatin and capecitabine regimen was independently associated with the development of TEE in the combined preoperative and postoperative period ($p=.034$).

Conclusion

Perioperative chemotherapy improves survival for operable esophageal cancer but comes at the price of toxicity. Perioperative chemotherapy for EAC increases the risk of TEE. However, chemotherapy-related preoperative TEE did not increase the risk of postoperative TEE, nor did it increase postoperative hospital stay, justifying its use in clinical practice.

INTRODUCTION

In the Western world, esophageal adenocarcinoma (EAC) has become the most prevalent type of cancer in the esophagus, overtaking squamous cell carcinoma.[1-3] Perioperative treatment of patients with operable adenocarcinoma of the distal esophagus, gastroesophageal junction and stomach with chemotherapy consisting of epirubicin, cisplatin and 5-fluorouracil improves overall survival in patients when compared to surgery alone.[4] The regimen consists of three preoperative treatment cycles followed by surgery and 3 postoperative cycles. Oral administration of capecitabine as an alternative for prolonged intravenous infusion of 5-fluorouracil showed equal efficacy in patients with advanced or inoperable esophagogastric cancer.[5] The epirubicin, cisplatin and capecitabine (ECC) regimen can be used as a substitute for the epirubicin, cisplatin and 5-fluorouracil (ECF) regimen.[6] Our current standard for patients with operable esophageal and gastroesophageal junction adenocarcinoma is perioperative chemotherapy with the ECC regimen combined with radical esophagectomy and gastric conduit reconstruction.

Gastroesophageal cancers are associated with the highest risk of venous thrombosis.[7] The reported incidence of thromboembolic events (TEE) during chemotherapy for advanced gastroesophageal cancer is approximately 12% and even higher when patients are treated with cisplatin.8 Another important risk factor for TEE is major (oncologic) surgery.[9,10]

Still, reports of TEE during chemotherapy are limited to series of patients with advanced inoperable gastroesophageal cancer. Little is known about the incidence and consequences of TEE during *perioperative* chemotherapy in patients with cancer of the upper gastrointestinal tract. This study shows our experience of the past 4 years during which the ECC protocol was applied in patients with EAC. The thromboembolic toxicity profile of patients receiving perioperative ECC chemotherapy was analyzed and compared with a group of patients from the same time period who were treated with surgery alone.

METHODS

DATA COLLECTION

A prospective database of all esophageal resections in our tertiary referral center (University Medical Center Utrecht, Utrecht, the Netherlands) is collected continuously. The research protocol was in accordance with the guidelines of the medical ethics committee. Database entries include standard patient characteristics, as well as prospectively collected intra- and postoperative data. The database enables entry of various complications among which deep venous thrombosis, pulmonary embolism and myocardial infarction form separate entries. Atrial fibrillation was also documented in the database because this condition is associated with both esophagectomy and an increased

risk of postoperative thromboembolic complications. Complications that occurred during preoperative chemotherapy were retrieved from the patient records of the medical oncology department.

INCLUSION CRITERIA

The study population consisted of all consecutive patients who underwent esophageal resection with gastric conduit reconstruction for EAC complemented with perioperative ECC therapy (January 1, 2007, to February 1, 2011). To eliminate historical bias, the control group comprised patients who underwent the same surgical treatment without chemotherapy in that same time period (surgery-alone group). Reasons for not commencing preoperative chemotherapy were T1 disease, weight loss >10%, World Health Organization performance status[11] >2, and Groningen Frailty Index >3 (for patients above 70 years of age[12]).

CHEMOTHERAPY

Patients eligible for perioperative ECC chemotherapy received three preoperative treatment cycles and another three postoperative treatment cycles. One chemotherapy cycle consisted of intravenous administration of epirubicin and cisplatin on day 1 (50 and 60 mg/m^2, respectively) followed by 625 mg/m^2 of capecitabine twice daily for 21 days. Adaptations to the regimen (i.e. dose reduction or change of regimen) were applied when necessary on the basis of the occurrence of adverse events during therapy as defined by the Common Terminology Criteria for Adverse Events (CTCAE) and the Common Toxicity Criteria.[13]

After completion of the second cycle, a computed tomography (CT) scan was performed to evaluate response. In case of tumor progression, a third cycle was not administered, and surgery was rescheduled accordingly. In case of a TEE during preoperative chemotherapy, anticoagulative therapy was initiated with low molecular weight heparin (Fragmin, Pfizer, New York, NY; daily; subcutaneous; < 80 kg, 2500 IU; > 80 kg, 5000 IU).

SURGICAL THERAPY

Surgical therapy consisted of esophageal resection with gastric conduit reconstruction through a transthoracic or transhiatal approach performed by a single experienced surgeon. Preferably, esophagectomy was performed with the use of minimally invasive surgery. Transthoracic surgery included two-field lymphadenectomy. Abdominal lymphadenectomy was similar for all patients (truncal and perigastric). Operative approach was matched to patient physiology and tumor characteristics. All esophagogastric anastomoses were hand sewn and situated cervically on the left side. Perioperatively, all patients received standard thrombotic prophylaxis with low molecular weight heparin (Fragmin, Pfizer; daily; subcutaneous; < 80 kg, 2500 IU; > 80 kg, 5000 IU) and stockings.

PRIMARY OUTCOME

The primary outcome was defined as the occurrence of a TEE during one of three distinct periods as follows: (1) the preoperative period, defined as the day of diagnosis until the day of surgery; (2) the postoperative period, defined as during the hospital stay or until the 30th day after surgery; and (3) the postoperative chemotherapy period, from the day on which postoperative chemotherapy was resumed until 30 days after the last administration of chemotherapy. The latter period was only applicable to those patients who received postoperative chemotherapy. TEEs were defined and graded according to CTCAE version 4.03.13 Only events graded 2 or higher were included for analysis (table 1).

Table 1 TEE grades as defined by the CTCAE

CTCAE grade	Definition
1*	Venous thrombosis (e.g., superficial thrombosis)
2	Venous thrombosis (e.g., uncomplicated deep vein thrombosis), medical intervention indicated
3	Thrombosis (e.g., uncomplicated pulmonary embolism [venous], non-embolic cardiac mural [arterial] thrombus, medical intervention indicated
4	Life-threatening (e.g., pulmonary embolism, cerebrovascular event, arterial insufficiency), hemodynamic or neurologic instability, urgent intervention indicated
5	Death

* Grade 1 not included for analysis

STATISTICAL ANALYSIS

Data were analyzed by SPSS statistical software version 15.0 (SPSS, Chicago, IL). Cross-tabulation with Chi-square testing was used to identify differences between cohorts regarding baseline characteristics and the occurrence of TEE during separate and combined treatment periods. The Student's t-test was used for continuous variables. Associations between risk factors and TEE were analyzed with univariate regression analysis. Multivariate logistic regression was applied to correct for confounders. The following risk factors and possible confounders with respect to the development of TEE were identified before analysis: age above 60 years, gender, body mass index above 27, American Society of Anesthesiologists score, chronic obstructive pulmonary disease (COPD), diabetes, hypertension, previous TEE, history of smoking, length of hospital stay, operative approach, operative time, presence of tumor-positive lymph nodes, and tumor differentiation grade. Anticoagulant use at the time of diagnosis was included as a protective factor. A *P value* of <.05 was considered statistically significant.

RESULTS

Between January 1, 2007, and February 1, 2011, a total of 107 patients underwent esophagectomy with gastric conduit reconstruction for EAC. After exclusion of patients who received chemoradiotherapy or primary chemotherapy other than ECC, 87 patients remained. Mean age was 65 years, and the ratio of men to women was approximately 4:1.

COHORTS AND BASELINE

Perioperative chemotherapy in the form of ECC was administered to 52 patients. The remaining 35 patients did not receive chemotherapy. Reasons for not giving chemotherapy are presented in table 2. Both age and gender were evenly distributed between the surgery-alone group and the group receiving ECC. In the surgery-alone group more patients had a history of COPD (28.6% vs. 11.5%, χ^2 test p=.044). With regard to other relevant medical history, no statistically significant differences were identified between the two groups (table 3; baseline characteristics).

Preoperative tumor staging with endoscopic ultrasound (uT) showed more patients with early stage disease (uTis-1, n=11) in the surgery-alone group, as opposed to none in the chemotherapy group. The number of patients with advanced (uT3-4) disease was evenly dispersed over the groups. Postoperative pathological tumor staging (pT) showed no statistically significant difference between the two groups. An equal spread of differentiation grades was observed. The number of patients with positive lymph nodes at pathological examination was slightly higher in the group that did not receive chemotherapy (65.7% vs. 46.2% in the chemotherapy group, χ^2 test p=.073). The majority of patients (73.1%) in the chemotherapy group were operated on via a transthoracic approach, as opposed to approximately half of the patients (51.4%) in the surgery-alone group (χ2 test p=.039).

Table 2 Reasons for not receiving neo-adjuvant chemotherapy in 35 of 87 patients

	n
Patients unfit for perioperative chemotherapy*	17
Previous chemotherapy with serious toxicity	2
Weight loss >10%	2
Refused patient consent	2
Esophageal perforation requiring immediate intervention	1
Early-stage disease (uT ≤1)	11

* Patients with a WHO performance status >2(corresponding to a Karnofsky score of <70), or a Groningen Frailty Index >3 (for patients older than 70).

Table 3 Baseline characteristics before treatment selection and after surgery of patients treated with surgery alone versus patients treated with surgery and perioperative chemotherapy according to the ECC regimen

	Surgery alone (n=35)	Surgery & ECC (n=52)	*P value*
Pretreatment Data			
Gender			*0.751*
Male	28 (80.0%)	43 (82.7%)	
Female	7 (20.0%)	9 (17.3%)	
Age, years			
Mean (SD)[b]	66.9 (9.6)	64.4 (8.4)	*0.198*
>60yr	27 (77.1%)	35 (67.3%)	*0.320*
BMI >27	17 (48.6%)	26 (50.0%)	*0.896*
Medical history			
DM	7 (20.0%)	7 (13.5%)	*0.416*
Hypertension	12 (34.3%)	22 (42.3%)	*0.452*
COPD	10 (28.6%)	6 (11.5%)	***0.044***
TEE	13 (37.1%)	12 (23.1%)	*0.155*
Anticoagulant therapy	3 (8.6%)	5 (9.6%)	*0.869*
Smoking	18 (51.4%)	29 (55.8%)	*0.217*
Ultrasound T stage			***0.003***
uTis	3 (8.6%)	0 (0.0%)	
uT1	8 (22.9%)	0 (0.0%)	
uT2	3 (8.6%)	12 (23.1%)	
uT3	18 (51.4%)	36 (69.2%)	
uT4	2 (5.7%)	2 (3.8%)	
uTx	1 (2.9%)	2 (3.8%)	
Surgical Data			
ASA score			*0.116*
1	8 (22.9%)	13 (25.0%)	
2	17 (48.6%)	34 (65.4%)	
3	9 (25.7%)	5 (9.6%)	
4	1 (2.9%)	0 (0.0%)	
Operative approach			***0.039***
Transhiatal	17 (48.6%)	14 (26.9%)	
Transthoracic	18 (51.4%)	38 (73.1%)	
Surgery type			
Minimally invasive surgery	27 (77.1%)	41 (78.8%)	*0.850*
Open surgery	8 (22.9%)	11 (21.2%)	
Tumor differentiation grade			*0.807*
Poor	14 (40.0%)	21 (40.4%)	
Moderate	13 (37.1%)	17 (32.7%)	
Well	1 (2.9%)	4 (7.7%)	
Unknown	7 (20.0%)	10 (19.2%)	
Pathological Tumor stage			*0.079*
pTis	1 (2.9%)	0 (0.0%)	
pT0	0 (0.0%)	6 (6.5%)	
pT1	11 (31.5%)	8 (15.4%)	
pT2	2 (5.7%)	7 (13.5%)	
pT3	19 (54.3%)	30 (57.7%)	
pT4	2 (5.7%)	1 (1.9%)	
Lymph nodes			
Total resected, median (range)[c]	19 (5-65)	20 (2-49)	*0.396*
Positive lymph nodes	23 (65.7%)	24 (46.2%)	*0.073*
Radicality (R0)	33 (94.3%)	50 (96.2%)	*0.683*
Length of hospital stay	17 (10-105)	15 (10-46)	*0.137*
Days, median (range)[c]			

a χ^2 test unless otherwise indicated. b two-sample t-test. c Mann-Whitney U-test

ECC - Epirubicin Cisplatin & Capecetabine; BMI - Body Mass Index; DM - Diabetes Mellitus; COPD - Chronic Obstructive Pulmonary Disease; TEE - Thromboembolic Event; ASA - American Society of Anesthesiologists; R0 - resection margin free of microscopic tumor cells.

TEE

Overall, 20 patients experienced a total of 22 TEEs at some stage during the three treatment periods (table 4). The observed TEEs included deep venous thrombosis (n=2), pulmonary embolism (n=9), aorta thrombosis (n=2), portal vein thrombosis (n=1), inferior caval vein thrombosis (n=1), myocardial infarction (n=2), cardiac mural thrombosis (n=1) and stroke (cardiovascular accident; n=4).

Within the group of patients receiving chemotherapy, 17 patients (32.7%) experienced a TEE. Three patients (7.5%) were diagnosed with TEE in the surgery-alone group (χ^2 test p<.01). The relative risk of developing a TEE during the whole treatment period for patients treated with chemotherapy was 3.8 (95%CI 1.2-12.0; χ^2 test p=.009). The relative risk of developing a TEE during the combined preoperative and postoperative period was 3.6 (95%CI 1.1-11.4; chemotherapy group: n=16; surgery-alone group: n=3; χ^2 test p=.014).

PREOPERATIVE PERIOD

In the preoperative period, 14 patients were diagnosed with a TEE. All of these TEEs occurred in the cohort of chemotherapy patients (26.9%). Patients receiving chemotherapy had a follow-up CT scan for evaluation of response after the second preoperative treatment cycle.

Table 4 Number of TEEs during treatment in control and ECC group

Treatment period	TEE grade	Surgery alone (n=35)	Surgery & ECC (n=52)
Preoperative	Total	0 (0.0%)	14 (29.9%)
	2	0	1
	3	0	9
	4	0	4
	5	0	0
Postoperative	Total	3 (8.6%)	3 (5.8%)
	2	0	0
	3	1	1
	4	2	2*
	5	0	0
Postoperative chemotherapy	Total	NA	2 (3.8%)
	2	NA	0
	3	NA	0
	4	NA	2**
	5	NA	0

* one patient also had a pre-operative TEE (grade 4). ** one patient also had a pre-operative TEE (grade 3). TEE - thromboembolic event; NA - not applicable.

This led to coincidental detection of TEE in 6 cases. In 8 patients, the TEEs were symptomatic. The median interval between CT scan for response evaluation and surgery was 57 (range 7-91) days for patients who received chemotherapy. The median time interval between the last preoperative CT scan and surgery was 59 (range 1-92) days for the surgery-alone group (Mann-Whitney U-test p=. 915).
In all cases of TEE anticoagulant therapy was initiated. For 6 patients with preoperative TEE, chemotherapy was stopped`, and in 2 cases, only cisplatin therapy was stopped (table 5). The other 6 TEEs were discovered after the third treatment cycle. None of the patients was excluded for surgery. The median time to surgery from the last day of chemotherapy was 34 (range 13-59) days for patients who did not have a preoperative TEE and 38.5 (range 15-91) days for patients with a preoperative TEE (Mann-Whitney U test; p=.239).
Median postoperative hospital stay for patients with a preoperative TEE was 14 (range 11-27) days and 17 (range 10-105) days for patients without TEE (Mann-Whitney U-test; p=.325).

POSTOPERATIVE PERIOD

During the postoperative period, three TEEs occurred in the chemotherapy group (5.8%). Another three events were recorded in the surgery-alone group (8.6%, χ^2 test p=.613). None of the patients with postoperative TEE was diagnosed with atrial fibrillation. One of the patients with postoperative TEE developed a thrombus in the left ventricle during preoperative chemotherapy. In the other 5 cases, no preoperative TEEs occurred.
Median postoperative hospital stay for patients with a postoperative TEE was 23 (range 14-78) days and 15 (range 10-105) days for patients without TEE (Mann-Whitney U-test; p=.126).

Table 5 Patients with preoperative TEEs and changes to their chemotherapy regimen

Change in preoperative regimen	n	Timing
Stop chemotherapy	1	During 1st cycle
	1	During 2nd cycle
	3	After 2nd cycle
	1	During 3rd cycle
Stop cisplatin, switch to oxaliplatin	1	After 1st cycle, completed 2nd cycle with oxaliplatin, no 3rd cycle
Stop Cisplatin	1	After 2nd cycle, completed 3rd cycle without Cisplatin
No change	6	TEE occurred after 3rd cycle*
Total	14	

* In 4 patients, capecetabin dosage was reduced for polyneuropathy

POSTOPERATIVE CHEMOTHERAPY PERIOD

After recovering from surgery, 21 of 52 patients continued chemotherapy. In 31 cases, postoperative chemotherapy was not administered for various reasons. Toxicity during preoperative chemotherapy formed the main reason for not receiving postoperative therapy (n=16). Other reasons were metastatic disease (n=4), poor patient condition (n=6), death (n=1), irradical resection (n=1) and withdrawn consent (n=3).

Two patients developed a grade 4 TEE (cardiovascular accident) during postoperative chemotherapy. One of these patients already had deep venous thrombosis and pulmonary embolism during preoperative therapy. Further postoperative chemotherapy was discontinued in both patients.

Table 6 Univariate and multivariate analysis of the association between risk factors and development of TEE during the combined preoperative & postoperative period

Characteristic	Unadjusted OR (95% CI) univariate	*P value*	Adjusted OR (95% CI)* multivariate	*P value*
Gender (male)	0.804 (0.226-2.853)	*0.735*		
Age (continuous)	0.988 (0.934-1.046)	*0.684*		
Age (>60 y)	0.840 (0.279-2.531)	*0.757*		
BMI (>27)	0.900 (0.325-2.492)	*0.839*		
TEE in history	1.620 (0.552-4.755)	*0.380*		
DM	0.549 (0.122-2.698)	*0.460*		
Hypertension	0.659 (0.224-1.944)	*0.450*		
COPD	0.454 (0.094-2.200)	*0.327*	0.687 (0.130-3.634)	*0.659*
Smoking	1.223 (0.474-3.151)	*0.677*		
Positive LNN	0.931 (0.336-2.580)	*0.891*	1.347 (0.445-4.070)	*0.598*
Tumor differentiation grade	0.958 (0.682-1.345)	*0.803*		
LOS	0.999 (0.964-1.036)	*0.967*	1.014 (0.975-1.054)	*0.501*
ASA score	0.534 (0.235-1.211)	*0.133*		
Operation time	1.002 (0.997-1.008)	*0.458*		
Transthoracic surgery	1.733 (0.559-5.378)	*0.341*	1.129 (0.334-3.817)	*0.845*
Open surgery	0.353 (0.074-1.687)	*0.192*		
Anticoagulant therapy	0.484 (0.056-4.199)	*0.510*		
Chemotherapy	4.741 (1.264-17.780)	***0.021***	4.937 (1.131-21.545)	***0.034***

* included for multivariate regression analysis were possible confounders (i.e. risk factors that showed univariate association (p<.100) with chemotherapy) OR – odds ratio; CI - confidence interval; TEE - thromboembolic event; BMI - body mass index; DM - diabetes mellitus; COPD - chronic obstructive pulmonary disease; LNN - lymph nodes; LOS - length of hospital stay; ASA - American Society of Anesthesiologists.

RISK FACTORS FOR THROMBOEMBOLIC COMPLICATIONS

Besides preoperative treatment with ECC, none of the risk factors showed statistically significant correlation with TEE in univariate logistic regression analysis. To test for possible confounders, all risk factors were analyzed for correlations with the use of chemotherapy by univariate logistic regression (data not shown). Factors with a correlation *P value* of <.100 were subjected to further analysis. A history of COPD and length of stay were inversely correlated with the use of preoperative chemotherapy (p=.051 and p=.042 respectively). Transthoracic surgery and the presence of positive lymph nodes in the resection specimen were positively correlated with the use of preoperative chemotherapy (p=.041 and p=.075 respectively). To correct for possible confounding, these factors were included in multivariate logistic regression analysis.

Table 6 provides an overview of the analyzed risk factors in univariate analysis as well as the possible confounding risk factors in multivariate analysis. Preoperative chemotherapy was an independent predictor for developing TEE during the combined preoperative and postoperative period (p=.034). None of the possible confounding risk factors contributed in a statistically significant way in multivariate analysis.

DISCUSSION

The current study shows that TEE is more frequent among patients selected for perioperative treatment with the ECC regimen compared to patients not receiving chemotherapy. The majority of preoperative TEEs were grade 3 and 4, which are potentially lethal. In all cases, TEE required medical intervention with anticoagulant therapy, and in most cases the chemotherapy regimen was adjusted or discontinued. However, preoperative TEE did not disqualify patients for surgery nor did it increase the risk of postoperative TEE.

Some studies have specifically reported on the prevalence and significance of venous thrombosis in gastroesophageal cancer patients receiving palliative chemotherapy.[14,15] To date, to our knowledge, no studies have reported on the effect of TEE on perioperative outcomes in patients receiving neoadjuvant ECC therapy. This study specifically focuses on patients with EAC comparing patients who receive a specific treatment regimen (ECC) with patients not receiving perioperative treatment in the same period. Hence, treatment standards (e.g., surgical expertise and intensive care unit protocols) were the same for all patients. However, selection bias, by which patients are selected for chemotherapy, could not be eliminated. Baseline characteristics show a less advanced endoscopically defined tumor stage (uT), but also a poorer patient condition in the surgery-alone group. Nonetheless, TEE rates are higher in the chemotherapy group. Postoperative pathologically defined tumor stage (pT), particularly for advanced disease stage (pT3), is equal in both groups. Hence, higher T-stage does not explain the difference in TEE incidence observed

in this study. Moreover, despite certain baseline differences, multivariate analysis identified chemotherapy as the only independent risk factor for preoperative and postoperative TEE. The MAGIC trial, did not describe excess thromboembolic complications during treatment.[4] The authors did recommend the use of prophylactic antithrombotic therapy. The MAGIC regimen used continuous intravenous fluorouracil administration during 21 consecutive days and recommended warfarin as a prophylaxis for intravenous catheter-associated thrombosis. However, it remains unclear what percentage of patients actually received prophylaxis during the trial. Also, we may have detected more subclinical TEEs as a result of the follow-up CT scan (for response evaluation) in the chemotherapy group. Patients from the surgery-alone group did not routinely undergo a second CT scan before surgery. Nevertheless, the time interval between the last preoperative CT scan and surgery was equal in both groups. Furthermore, 8 out of 14 preoperative TEEs were symptomatic. The observed TEE rate in our analysis of the preoperative chemotherapy group suggests that without prophylaxis, a high TEE rate can be expected.

All patients with preoperative TEE received anticoagulant therapy until surgery. The subsequent effect on postoperative TEE cannot be measured in this study. Careful monitoring by the medical oncologist and early intervention has proved to prevent worsening of thromboembolic disease and most probably prevented postoperative events. Only 1 patient with a preoperative TEE also developed a postoperative TEE. The other 5 patients with postoperative TEE did not use anticoagulant therapy in the preoperative period.

During the last year of the inclusion period, several candidates for perioperative therapy were given an alternative regimen in which cisplatin (ECC therapy) was replaced by oxaliplatin (EOC therapy). Oxaliplatin is as effective as cisplatin in the treatment of patients with untreated advanced gastroesophageal cancer, but it is associated with a lower incidence of thromboembolism.[8,16,17]

In 1 patient with preoperative TEE during the first treatment cycle, ECC was changed to EOC therapy. This patient did not receive a third treatment cycle. In another patient with TEE discovered after the second treatment cycle, cisplatin was not administered during the third cycle. In the other 6 cases, ECC therapy was changed to EOC therapy for reasons of ototoxicity (n=2) and nephrotoxicity (n=4). Because patients received ECC during the first treatment cycle, and based on the intention-to-treat principle, these patients were included in our analysis. This has possibly caused an underestimation of the TEE rate in the ECC cohort. Though speculative, it could be argued that without the alternative EOC regimen, preoperative and postoperative TEE rates would have turned out higher. It must, however, be noted that there is no evidence that oxaliplatin is preferred over cisplatin in the curative perioperative setting with respect to survival and toxicity.

Because transthoracic surgery takes more time and includes more extensive dissection than transhiatal surgery, it could be argued that operative approach might influence postoperative TEE rates. However, operative approach as a possible confounder was not

correlated with TEE in multivariate analysis. With respect to postoperative recovery, no negative effects of chemotherapy were noticed. The median hospital stay was similar in both study groups. In addition, experiencing TEE during the preoperative period was not associated with increased postoperative hospital stay. Though not statistically significant, median hospital stay was increased with 7 days for patients who experienced a postoperative TEE.

Because venous thrombotic events can extend beyond the inpatient recovery period, the postoperative period included inpatient hospital stay as well as the postdischarge period up to 30 days after initial surgery.[18] Despite this extended period, there was no observation of symptomatic TEE in the outpatient clinic. Moreover, there was no increased incidence of postoperative TEE within the group of patients who experienced a preoperative TEE. Perioperative treatment with the ECC regimen improves survival in patients with EAC, but this benefit is accompanied by toxicity that could seriously harm patients and their surgical outcomes. The current study showed that TEE was frequent and was independently associated with chemotherapy. Although they were treated adequately, preoperative TEEs did not greatly influence time to surgery, hospital stay, or the occurrence of postoperative TEE. Nonetheless, in each individual case, it should be evaluated whether the benefits of the ECC regimen outweigh the increased risk of TEE. All patients who receive preoperative chemotherapy according to the ECC regimen should undergo a follow-up CT scan after the second treatment cycle. Imaging should not only focus on treatment evaluation, but also on signs of thromboembolic disease.

REFERENCES

1. Cook MB, Chow WH, Devesa SS. Oesophageal cancer incidence in the United States by race, sex, and histologic type, 1977-2005. Br J Cancer. 2009;101(5)855-859.
2. Bosetti C, Levi F, Ferlay J, Garavello W, Lucchini F, Bertuccio P, Negri E, La VC. Trends in oesophageal cancer incidence and mortality in Europe. Int J Cancer. 2008;122(5)1118-1129.
3. Lepage C, Rachet B, Jooste V, Faivre J, Coleman MP. Continuing rapid increase in esophageal adenocarcinoma in England and Wales. Am J Gastroenterol. 2008;103(11)2694-2699.
4. Cunningham D, Allum WH, Stenning SP, Thompson JN, Van d, V, Nicolson M, Scarffe JH, Lofts FJ, Falk SJ, Iveson TJ, Smith DB, Langley RE, Verma M, Weeden S, Chua YJ, MAGIC TP. Perioperative chemotherapy versus surgery alone for resectable gastroesophageal cancer. N Engl J Med. 2006;355(1)11-20.
5. Evans TR, Pentheroudakis G, Paul J, McInnes A, Blackie R, Raby N, Morrison R, Fullarton GM, Soukop M, McDonald AC. A phase I and pharmacokinetic study of capecitabine in combination with epirubicin and cisplatin in patients with inoperable oesophago-gastric adenocarcinoma. Ann Oncol. 2002;13(9)1469-1478.
6. Rich TA, Shepard RC, Mosley ST. Four decades of continuing innovation with fluorouracil: current and future approaches to fluorouracil chemoradiation therapy. J Clin Oncol. 2004;22(11)2214-2232.
7. 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(8)1702-1709.
8. Starling N, Rao S, Cunningham D, Iveson T, Nicolson M, Coxon F, Middleton G, Daniel F, Oates J, Norman AR. Thromboembolism in patients with advanced gastroesophageal cancer treated with anthracycline, platinum, and fluoropyrimidine combination chemotherapy: a report from the UK National Cancer Research Institute Upper Gastrointestinal Clinical Studies Group. J Clin Oncol. 2009;27(23)3786-3793.
9. Agnelli G, Bolis G, Capussotti L, Scarpa RM, Tonelli F, Bonizzoni E, Moia M, Parazzini F, Rossi R, Sonaglia F, Valarani B, Bianchini C, Gussoni G. A clinical outcome-based prospective study on venous thromboembolism after cancer surgery: the @RISTOS project. Ann Surg. 2006;243(1)89-95.
10. Khorana AA, Connolly GC. Assessing risk of venous thromboembolism in the patient with cancer. J Clin Oncol. 2009;27(29)4839-4847.
11. Oken MM, Creech RH, Tormey DC, Horton J, Davis TE, McFadden ET, Carbone PP. Toxicity and response criteria of the Eastern Cooperative Oncology Group. Am J Clin Oncol. 1982;5(6)649-655.
12. Schuurmans H, Steverink N, Lindenberg S, Frieswijk N, Slaets JP. Old or frail: what tells us more? J Gerontol A Biol Sci Med Sci. 2004;59(9)M962-M965.
13. National Cancer Institute. Common Terminology Criteria for Adverse Events v.4.03. 2010.
14. Tesselaar ME, Ouwerkerk J, Nooy MA, Rosendaal FR, Osanto S. Risk factors for catheter-related thrombosis in cancer patients. Eur J Cancer. 2004;40(15)2253-2259.
15. Tetzlaff ED, Correa AM, Baker J, Ensor J, Ajani JA. The impact on survival of thromboembolic phenomena occurring before and during protocol chemotherapy in patients with advanced gastroesophageal adenocarcinoma. Cancer. 2007;109(10)1989-1995.
16. Cunningham D, Starling N, Rao S, Iveson T, Nicolson M, Coxon F, Middleton G, Daniel F, Oates J, Norman AR. Capecitabine and oxaliplatin for advanced esophagogastric cancer. N Engl J Med. 2008;358(1)36-46.
17. Montagnani F, Turrisi G, Marinozzi C, Aliberti C, Fiorentini G. Effectiveness and safety of oxaliplatin compared to cisplatin for advanced, unresectable gastric cancer: a systematic review and meta-analysis. Gastric Cancer. 2011;14(1)50-55.
18. Fleming FJ, Kim MJ, Salloum RM, Young KC, Monson JR. How much do we need to worry about venous thromboembolism after hospital discharge? A study of colorectal surgery patients using the National Surgical Quality Improvement Program database. Dis Colon Rectum. 2010;53(10)1355-1360.

CHAPTER 4

Fibrin-thrombin coated sealant increases strength of esophagogastric anastomoses in a rat model

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ABSTRACT

Background

Anastomotic leakage is a feared complication after esophagectomy. The purpose of this study was to investigate whether the use of a fibrin-thrombin coated collagen patch (TachoSil®, Nycomed, Zurich, Switzerland), applied as a sealant, would strengthen the esophagogastric anastomosis and stimulate anastomotic healing in a rat model.

Methods

Hand sewn, end-to-side esophagogastric anastomoses were performed in 54 rats. Animals were randomized for an unsealed or sealed anastomosis. Rats were sacrificed on postoperative days 0, 3, 5, and 7. Primary parameter was bursting pressure. Secondary outcomes were complications, weight and immunohistochemical staining for collagen formation and fibroblast activity.

Results

Bursting pressure at days 0 and 3 was significantly increased when a sealant was used (55.1±4.6 mmHg vs. 102.4±7.3 mmHg, p<.010; and 19.7±3.3 mmHg vs. 34.6±4.9 mmHg, p<.050 respectively). There was no difference in bursting pressure at days 5 and 7 between unsealed and sealed anastomoses (60.9±18.2 mmHg vs. 53.4±6.6 mmHg, p=.690; and 118.8±20.2 mmHg vs. 97.2±8.3 mmHg, p=.374 respectively). Application of sealant independently influenced bursting pressure (p<.010). Increased fibroblastic activity was noticed at day 7 in sealed anastomoses (p<.050). There were no differences in weight gain between groups.

Conclusions

Additional sealing of the anastomosis increased anastomotic strength during early postoperative recovery when anastomotic strength is at its' weakest. The findings indicate that sealing of the anastomosis has the potential to prevent leakage after esophagectomy in humans.

INTRODUCTION

The only curative treatment strategy for patients with locally invasive esophageal cancer is surgical resection.[1-3] Commonly, the stomach is used for restoring continuity of the upper intestinal tract. Gastric conduit reconstruction is most frequently performed by means of a hand sewn, end-to-side esophagogastric anastomosis.[4]

Anastomotic leakage is a feared complication after esophagectomy. The reported incidence of leakage ranges from 3% up to 41% of which approximately 70% concerns leakage of the esophagogastric anastomosis.[5,6] Clinical leakage of the anastomosis has significant consequences during postoperative recovery[7], but also increases the risk of benign cervical strictures.[8] Moreover, leak-associated mortality rates are high (14.7%) compared to the overall in-hospital mortality rate after esophagectomy (4.1%).[5] Various techniques for constructing an esophagogastrostomy have been proposed in the literature and even comparative studies have not yet led to consensus.[6,9-11] This warrants further research on how to improve the anastomotic healing process and improve outcomes for patients after esophagectomy.

Several studies have proposed the use of a fibrin coated patch on colon anastomoses.[12-14] We hypothesized that the use of such a patch would have beneficial effects on esophagogastric anastomoses. The purpose of this study was to investigate whether the use of a fibrin-thrombin coated collagen patch (TachoSil®, Nycomed, Zurich, Switzerland), applied as a sealant, would strengthen the esophagogastric anastomosis and stimulate anastomotic healing in a rat model.

METHODS

Male wistar rats (n=54, weight 350-400 gram) were housed and cared for following Dutch Animal Welfare regulations at the Central Laboratory Animal Research Facility Utrecht. Food and water was available ad libitum. The study protocol was approved by the Animal Ethics Committee of Utrecht University. Primary outcome was defined as the maximum pressure of the esophagogastric anastomosis (in mmHg). Measurements were performed at varying intervals from operation in 4 different survival groups to assess longitudinal changes in bursting pressure during the anastomotic healing process: directly postoperative (0 days), 3 days postoperatively, 5 days postoperatively and 7 days postoperatively.

SURGICAL PROCEDURE

There was no preoperative fasting period. Rats were anesthetized with isoflurane gas. Antibiotics were not administered. Analgesia was managed with intramuscular bolus injections of carprofen at a dose of 5.0 mg/kg (24 hours preoperatively, preoperatively, postoperatively, 24 hours and 48 hours postoperatively). Atropine was administered with

bolus injections of 0.05mg/kg (preoperatively and postoperatively) to reduce salivary excretion and the risk of aspiration.

The surgical procedure was adapted from the method described by Cui *et al.*[15] Access to the upper abdomen was obtained through a 3-cm midline laparotomy under sterile conditions. Flimsy adhesions between the proximal stomach and the liver were sharply divided. The intra-abdominal distal esophagus was bluntly dissected and mobilized. Adequate mobilization was required for a tension-free anastomosis. The vagal nerve was identified and preserved to prevent postoperative gastroparesis. A 5mm hemoclip was placed distally on the gastroesophageal junction. The esophagus was dissected transversely approximately 3-5mm above the hemoclip. Supportive 7-0 polyamid sutures (Ethicon, Somerville, NJ, USA) were placed at 9, 12 and 3 o'clock to manipulate the esophagus and obtain optimal view of the mucosa. In the glandular portion of the proximal anterior stomach, between the first and second major branches of the left gastric artery, a 4-5mm oblique gastrotomy was made using a monopolar coagulation knife (ERBE, Tübingen, Germany). Adequate size of the opening was verified with a 14 gauge catheter, of which insertion should be without resistance.

An end-to-side single-layer anastomosis was constructed using continuous 7-0 polypropylene sutures (Ethicon, Somerville, NJ, USA). The sutures were placed through all layers of the stomach and the esophagus. Suturing began by placing a suture between the 9 o'clock end of the esophagotomy and 10 o'clock end of the gastrotomy. A continuous suture was placed to the 3' o clock end of the esophagus completing the posterior aspect of the anastomosis. A second suture was placed near the 9 o'clock end of the esophagotomy and the 10 o'clock end of the gastrotomy. The anterior side of the anastomosis was then completed similarly. The posterior and anterior running sutures were then tied where they joined together at the 3'o clock end of the anastomosis.

To exclude surgeon bias, randomization took place after completion the anastomosis. Animals were assigned to the control group (unsealed anastomosis) or the intervention group (sealed anastomosis) within each survival group though randomization with sealed envelopes.

In case of sealing the anastomosis, a fibrin thrombin coated collagen patch (TachoSil®, Nycomed, Zurich, Switzerland) was cut into tiles of approximately 5mm x 10mm. Tiles were placed around the anastomosis one by one. Each tile was gently pressed on the esophagogastrostomy for 30 seconds to ensure adhesion. In both groups, the omentum was pulled upwards and positioned between the anastomosis and the liver to reduce adhesions to the liver. The linea alba was closed with a continuous 4-0 polyglactin suture (Ethicon, Somerville, NJ, USA). The skin was closed intracutaneously using a running horizontal mattress technique.

Animals were allowed free access to water and soaked food after recovery from anesthesia.[16] To prevent the rats from eating their cage bedding (wood shaving) due to stress, they were placed on a grid during the first two days postoperatively. Animals were

examined daily to evaluate overall condition, weight and any signs of complications. Housing conditions and endpoints were applied as formulated by the Dutch regulations for animal welfare.

MACROSCOPIC ANALYSIS; BURSTING PRESSURE

Each animal was sacrificed by cardiac puncture under general anesthesia (isoflurane) on the assigned day depending on survival group. The esophagus was transected above the level of the diaphragm and resected en-bloc together with the stomach and proximal duodenum. A 4 mm oblique incision was made through the pyloric sphincter. The stomach was flushed with saline under low pressure to remove gastric contents. A 10 gauge blunt canule was then inserted through the duodenum into the distal stomach. A custom made clamp was placed over the distal stomach to secure the canule. Proximally, the esophagus was closed airtight with a hemoclip.

The canule was attached to a calibrated pressure system. Submerged in saline, the specimen was inflated with air at 200ml/h. Intraluminal pressure was monitored using a Spacelabs patient monitor (Spacelabs Healthcare, Issaquah, WA, USA). Data was transferred from the monitor to a laptop with the LabJack U12 data acquisition device (Meilhaus Electronic, Puchheim, Germany). DAQFactory software (AzeoTech, Ashland, OR, USA) was used for recording pressure. The bursting pressure was defined as the maximum pressure before air bubbles appeared. After acquisition, data were imported into the statistical graphing software GraphPad Prism 5 (GraphPad Software, La Jolla, CA, USA) and converted to mmHg.

MICROSCOPIC ANALYSIS; IMMUNOHISTOCHEMISTRY

Directly after measuring bursting pressure, specimens were fixed in formalin. For immunohistochemical analysis, tissue sections (4 µm thick) were prepared from formalin-fixed paraffin-embedded blocks.

Sections were stained with α-SMA monoclonal antibody (Sigma Aldrich, St Louis, MO, USA; 1:32000 dilution) with the use of an automated immunostainer (Bond-Max immunostainer, Leica Microsystems, Wetzlar, Germany). For Sirius Red staining, slides were deparaffinized in xylene (15 minutes) and dehydrated in serial ethanol dilutions (15 minutes). Between all steps, slides were rinsed with distilled water. After pre-treatment with 0.2% phosphomolybdic acid and rinsing, slides were incubated with Sirius Red (90 minutes). Slides were then treated with hydrochloric acid (2 minutes), dehydrated in serial ethanol dilutions and washed with xylene.

Two independent observers quantified the α-SMA and Sirius Red expression in stroma. Slides were reviewed under light microscopy at 200× magnification. Staining intensity in 10 selected regions was assigned a score of 0 to 4 (0 indicating no staining, 4 indicating maximal staining). For evaluation of α-SMA staining, vessel staining was not included in score assignment. Scores were added to result in a total score.

STATISTICAL ANALYSIS & SAMPLE SIZE

Bursting pressures were compared with one-way ANOVA (longitudinal) and Student *t* test (within each survival group). Effect size was estimated at 80% based on reported bursting pressures in sealed colonic anastomoses in rat models.[13,17-20] Including a multiple testing correction (Bonferroni), subsequent power calculation yielded 6 animals per randomization group for each postoperative interval (power 0.80, alpha 0.05). Accounting for a loss of 10%, sample size was set at 7 subjects per treatment group per survival group. The survival group of 0 days was set at 6 animals per treatment group, because postoperative loss was not relevant for this group. Immunohistochemical scoring was compared with Student *t* test. Inter-observer variation was analyzed with Pearson's R. Data are denoted as mean(±SEM) and were analyzed using standard statistical software (SPSS, version 17.0; SPSS inc, Chicago, IL, USA). *P values* <0.050 were considered statistically significant.

RESULTS

CLINICAL OUTCOMES

Postoperative weight is shown in figure 1. Weight reduced postoperatively until day 3. After day 3, the animals surviving 5 and 7 days regained weight. Changes in weight were comparable between both treatment groups.

Respiratory insufficiency (RI) was observed in 3 animals (table 1), most probably caused by aspiration of gastric contents after surgery. Due to the nature of the surgical intervention, the distal esophagogastric sphincter function was impaired. In combination with a horizontally orientated esophagus, this may have led to aspiration.

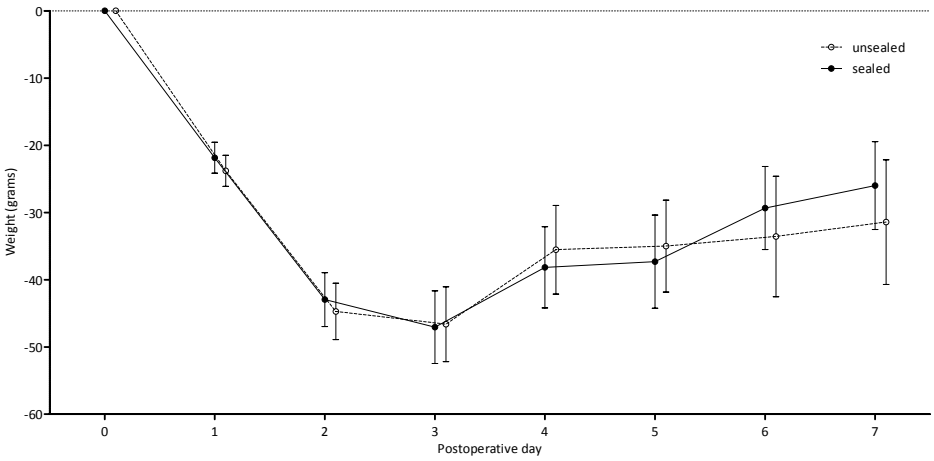

Figure 1 Postoperative weight gain. Data are mean (±SE).

Furthermore, 2 subjects were excluded from analysis in the acute group (0 days) due to incorrect application of the sealant. In the 3 days survival group, 1 animal with a sealed anastomosis died on postoperative day 1 without an identifiable cause at autopsy. Another animal from the sealed group was excluded from analysis since its burst pressure was an outlier (2.0 mmHg) compared to other measurements in both the sealed and unsealed group.

Table 1 Inclusion and exclusion of animal subjects per survival and randomization group

Survival group	Randomization	Included (n)	Exclusion or drop-out (n)	Reasons for exclusion
0	unsealed	6		
	sealed	4	2	Incorrect application of sealant (n=2)*
3	unsealed	6	1	Respiratory insufficiency on day 2 (n=1)†
	sealed	5	2	Unexplained death on day 1 (n=1), Extreme outlier (n=1)
5	unsealed	6	1	Respiratory insufficiency on day 3 (n=1)†
	sealed	7		
7	unsealed	7		
	sealed	6	1	Respiratory insufficiency on day 2 (n=1)†

* In two sealed subjects, the sealant was incorrectly applied. The sealants did not cover the anastomosis. Therefore, measured bursting pressures represented unsealed anastomoses instead of sealed anastomoses. When these two measurements (57.9 mmHg and 45.2 mmHg, respectively) were included for analysis, the *P value* for survival group 0 was P=0.037. † Animals which suffered from respiratory insufficiency were sacrificed following the Dutch Animal Welfare regulations and autopsy was performed.

In 2 subjects it was noticed at sacrifice on day 5 that cage bedding (wood chips) had been eaten, probably due to stress. The ingested bedding probably caused gastric obstruction which increases the chance of aspiration. Hereafter, animals were placed on a mesh instead of wood chips bedding for the first 2 postoperative days. When signs of distress were absent and intake of water and soaked food was sufficient, they were placed on standard cage bedding again.

MACROSCOPIC ANALYSIS

In 1 animal from the 7 days survival group (unsealed anastomosis), an abscess had formed at the anastomosis. This animal had shown no clinical signs of respiratory insufficiency or other forms of clinical distress and its weight had remained constant after day 3. None of the other animals showed signs of infection or leakage upon sacrifice. Inspection of the anastomosis in the animals which were sacrificed earlier due to respiratory insufficiency (n=3) did not reveal any signs of anastomotic leakage at autopsy. Instead, there was increased mucus secretion and pulmonary edema.

BURSTING PRESSURE

Bursting pressure directly postoperative was 55.1(±4.6) mmHg in unsealed anastomoses and 102.4(±7.3) mmHg in sealed anastomoses (p=0.000). At 3 days, mean bursting pressure was 19.7(±3.3) mmHg in unsealed anastomoses and 34.6(±4.9) mmHg in sealed anastomoses (p=0.029). At 5 days, unsealed anastomoses had a mean bursting pressure of 60.9(±18.2) mmHg and sealed anastomoses 53.4(±6.6) mmHg (p=.690). At 7 days, mean bursting pressures were 118.8(±20.2) mmHg and 97.2(±8.3) mmHg respectively (p=0.374). After log transformation to reduce heterogeneities in variance for one-way ANOVA, time and randomization (sealed vs. unsealed) independently influenced bursting pressure (p=0.031 and p=0.000 respectively). Bursting pressures are presented in figure 2.

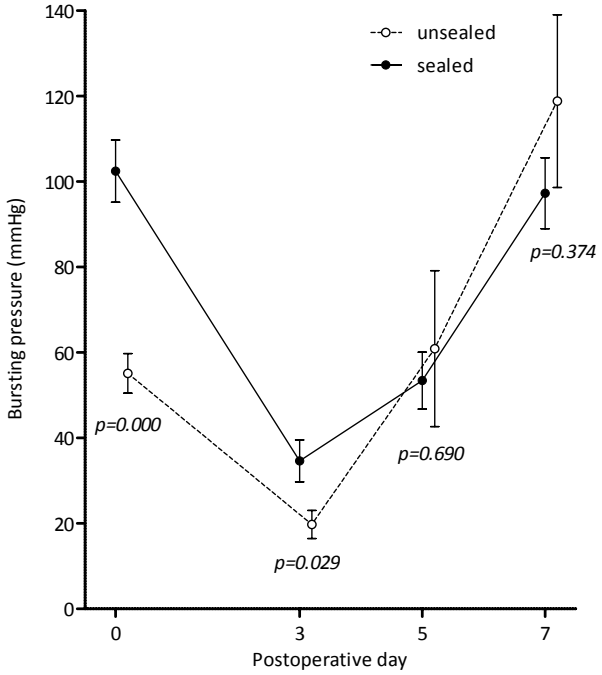

Figure 2 Postoperative bursting pressures. Data are mean (±SE). The *P values* of Student t test at different postoperative time intervals are shown.

IMMUNOHISTOCHEMISTRY

For immunohistochemical analysis, specimens from the 3 days and 7 days survival groups were first sectioned and H&E stained. Macrophage activity and inflammatory response were equally observed in both sealed and unsealed subjects. There were no concealed abscesses visible in the sealed specimens. In some specimens the anastomotic integrity was partially lost due to measurement of bursting pressure. From both survival groups, 7 sections were selected for Sirius Red and α-SMA staining (unsealed n=3, sealed n=4).

Scoring of immunohistochemical staining were significantly correlated between observers for Sirius Red (Pearson's R=0.900, SE=0.033, p=.000) as well as α-SMA (Pearson's R=0.951, SE=0.013, p=0.000). The mean staining score of Sirius Red was 4.33(±0.53) for unsealed anastomoses and 3.06(±0.32) for sealed anastomoses at 3 days postoperatively (p=0.051). At 7 days postoperatively, the mean scores were 7.17(±0.46) and 7.25(±0.33) respectively (p=0.882). The mean α-SMA staining score for unsealed anastomoses was 2.3(±0.38) and 2.25(±0.21) for sealed anastomoses (p=0.841). At 7 days postoperatively, the mean scores were 11.5(±2.29) and 17.6(±1.55) respectively (p=0.042). Increased fibroblastic activity at day 7 visualized with α-SMA staining is illustrated in figure 3.

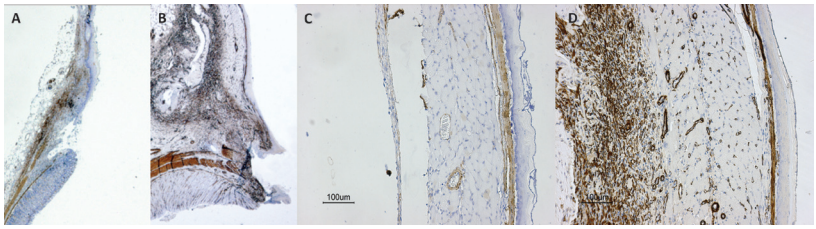

Figure 3 Immunohistochemical α-SMA staining at postoperative day 7 illustrating fibroblastic activity. Legend: A) An unsealed esophagogastric anastomosis (magnification 10x). B) A sealed esophagogastric anastomosis showing increased fibroblastic activity (magnification 10x). C) Detail of the esophageal wall showing the muscular layers and squamous epithelium (magnification 100x). D) Detail of the esophageal wall (similar to C) with the sealant applied showing increased fibroblastic activity (magnification 100x).

DISCUSSION

This is the first study to evaluate the effects of additional anastomotic sealing of an esophagogastrostomy with a fibrin thrombin coated patch in a validated rat model.[15,21,22] Bursting pressure was significantly higher postoperatively until at least 3 days after surgery when the anastomosis was sealed with a fibrin thrombin coated patch. In both groups, the lowest bursting pressures were measured 3 days postoperatively, indicating a strong degradation of extracellular matrix as described in earlier studies regarding anastomotic healing.[22-24] At day 5 the remodeling phase had already started, illustrated by an increase of bursting pressure in both groups. Remodeling continued until at least day 7, where bursting pressure returned to its' physiological range. There was no difference in bursting pressure at postoperative days 5 and 7 between sealed and unsealed anastomoses. Longitudinal analysis with one-way ANOVA showed that application of the sealant significantly increased the anastomotic strength during postoperative recovery. Histological analysis of the anastomoses from animals sacrificed at postoperative day 7 showed that the patch was still visible, but already partly absorbed. Both collagen content

(visualized by Sirius Red staining) and fibroblastic activity (visualized by α-SMA staining) were significantly increased on day 7 when compared to day 3. Between groups (sealed vs. unsealed), the collagen content was comparable at both postoperative days 3 and 7. These findings correspond to results of Schreinemacher *et al*. in colonic anastomoses.[25] This indicates that the fibrin thrombin coated patch does not increase formation of new collagen during the remodeling phase. We observed equal α-SMA staining at day 3 in sealed and unsealed anastomoses, but significantly increased fibroblastic activity at day 7 in the sealed group. This is comparable with findings in another study of colonic anastomoses.[26] The cellular response of fibroblasts is important for anastomotic healing. The observed early difference in bursting pressures and the absence of early histological differences imply that the increased anastomotic strength in the sealed group is mainly attributable to a mechanical strengthening of the anastomosis by the sealant and not so much by a cellular response.

The findings suggest that the sealant provides significant support of the anastomosis and the sutures during early recovery. Rats have the ability to recover quickly after surgery. Even after the highly invasive and complex procedure of this study, the animals regained weight very soon after surgery. We observed only 1 animal with macroscopic signs of leakage. The observed complications were due to aspiration, not anastomotic leakage. Furthermore, rats have powerful immune response, which causes fast resorption of the sealant. This might explain the absence of differences in bursting pressure at postoperative days 5 and 7. Moreover, in humans, clinical presentation of anastomotic leakage is usually observed within the first week.[24,31] The results show that the sealant strengthens the anastomosis when anastomotic strength is at its' weakest. It must be noted that increased bursting pressure does not equate with a reduced risk of leakage. Measurement of bursting pressure is impossible in humans. However, anastomotic sealing may have the potential to reduce leakage rates by strengthening the anastomosis during the weakest phase in early postoperative recovery. This hypothesis should be evaluated in a clinical study.

Stumpf *et al*. suggest that increased fibrosis may lead to obstruction of the anastomosis.[27] Although we observed higher fibroblastic activity in sealed anastomoses, we did not observe any strictures. In the studied subjects there was no increased complication rate in animals with sealed anastomoses. Moreover, weight gain was equal in both groups indicating sufficient intake and absence of esophagogastric strictures. Some studies report increased obstruction rates when wrapping a sealant around colonic[25] and ileal[27,28] anastomoses. This could be due to differences in tissue and techniques. Schreinemacher *et al*. and Chmelnik *et al*. studied the effects of sealants in large and small bowel, which have thinner and less muscular walls.[25,28] Furthermore, we used an end-to-side anastomotic technique as opposed to end-to-end. The end-to-side technique makes it more challenging to apply the sealant. However, we experienced that the application of the sealant was relatively easy to perform using the tile-like method as described earlier in vascular end-to-side anastomoses by van Doormaal *et al*.[29] Moreover, the tile-technique eliminates the

risk of constricting the anastomosis as could occur when wrapping the anastomosis at once like a scarf or sleeve.

The long term effects of esophagogastric sealing are yet unknown. One study with a fibrin coated collagen patch applied on porcine small bowel reported a 6 week follow-up and showed no difference in stenosis.[30] The patch was reported to be safe, but bursting pressures were comparable in sealed and unsealed subjects. However, the majority of anastomotic leakages and subsequent morbidity occur during early in-hospital recovery.[31] Still, a future clinical study should pay attention to the possible risks of stenosis. Careful endoscopic assessment of the esophagogastric lumen should be included in the design of such a study.

Some limitations of the current study must be mentioned. Since rat anatomy is not suitable for esophagectomy and gastric conduit reconstruction, the anastomosis was formed intra-abdominally. There was no alternative, in this animal model, to test the effect of sealants on esophagogastrostomies. An advantage of not using a model with gastric conduit reconstruction is that the influence of gastric blood flow on anastomotic healing is not of concern. Sufficient arterial blood flow[32,33] and venous outflow[34,35] are crucial for healing of the anastomosis. Since these factors did not play a role in the current study, the observed effects are only attributable to the sealant.

This study showed that bursting pressure was significantly increased during early postoperative recovery when a fibrin thrombin coated collagen patch was applied around the esophagogastric anastomosis. Although anastomotic healing is dependent on multiple factors, the current findings indicate that sealing of the anastomosis has the potential to prevent leakage after esophagectomy in humans. While manifestation of leakage occurs mainly during the first postoperative week when anastomotic strength is at its weakest, increased anastomotic strength during early recovery may be beneficial. Therefore, the applicability and effectiveness of esophagogastric sealing should be further examined in a clinical phase II study.

REFERENCES

1. Graham AJ, Shrive FM, Ghali WA, Manns BJ, Grondin SC, Finley RJ, Clifton J. Defining the optimal treatment of locally advanced esophageal cancer: a systematic review and decision analysis. Ann Thorac Surg. 2007;83(4)1257-1264.
2. Hulscher JB, van Sandick JW, de Boer AG, Wijnhoven BP, Tijssen JG, Fockens P, Stalmeier PF, Ten Kate FJ, van Dekken H, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the esophagus. N Engl J Med. 2002;347(21)1662-1669.
3. Verhage RJ, Hazebroek EJ, Boone J, van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64(2)135-146.
4. Boone J, Livestro DP, Elias SG, Borel Rinkes IH, van Hillegersberg R. International survey on esophageal cancer: part I surgical techniques. Dis Esophagus. 2009;22(3)195-202.
5. Sarela AI, Tolan DJ, Harris K, Dexter SP, Sue-Ling HM. Anastomotic leakage after esophagectomy for cancer: a mortality-free experience. J Am Coll Surg. 2008;206(3)516-523.
6. Nederlof N, Tilanus HW, Tran TC, Hop WC, Wijnhoven BP, de Jonge J. End-to-End Versus End-to-Side Esophagogastrostomy After Esophageal Cancer Resection: A Prospective Randomized Study. Ann Surg. 2011;254(2)226-33.
7. Alanezi K, Urschel JD. Mortality secondary to esophageal anastomotic leak. Ann Thorac Cardiovasc Surg. 2004;10(2)71-75.
8. van Heijl M, Gooszen JA, Fockens P, Busch OR, van Lanschot JJ, van Berge Henegouwen MI. Risk factors for development of benign cervical strictures after esophagectomy. Ann Surg. 2010;251(6)1064-1069.
9. Kim RH, Takabe K. Methods of esophagogastric anastomoses following esophagectomy for cancer: A systematic review. J Surg Oncol. 2010;101(6)527-533.
10. Orringer MB, Marshall B, Iannettoni MD. Eliminating the cervical esophagogastric anastomotic leak with a side-to-side stapled anastomosis. J Thorac Cardiovasc Surg. 2000;119(2)277-288.
11. Dai JG, Zhang ZY, Min JX, Huang XB, Wang JS. Wrapping of the omental pedicle flap around esophagogastric anastomosis after esophagectomy for esophageal cancer. Surgery. 2011;149(3)404-410.
12. Carbon RT, Baar S, Kriegelstein S, Huemmer HP, Baar K, Simon SI. Evaluating the in vitro adhesive strength of biomaterials. Biosimulator for selective leak closure. Biomaterials. 2003;24(8)1469-1475.
13. Ozel SK, Kazez A, Akpolat N. Does a fibrin-collagen patch support early anastomotic healing in the colon? An experimental study. Tech Coloproctol. 2006;10(3)233-236.
14. de Stefano A, Bettarini F, Di Mare G, Neri A. Enteric anastomosis and Tachosil(R). Minerva Chir. 2011;66(3)183-188.
15. Cui Y, Urschel JD, Petrelli NJ. Esophagogastric anastomoses in rats--an experimental model. J Invest Surg. 1999;12(5)295-298.
16. Tadano S, Terashima H, Fukuzawa J, Matsuo R, Ikeda O, Ohkohchi N. Early postoperative oral intake accelerates upper gastrointestinal anastomotic healing in the rat model. J Surg Res. 2011;169(2)202-208.
17. Akgun A, Kuru S, Uraldi C, Tekin O, Karip B, Tug T, Ongoren AU. Early effects of fibrin sealant on colonic anastomosis in rats: an experimental and case-control study. Tech Coloproctol. 2006;10(3)208-214.
18. Shinohara K, Kobayashi E, Yoshida T, Toyama N, Kiyozaki H, Fujimura A, Miyata M. Effect of fibrin glue on small and large bowel anastomoses in the rat. Eur Surg Res. 1998;30(1)8-12.
19. Kanellos I, Mantzoros I, Goulimaris I, Zacharakis E, Zavitsanakis A, Betsis D. Effects of the use of fibrin glue around the colonic anastomosis of the rat. Tech Coloproctol. 2003;7(2)82-84.
20. van der Ham AC, Kort WJ, Weijma IM, van den Ingh HF, Jeekel H. Healing of ischemic colonic anastomosis: fibrin sealant does not improve wound healing. Dis Colon Rectum. 1992;35(9)884-891.
21. Cui Y, Urschel JD. Esophagogastric anastomotic wound healing in rats. Dis Esophagus. 1999;12(2)149-151.

22. Cui Y, Chen H. The effect of tension on esophagogastric anastomotic wound healing in rats. J Cardiovasc Surg . 2003;44(6)775-778.

23. Martens MF, Hendriks T. Postoperative changes in collagen synthesis in intestinal anastomoses of the rat: differences between small and large bowel. Gut. 1991;32(12)1482-1487.

24. Thompson SK, Chang EY, Jobe BA. Clinical review: Healing in gastrointestinal anastomoses, part I. Microsurgery. 2006;26(3)131-136.

25. Schreinemacher MH, Bloemen JG, van der Heijden SJ, Gijbels MJ, de Jong CH, Bouvy ND. Collagen fleeces do not improve colonic anastomotic strength but increase bowel obstructions in an experimental rat model. Int J Colorectal Dis. 2011;26(6)729-735.

26. Hoeppner J, Willa K, Timme S, Tittelbach-Helmrich D, Hopt UT, Keck T, Marjanovic G. Reinforcement of colonic anastomoses with a collagenous double-layer matrix extracted from porcine dermis. Eur Surg Res. 2010;45(2)68-76.

27. Stumpf M, Junge K, Rosch R, Krones C, Klinge U, Schumpelick V. Suture-free small bowel anastomoses using collagen fleece covered with fibrin glue in pigs. J Invest Surg. 2009;22(2)138-147.

28. Chmelnik M, Lasch L, Weih S, Wink E, Romero P, Holland-Cunz S. Anastomotic sealing with a fibrin-coated collagen patch in small-diameter bowel. Langenbecks Arch Surg. 2011;396(5)685-691.

29. van Doormaal TP, van der ZA, Verweij BH, Biesbroek M, Regli L, Tulleken CA. Experimental simplification of the excimer laser-assisted nonocclusive anastomosis (ELANA) technique. Neurosurgery. 2010;67(3 Suppl Operative)ons283-ons290.

30. Nordentoft T, Romer J, Sorensen M. Sealing of gastrointestinal anastomoses with a fibrin glue-coated collagen patch: a safety study. J Invest Surg. 2007;20(6)363-369.

31. Boone J, Borel Rinkes IH, van Leeuwen M, van Hillegersberg R. Diagnostic value of routine aqueous contrast swallow examination after oesophagectomy for detecting leakage of the cervical oesophagogastric anastomosis. ANZ J Surg. 2008;78(9)784-790.

32. Pierie JP, De Graaf PW, Poen H, van der Tweel I, Obertop H. Impaired healing of cervical oesophagogastrostomies can be predicted by estimation of gastric serosal blood perfusion by laser Doppler flowmetry. Eur J Surg. 1994;160(11)599-603.

33. Boyle NH, Pearce A, Hunter D, Owen WJ, Mason RC. Intraoperative scanning laser Doppler flowmetry in the assessment of gastric tube perfusion during esophageal resection. J Am Coll Surg. 1999;188(5)498-502.

34. Buise MP, Ince C, Tilanus HW, Klein J, Gommers D, van Bommel J. The effect of nitroglycerin on microvascular perfusion and oxygenation during gastric tube reconstruction. Anesth Analg. 2005;100(4)1107-1111.

35. van der Ham AC, de Jonge J, Buise MP, Specht P, van Genderen M, Gommers D. The effects of intravenous nitroglycerine and norepinephrine on gastric microvascular perfusion in an experimental model of gastric tube reconstruction. Surgery. 2010;148(1)71-77.

CHAPTER 5

Reduced local immune response with CPAP during single lung ventilation for esophagectomy

submitted

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ABSTRACT

Background

Transthoracic esophagectomy requires prolonged single lung ventilation causing systemic and local inflammatory response. Application of continuous positive airway pressure (CPAP) to the collapsed lung potentially reduces pulmonary damage, hypoxia, and consequent inflammation. This randomized controlled trial studied the influence of CPAP applied to the collapsed right lung during thoracoscopic esophagectomy on local and systemic inflammatory response.

Methods

Broncho-alveolar lavage fluid (BALF) from the right collapsed and left ventilated lung, and serum samples were obtained during surgery from 30 patients undergoing thoracoscopic esophagectomy for cancer who were randomized for complete right lung collapse or collapse with CPAP. Concentrations of cytokines and chemokines, in BALF and serum, were determined with Luminex.

Results

Patients from the control group had significantly increased concentrations of interleukin (IL)-1α, IL-1β, IL-10, tumor necrosis factor-alpha (TNFα), macrophage inflammatory protein (MIP)-1α, pulmonary and activation-regulated chemokine (PARC) and IL-8 in the right collapsed lung when compared to patients from the CPAP group ($p<0.050$). The left ventilated lung of the control group showed increased concentrations of monocyte chemoattractant protein (MCP)-1 and MIP-1α ($p<0.050$). Serum concentrations of cytokines and chemokines increased during surgery, but did not differ between the control and CPAP group.

Conclusions

A significantly lower local immune response was observed during single lung ventilation when CPAP was applied to the collapsed lung. The findings support the use of CPAP on the collapsed lung during esophagectomy with single lung ventilation.

INTRODUCTION

Esophagectomy is accompanied by significant morbidity and mortality. Reported pulmonary complications after transthoracic esophagectomy are in the range of 50-60%.[1] Mortality rates are the range of 1-5%, but pneumonia and respiratory failure after esophagectomy are associated with an increased mortality risk up to 20%.[2] Prolonged single lung ventilation, manipulation of the collapsed lung, extensive dissection of the mediastinum and reinsufflation of the collapsed lung are held responsible for the high incidence of respiratory complications. Various pathophysiological models for pulmonary complications after esophagectomy have been proposed.[3-6] A systemic inflammatory response syndrome (SIRS) probably plays an important role. A relationship between local (lung) and systemic (serum) inflammatory mediators and the development of pulmonary infections has been described.[5,6] Thoracic surgery, esophagectomy in particular, is accompanied by a local and systemic increase of pro- and anti-inflammatory mediators such as interleukin-6 (IL-6), IL-8, and IL-10.[7-10] Such response has been found to correlate with postoperative septic events.[11] The duration of surgery and amount of blood loss further influence the immunological response.[3] Pulmonary damage is caused by manipulation of the collapsed lung during surgery[3] and reinsufflation of the lung after surgery. Animal experiments have shown that reexpansion pulmonary edema (RPE) after reinsufflating a collapsed lung is associated with upregulation of IL-8, MCP-1[12], TNFα and IL-1β.[13] Other experiments have shown that positive end expiratory pressure (PEEP) during single lung ventilation leads to reduced lung damage.[14] Continuous positive airway pressure (CPAP), as a non invasive ventilation technique, has been proven to prevent or treat respiratory failure in the postoperative clinical setting.[15] The application of CPAP improves gas exchange, alveolar recruitment and lung capacity. In the operative setting, it can also be applied on the collapsed lung during single lung ventilation for thoracic surgery. CPAP potentially reduces peroperative hypoxia in the collapsed lung as well as mechanical stress by keeping the alveoli open. Theoretically, improved oxygenation and reduced trauma of prolonged collapse would translate into reduced inflammatory response during and after surgery.
The aim of this study was to examine whether CPAP on the collapsed right lung during thoracoscopic esophagectomy would reduce local and systemic cytokine production when compared to complete collapse.

METHODS

The study was performed in a tertiary referral center for esophageal cancer. The study protocol was approved by the institutional ethical review board and registered with the Netherlands National Trial Register (NTR645). Written consent was obtained from all participants before enrolment in the study.

PATIENTS

Eligible patients had histologically confirmed adenocarcinoma or squamous cell carcinoma of the mid-to-distal esophagus or adenocarcinoma of the gastric cardia involving the distal esophagus. Patients were candidates for thoracolaparoscopic esophagectomy with two-field lymphadenectomy. Evidence of unresectable local disease (cT4) or distant metastases (cM1b) excluded patients for surgery. Patients had to be older than 18 years of age and in adequate physical condition to undergo surgery. Exclusion criteria are shown in table 1. All other conditions or events were handled according to the intention to treat principle.

Table 1 Exclusion criteria

Exclusion criteria*
Preoperatively established
ASA score >3
FEV1<80%
Peroperatively established
Unsuccessful placement of epidural catheter
Insufficient lung collapse requiring conversion to transhiatal procedure
Extensive pleural adhesions requiring conversion to transhiatal procedure
Irresectability of tumor established during surgery

* All other conditions were handled according to the intention to treat principle. Definition of abbreviations: ASA = American Society of Anesthesiologists; FEV1 = Forced Expiratory Volume in 1 second.

ANESTHETIC MANAGEMENT

Anesthetic management was standardized including an epidural catheter for intra- and postoperative analgesic management between the 5th and 8th thoracic vertebrae. Patients were intubated with a left-sided double lumen tube (Broncho-Cath Left, Mallinckrodt Medical, Athlone, Ireland) under fiberoptic control. Cuff pressure was measured to prevent mucosal damage. General anesthesia was achieved with induction doses of propofol, sufentanil and rocuronium and maintained with continuous infusion of propofol, continuous infusion of atracurium and additional doses of sufentanil. Lung protective mechanical ventilation was pressure-controlled with a maximum pressure of 20 cm H_2O. During single lung ventilation, maximum pressure was tolerated up to 25 cm H_2O. Tidal volume was reduced to approximately 6 ml/kg predicted body weight and 5 cm H_2O PEEP was routinely used. The lowest possible fraction of inspired oxygen (FiO_2) was delivered to prevent oxidative damage and postoperative Acute Lung Injury with a minimum of 70% aiming at a SaO_2 of >92% (permissive hypoxia with a minimum of 88% SaO_2). Ventilation rate was adjusted whilst keeping $ETCO_2$ levels below 7.0% (permissive hypercapnia) aiming at CO_2 levels below 6.0%. Antibiotic prophylaxis was provided by intravenous administration of 2000 mg cefazolin and 500 mg metronidazole. Thirty minutes before incision, 10 mg/kg methylprednisolone (Pfizer, New York, NY, USA) was administered to minimize postoperative pulmonary complications.[16]

SURGICAL PROCEDURE

All patients underwent esophagolymphadenectomy through a robot assisted thoraco-laparoscopic approach as described previously.[17,18] For the thoracoscopic phase, patients were positioned in a left lateral decubitus position (tilted 45˚ towards prone position). Adequate tube placement was again verified by flexible bronchoscopy. At insertion of the thoracoscopic trocars, the right lung was selectively deflated using the double lumen tube. Thoracoscopic esophageal mobilization and lymphadenectomy were performed with the use of a robotic system (DaVinci, Intuitive Surgical Inc., Sunnyvale CA, USA). After the thoracic phase, the patient was positioned in the supine position for laparoscopic mobilization of the stomach and lymphadenectomy of truncal and perigastric nodes. The stomach was used for gastric conduit reconstruction with a cervical hand sewn end-to-side esophagogastrostomy.

POSTOPERATIVE CARE

Postoperatively, patients were transferred to the ICU department. Criteria for weaning from mechanical ventilation were hemodynamic stability without high dose positive inotropic or vasoconstrictive agents, core temperature >36°C, peripheral temperature >31°C, SaO_2 >94% with FiO_2 ≤40% and PEEP ≤8 cm H_2O, physiological respiratory impulse (respiratory frequency 10-20 per minute) and adequate consciousness.

COLLECTION OF PER- AND POSTOPERATIVE DATA

All operative and clinical parameters were prospectively collected in an MS Access database (Microsoft Corp., Redmond, WA, USA). Operative data were recorded during surgery and included duration of surgery and thoracoscopic dissection, and blood loss. Clinical parameters were recorded until discharge and included duration of length of ICU and hospital stay, duration of mechanical ventilation, and postoperative morbidity. Postoperative pneumonia was defined as an infiltrate on chest X-ray in combination with a positive sputum culture.

RANDOMIZATION AND INTERVENTION

Patients were randomly assigned to either the control group (complete right lung collapse) or the CPAP group (right lung collapse with CPAP). Randomization took place with sealed envelopes after placement of trocars for the thoracoscopic phase and before commencing dissection. In the CPAP group, positive pressure on the right lung was maintained at 5cm H_2O.

SAMPLE COLLECTION

EDTA-blood samples and selective broncho-alveolar lavage fluid (BALF) samples from the left and right lung were collected at the following moments: (t1) preoperatively, (i.e. directly after intubation); (t2) two hours after collapse of the right lung (i.e. two hours after

randomization); (t3) two hours after completely reinsufflating the right lung (i.e. two hours after ending the thoracic phase of the procedure); (t4) postoperatively after closure of incisions. Two additional blood samples were collected at (t5) 24 hours and (t6) 48 hours after completely reinsufflating the right lung (i.e. 22 and 46 hours after t3). The sequence of sampling is presented in figure 1B. For BALF collection, a bronchoscope was inserted selectively in the left lung first to the deepest bronchial level possible. Twenty milliliters of sterile 0.9% NaCl were instilled through the scope and aspirated by suction. The same procedure was repeated in the right lung. In between sampling, the bronchoscope was cleaned with sterile 0.9% NaCl. Collected blood and BALF samples were directly put on ice and transported to the laboratory. BALF samples were filtered during centrifugation at 1500 rpm for 3 minutes at 4˚C. Cells, debris and mucous were removed by centrifugation at 3000 rpm for 5 minutes. Blood samples were separated with centrifugation at 3000 rpm for 5 minutes. All samples were stored at -80˚C until the time of analysis.

CYTOKINE MEASUREMENTS

Cytokines and chemokines were measured using xMAP technology (Luminex Corporation, Austin, TX, USA) on a Bioplex 100 instrument as described previously.[19,20] Data analysis was performed with Bioplex Manager software version 4.1 (Bio-Rad Laboratories, Hercules, CA, USA). An eight point standard curve in duplicate, as well as appropriate controls, were included on every 96-well plate. Based on a pilot experiment in which a total of 27 cytokines and chemokines were analyzed, the following cytokines were measured: interleukin-1α (IL-1α), IL-1β, IL-6, IL-10, IL-12p70, tumor necrosis factor-alpha (TNFα), as well as the following chemokines: monocyte chemoattractant protein-1 (MCP-1, CCL2), macrophage inflammatory protein-1α (MIP-1α, CCL3), Regulated upon Activation, Normal T-cell Expressed, and Secreted (RANTES, CCL5), Eotaxin (CCL11), pulmonary and activation-regulated chemokine (PARC; CCL18), and IL-8 (CXCL8). All cytokine and chemokine concentrations are expressed in pg/ml.

STATISTICAL ANALYSIS

Primary outcome of the study for the power analysis was the postoperative IL-8 response with a reduction of response estimated at 50% (based on the results of the pilot experiment). For a two-tailed hypothesis with alpha set at 0.050 and 80% power the required sample size was 30 patients (15 per group) taking into account a 10% loss due to incomplete sampling. Because the nature of the surgical technique could change during the intervention, causing exclusions during surgery (e.g. exclusion based on irresectable tumor), an additional 20% of patients was included. All data were analyzed according to the intention to treat principle. Non-normally distributed cytokine concentrations were normalized by natural log transformation of absolute concentrations. The cytokine concentration change was measured as the change in concentration compared to the preoperative baseline measurement (t1). Differences in concentration change between

the CPAP group and control group at a specific sample moment were evaluated by Student *t* test. Longitudinal changes in cytokine levels were analyzed with repeated measurement analysis using a general linear model. Secondary outcome of the study was occurrence of postoperative pneumonia. Binary data was compared with the Chi-square test. Normally distributed continuous variables were analyzed with the Student *t* test, while non-parametric data was analyzed using the Mann-Whitney U test. *P values* <0.050 were considered statistically significant. All analyses were performed using standard statistical software (SPSS, Version 17.0, SPSS Inc. Chicago, IL, USA).

RESULTS

PATIENT CHARACTERISTICS

In total, 40 patients were randomized of which 10 patients were excluded intraoperatively (a flowchart and the reasons for exclusion are presented in figure 1A). Baseline characteristics are presented in table 2. The study population mainly consisted of males (83%). Mean age and BMI were comparable in both groups. No significant differences were found for ASA score and relevant medical history.

PEROPERATIVE COURSE

Conversion during thoracoscopy to thoracotomy was necessary in 2 patients (13%) of the control group and in 3 patients (20%) of the CPAP group (Chi-square test; p=0.624). Conversions in the CPAP group were related to limited visibility after bleeding (n=1), thoracic kyfosis limiting robot positioning (n=1) or suspicion of tumor ingrowth into the azygos vein (n=1). In 2 patients from the control group, conversion was necessary for inadequate surgical visibility (n=2). Operative time, thoracic dissection time and blood loss were comparable between the two groups (table 3). In the CPAP group, the median duration of CPAP was 105 (range 10-160) minutes. CPAP was discontinued in 4 patients during the course of surgery to improve visibility of the operative field. CPAP was paused in 4 patients and paused 3 times in 1 patient. In the 9 patients in whom CPAP was discontinued or paused, the median duration of CPAP was 75 (range 10-145) minutes. In the control group, 1 patient required CPAP on the collapsed lung because of oxygen desaturation during the thoracoscopic phase (CPAP time 155 minutes). The duration of CPAP and duration of complete single lung ventilation were significantly different between groups (table 3).

LUNG CYTOKINE PATTERNS

Data from BALF samples is presented in table 4. Figures 2 and 3 show the concentration changes in the left and right lungs for those cytokines and chemokines that showed a significant difference in change from baseline concentration at t4.

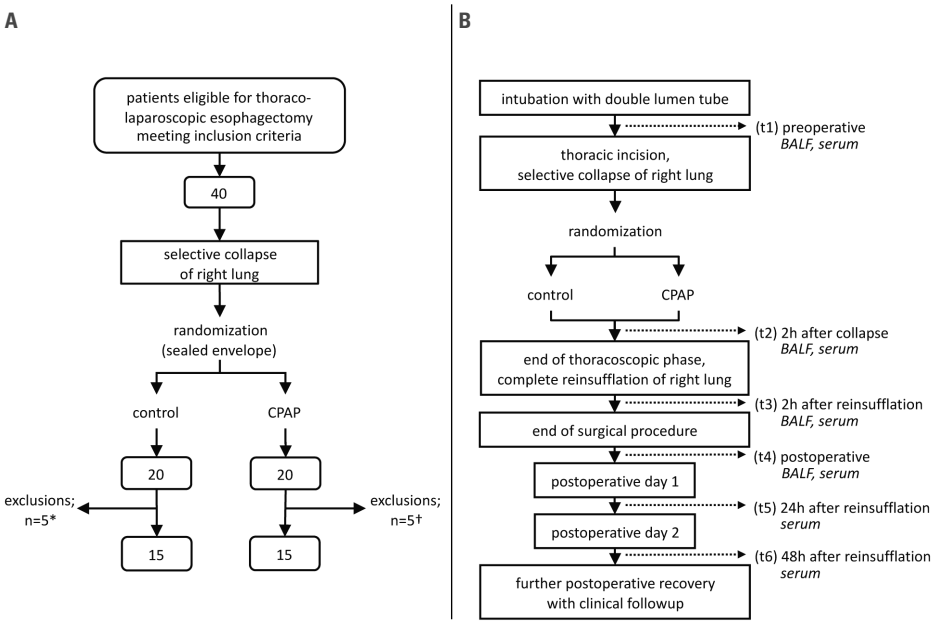

Figure 1 Trial design
Legend: A. Flowchart of trial design, inclusion and exclusion. B. Sequence of randomization and sampling. * Exclusions from the control group: Extensive pleural adhesions, converted to transhiatal procedure (n=1); Insufficient collapse of the right lung, converted to transhiatal procedure (n=2); Irresectable tumor (n=1); Gastrectomy instead of esophagectomy (n=1). † Exclusions from the CPAP group: Irresolvable hardware issues with robotic system, converted to transhiatal procedure (n=2); Extensive pleural adhesions, converted to transhiatal procedure (n=1); Irresectable tumor (n=2).

Right Lung (collapsed)

Concentration changes at t4 in the right lung were significantly lower in the CPAP group for IL-1α, IL-1β, IL-10, TNFα, MIP-1α, PARC and IL-8 (Student t-test; *P values* <0.050). There was a borderline different increase noticed for IL-6 (p=0.061). IL-12p70, MCP-1, RANTES and Eotaxin levels did not differ between the control and the CPAP group.

Left Lung (ventilated)

In the CPAP group, the concentration increase at t4 of MCP-1 and MIP-1α was significantly lower compared to the control group (p<0.050). Concentration changes at t4 in the left lung of IL-1α, IL-1β, IL-6, IL-10, IL-12p70, TNFα, RANTES, Eotaxin, PARC and IL-8 were not statistically different between the control and the CPAP group.

SERUM CYTOKINE PATTERNS

There was no statistical difference between the control and CPAP group in the change of serum concentrations of the tested cytokines at t4, t5 and t6 (table 5). Repeated

measurement analysis showed that *the moment of sampling* (t1-t6) was an independent factor influencing cytokine concentrations. Significant changes of cytokine concentrations over time were present for IL-1β, IL-6, IL-10, MCP-1, MIP-1α, RANTES, Eotaxin, PARC and IL-8 (repeated measurement analysis; all *P values* <0.010; data not shown).

CLINICAL OUTCOMES

Median length of ICU stay was 2 (1-40) days for the control group and 3 (1-65) days for the CPAP group (Mann-Whitney U test; p=0.492). The median number of ventilation days was equal in both groups as well (both 1 day, Mann-Whitney U test; p=0.761). Median hospital length of stay was 21 (10-80) days for control patients and 20 (12-87) days for CPAP patients (Mann-Whitney U test; p=0.724). Pneumonia occurred in 5 patients (33%) of the control group and in 4 patients (27%) of the CPAP group (RR .800, CI .266-2.410, Chi-square test; p=0.690).

Table 2 Baseline characteristics

		Control (n=15)	**CPAP (n=15)**	***P value****
Age (years)		64.4 (9.3)	61.7 (8.0)	0.402
Male		13 (86.7)	12 (80.0)	0.624
BMI (kg/m^2)		24.0 (3.7)	24.8 (2.6)	0.508
ASA score	1	4 (26.7)	8 (53.3)	0.270
	2	8 (53.3)	6 (40)	
	3	3 (20.0)	1 (6.7)	
Preoperative CTX		2 (13.3)	5 (33.3)	0.195
History of				
COPD		0 (0)	1 (6.7)	0.309
DM		2 (13.3)	2 (13.3)	1.000
Cardiologic disease		2 (13.3)	2 (13.3)	1.000
Malignant disease		4 (26.7)	1 (6.7)	0.142
Operation for malignancy		1 (6.7)	1 (6.7)	1.000
Laparotomy		0 (0)	0 (0)	1.000
Barrett's disease		0 (0)	2 (13.3)	0.143
Smoking		6 (40)	6 (40)	1.000

Data are n(%) or mean (SD).

* Mean values are analyzed with the Student t test, all other data with the Chi-square test. Definition of abbreviations: BMI = body mass index; ASA = American Society of Anesthesiologists; CTX = chemotherapy; COPD = chronic obstructive pulmonary disease; DM = diabetes mellitus.

Table 3 Per- and postoperative characteristics

	Control	CPAP	*P value**
Operative time total (minutes)	455 (349-505)	420 (321 – 545)	*0.648*
Thoracic dissection time (minutes)	170 (95-190)	150 (100-210)	*0.213*
Bloodloss total (milliliters)	350 (100-1400)	425 (200-3800)	*0.584*
Bloodloss thoracoscopy (milliliters)	175 (50-450)	200 (100-3050)	*0.440*
Duration of CPAP (minutes)†	0 (0-155)	105 (10-160)	***0.000***
Duration of complete single lung ventilation (minutes) ‡	170 (46-220)	55 (14-168)	***0.000***
LOS ICU (days)	2 (1-40)	3 (1-65)	*0.492*
Ventilation days (days)	1 (0-32)	1 (0-64)	*0.761*
LOS hospital (days)	21 (10-80)	20 (12-87)	*0.724*

Data are median (range). * *P value* of Mann-Whitney U test. † In the control group, CPAP was applied in 1 patient (155 minutes). ‡ In the CPAP group, CPAP was discontinued in 4 patients, paused in 4 patients, paused 3 times in 1 patient (all for reasons of surgical visibility). Definition of abbreviations: CPAP = continuous positive airway pressure; LOS = length of stay; ICU = intenstive care unit.

DISCUSSION

Surgical intervention inevitably leads to activation of the immune system.[21,22] Some studies have specifically described the immune response to thoracic surgery and esophagectomy.[3,5,23,24] However, only few longitudinal studies combine the analysis of systemic and local response in both the collapsed and ventilated lung separately during esophagectomy.[10] This is the first study demonstrating the influence of CPAP on the differential inflammatory response between ventilated and collapsed lungs.

The presented results show a significantly reduced local response of IL-1α, IL-1β, IL-10, TNFα, MIP-1α, PARC and IL-8 in the collapsed right lung when CPAP is applied during the thoracoscopic phase of esophagectomy. Furthermore, a reduced response of the lung specific chemokines MCP-1 and MIP-1α was observed in the left ventilated lung of patients who received CPAP on the right lung. The exact mechanism underlying the observed contralateral effect in the left lung of patients from the CPAP group is unclear. The left and right lungs form a single organ system and are simultaneously controlled by the same regulatory neural and hormonal systems. Pro- or anti-inflammatory processes in the collapsed lung may have triggered a similar response in the ventilated lung.[25]

Cree *et al*. report on the plasma and lavage concentrations of IL-8 and VEGF from postoperatively collected samples after esophagectomy. The authors compare the cytokine concentrations of the lavage fluid with the concentration in the plasma sample and report a significantly higher concentration of IL-8 and VEGF in BALF.[5] Moreover, it appears from their data that the IL-8 concentration was substantially higher in collapsed lungs when compared to ventilated lungs. In contrast, Zingg *et al*. report a trend towards higher IL-6 and IL-8 responses in the ventilated lung.[10] In our study we did not observe a

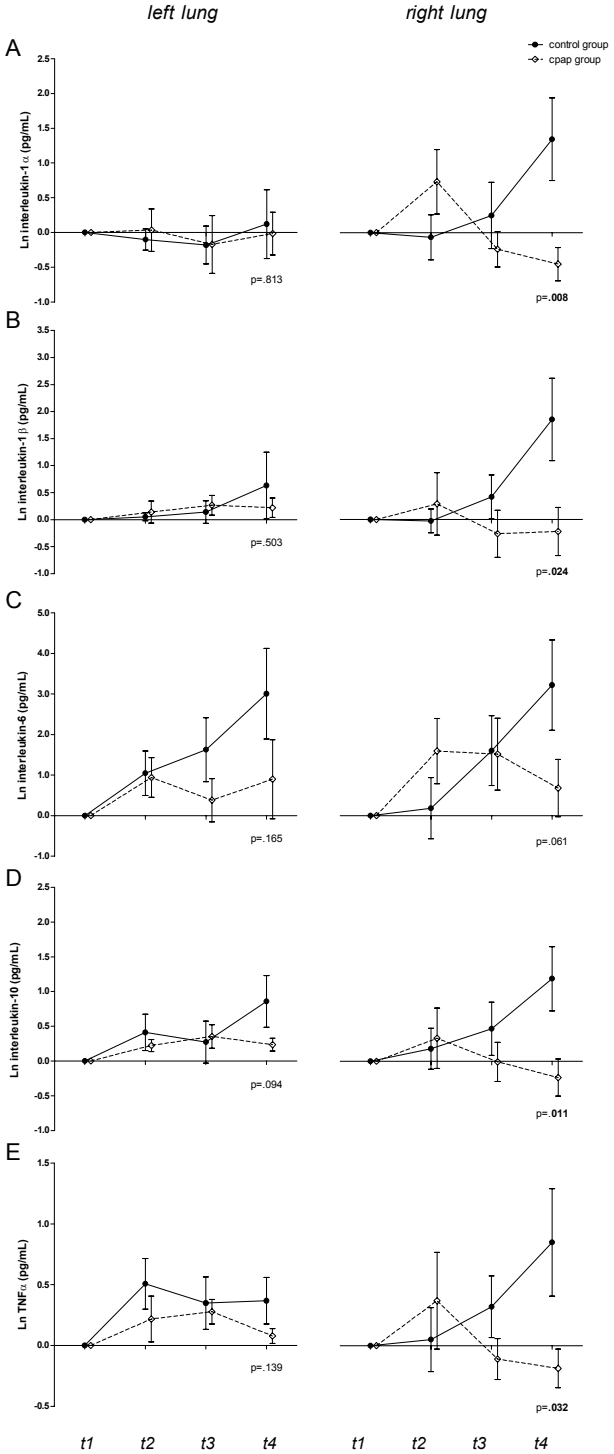

Figure 2 Longitudinal concentrations of cytokines (IL-1α, IL-1β, IL-6, IL-10 and TNFα) as change from baseline (preoperative) in the left and right lung of patients with and without CPAP.
Legend: Data represent the difference of the Ln concentration from t1 (mean ±SE). *P values* indicate the difference between the control group and the CPAP group at t4. Sample moments: t1 = preoperative; t2 = 2 hours after collapse; t3 = 2 hours after reinsufflation; t4 = postoperative.

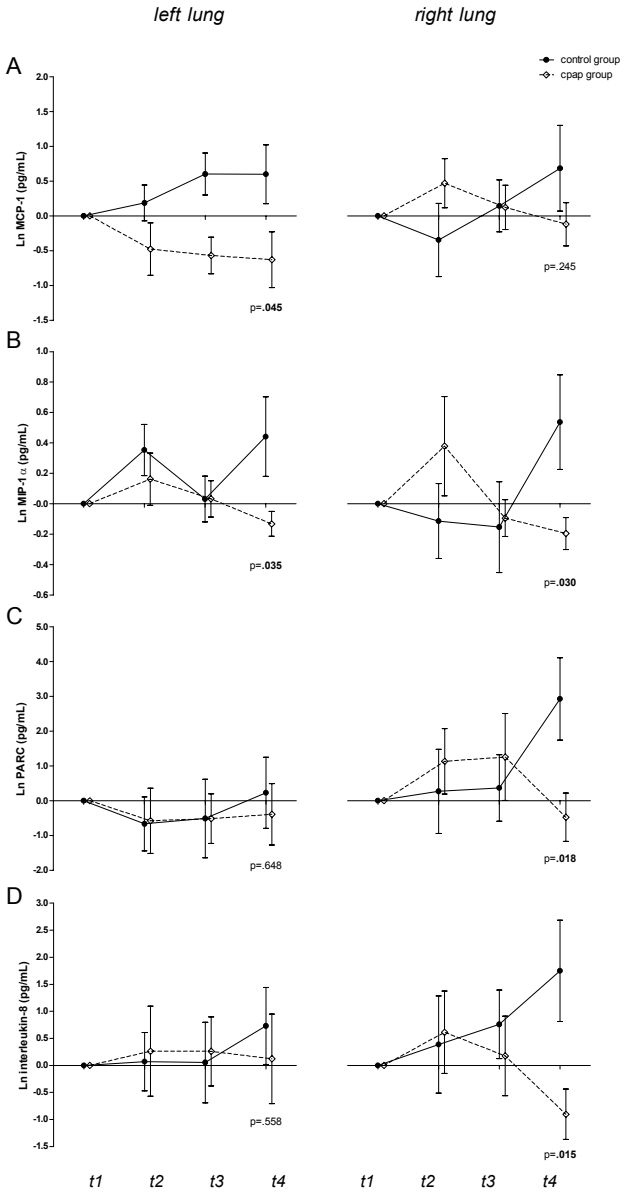

Figure 3 Longitudinal concentrations of chemokines (MCP-1, MIP-1α, PARC and IL-8) as change from baseline (preoperative) in the left and right lung of patients with and without CPAP.
Legend: Data represent the difference of the Ln concentration from t1 (mean±SE). *P values* indicate the difference between the control group and the CPAP group at t4. Sample moments: t1 = preoperative; t2 = 2 hours after collapse; t3 = 2 hours after reinsufflation; t4 = postoperative.

more pronounced response in the ventilated lung. The local immune response was most prominent in the collapsed lungs, particularly when compared to collapsed lungs which received CPAP.

The observed differences in local immune response between the control and CPAP group were most apparent at t4 (directly postoperatively). At t2 (2 hours after collapse), there is a trend towards increased cytokine response in the right lungs of patients who received CPAP. Hereafter, the response diminishes again (see the right columns of figures 2A-E and 3A-D). The right lungs of these patients had undergone desufflation followed shortly thereafter by mild insufflation when CPAP was applied. This might have caused the modest peak in cytokine response at t2 in CPAP patients. It must be noted though that there were no statistically significant differences between the control and CPAP group at t2 (data not shown). The modest t2 peak in the CPAP group or the absence of such a peak in the control group could possibly also be an artifact of the BALF sampling process. It could be argued that the right lungs of patients in the control group were less open at t2, due to complete collapse, compared to the lungs of patients in the CPAP group. We did not experience any difficulties with reaching the most distal airways with the sampling bronchoscope in neither of the two groups at t2. Moreover, a concern for possible sampling artifacts is not relevant for t4, where the difference of response was most pronounced. At t4 all patients were ventilated on both lungs.

The cytokine response pattern of the left and right lungs of patients in the control group was found to be different from that of the CPAP patients. In the control patients, there was a less distinct increase of pulmonary cytokines at t2, but a significant and ongoing increase was observed after reinsufflation of the right lung (t3-t4). This suggests that, within the given timeframe of the surgical procedure (t1-t4), the right lungs of patients from the control group experienced relatively more mechanical or hypoxic stress than the right lungs of patients who were randomized for CPAP.

The trend of a modest cytokine response after application of CPAP (i.e. mild insufflation) in the CPAP group, and the observation of a powerful local response after complete reinsufflation in the control group are in line with the hypothesis that pulmonary damage is caused by reinsufflation rather than by desufflation of the lung.[12,13] However, in the CPAP group, an distinct peak response to complete reinsufflation at the end of the thoracoscopic phase was absent (t3-t4). It appears that CPAP reduced the potential harmful mechanical consequences of complete reinsufflation after prolonged collapse. In addition, it could be argued that CPAP reduced the duration of hypoxia in the collapsed lung, thereby dampening the immune response to hypoxic stress. A combination of these two effects seems plausible.

In this study, we also measured the systemic inflammatory response in serum. The decreased inflammation in the right lung of CPAP patients did not correlate with a systemic suppression of inflammatory mediators. The systemic effect of surgical trauma was clearly noticed in both groups, demonstrated by significantly changing concentrations

of both pro- and anti-inflammatory cytokines over time in both groups (data not shown). It appears that the moderated local inflammatory response, as a result of CPAP, is unable to affect the surgery induced systemic response.

It has been described that an imbalance of pro- and anti-inflammatory response, leaning towards an overactive anti-inflammatory response leads to immune paralysis, which weakens host defense mechanisms and leads to organ failure.[26] In our study population, there was no noticeable effect of CPAP on the incidence of postoperative pneumonia. However, it must be noted that this study was primarily powered to demonstrate a reduction of cytokine response. To detect a significant effect on postoperative pneumonia or other clinical endpoints, larger (prospective) cohorts are required.

We have measured the local immune response in the lung by BALF sampling. A limitation of the applied method for acquisition of BALF lies within the retrieval of lavage fluid. Although frequently used in comparable studies, measured concentrations might vary between samples due to difficulties with the aspiration of lavage fluid.[24,27,28] In our hands the method for BALF collection was relatively easy to perform. Although the exact amounts of fluid were not measured during the study, we did not experience large fluctuations in retrieved BALF. Collection of exhaled breath condensate (EBC) is an allegedly more consistent and less invasive way to retrieve pulmonary fluids.[29,30] However, results from BALF and EBC do not correlate and the validity of this technique for the specific purpose of this study has yet to be confirmed.[31] Another alternative could be found in microdialysis.[32] Besides the possibility to acquire longitudinal data continuously, this technique has not proven superior to BALF.

The observed local inflammatory response in the collapsed lung pleads for an operative technique that does not require single lung ventilation. Several studies have reported on minimally invasive esophagectomy (MIE) in the prone position. In the prone position, a more dorsal approach is used compared to the lateral approach in the lateral decubitus position. The technique uses single lumen intubation instead of double lumen intubation. Positive pressure is maintained inside the thoracic cavity with CO_2 insufflation to induce collapse of the right lung, whilst maintaining ventilation of both lungs. Surgeons supporting this technique claim a reduced need for lung retraction and lower incidences of postoperative pneumonia.[33] However, this has not been demonstrated yet in well-designed comparative studies.[34] In addition, an important disadvantage of the prone position is the limited access in case of conversion to open surgery. With uncontrollable hemorrhage, there is only little time for conversion. The prone position limits the surgeon's ability to gain access to the thorax and control of the bleeding. Repositioning of the patient to the lateral decubitus is then required. We therefore prefer the left lateral decubitus position.

The application of CPAP disturbed visibility during 9 procedures. These mainly occurred temporarily during paratracheal and carinal dissection in the upper mediastinum. With the patient in the left lateral decubitus position, the partially inflated lung tended to move into

the operative field disturbing the surgeon's vision. More recently, we have overcome this by creating positive pressure inside the thoracic cavity with 8 mmHg CO_2 insufflation. The presented results show a significantly lower local immune response after single lung ventilation when CPAP is applied to the collapsed lung. This finding supports the use of CPAP during esophagectomy with single lung ventilation. Further studies are needed to address the correlation between clinical outcomes and the local inflammatory response to CPAP or double lung ventilation as opposed to complete single lung ventilation.

Table 4 Lung cytokine (ln) concentration at t4 (postoperatively) as difference from baseline in the left and right lung

	lung	Control Ln pg/mL		CPAP Ln pg/mL		*P value**
IL-1α	left	0.120	(0.495)	-0.015	(0.308)	*0.813*
	right	1.344	(0.593)	-0.455	(0.239)	***0.008***
IL-1β	left	0.631	(0.617)	0.220	(0.181)	*0.503*
	right	1.854	(0.763)	-0.221	(0.444)	***0.024***
IL-6	left	3.006	(1.117)	0.896	(0.976)	*0.165*
	right	3.218	(1.114)	0.677	(0.707)	*0.061*
IL-10	left	0.859	(0.371)	0.236	(0.091)	*0.094*
	right	1.190	(0.461)	-0.234	(0.267)	***0.011***
IL-12p70	left	0.019	(0.140)	0.199	(0.107)	*0.311*
	right	0.206	(0.217)	0.062	(0.046)	*0.506*
TNFα	left	0.368	(0.192)	0.079	(0.061)	*0.139*
	right	0.850	(0.443)	-0.187	(0.158)	***0.032***
MCP-1	left	0.601	(0.423)	-0.629	(0.402)	***0.045***
	right	0.684	(0.616)	-0.119	(0.311)	*0.245*
MIP-1α	left	0.441	(0.261)	-0.133	(0.081)	***0.035***
	right	0.536	(0.311)	-0.196	(0.105)	***0.030***
RANTES	left	-0.214	(0.471)	0.142	(0.147)	*0.451*
	right	-0.072	(0.373)	-0.014	(0.224)	*0.893*
Eotaxin	left	0.555	(0.737)	-0.325	(0.474)	*0.312*
	right	-0.005	(0.566)	-0.167	(0.241)	*0.790*
PARC	left	0.229	(1.019)	-0.390	(0.882)	*0.648*
	right	2.929	(1.186)	-0.476	(0.695)	***0.018***
IL-8	left	0.730	(0.712)	0.121	(0.827)	*0.588*
	right	1.749	(0.937)	-0.905	(0.465)	***0.015***

Data are mean (±SE) measured as Ln concentration difference from baseline (t1 - preoperatively) at t4 - postoperatively.
* *P value* of Student *t* test

Table 5 Serum cytokine concentrations as difference from baseline at t4-6

	t	Control		CPAP		*P* value*
		pg/mL		pg/mL		
IL-1α	t4	0.0	(0.0)	-2.8	(2.4)	*0.284*
	t5	0.1	(0.2)	-3.2	(2.9)	*0.261*
	t6	0.1	(0.2)	-1.9	(1.7)	*0.249*
IL-1β	t4	1.1	(0.6)	0.1	(0.1)	*0.127*
	t5	1.9	(1.1)	0.1	(0.1)	*0.130*
	t6	1.1	(0.6)	0.2	(0.2)	*0.185*
IL-6	t4	390.1	(170.6)	235.9	(75.1)	*0.405*
	t5	483.4	(348.6)	52.3	(20.3)	*0.227*
	t6	370.4	(237.4)	614.2	(445.4)	*0.633*
IL-10	t4	37.8	(12.4)	34.3	(6.6)	*0.799*
	t5	21.6	(21.2)	-0.2	(0.7)	*0.315*
	t6	5.8	(6.3)	8.5	(7.3)	*0.777*
IL-12p70	t4	945.6	(540.0)	81.3	(102.6)	*0.116*
	t5	1360.1	(886.9)	202.6	(121.5)	*0.207*
	t6	1030.4	(743.6)	217.5	(100.8)	*0.296*
TNFα	t4	5.9	(5.6)	-0.1	(0.2)	*0.274*
	t5	18.8	(18.6)	-0.7	(0.8)	*0.304*
	t6	8.9	(8.9)	-0.7	(0.9)	*0.291*
MCP-1	t4	-4.5	(4.0)	-14.2	(5.3)	*0.158*
	t5	25.0	(10.3)	27.4	(14.7)	*0.893*
	t6	50.5	(6.9)	59.7	(10.5)	*0.468*
MIP-1α	t4	-47.3	(46.0)	-28.8	(11.1)	*0.691*
	t5	159.0	(130.0)	94.2	(61.3)	*0.655*
	t6	96.9	(105.6)	216.0	(185.5)	*0.581*
RANTES	t4	-386.0	(744.8)	206.1	(127.2)	*0.425*
	t5	-1376.4	(1285.3)	-291.5	(231.7)	*0.413*
	t6	-1333.3	(1290.4)	-493.3	(254.2)	*0.528*
Eotaxin	t4	-44.0	(21.6)	-58.9	(7.8)	*0.509*
	t5	-31.7	(26.6)	-47.9	(7.9)	*0.565*
	t6	-20.1	(23.7)	-33.6	(5.9)	*0.554*
PARC	t4	-31060.1	(14543.2)	-10440.9	(4850.6)	*0.178*
	t5	-18555.4	(14686.2)	-1471.0	(8174.5)	*0.318*
	t6	-16095.6	(9123.7)	1015.8	(7331.3)	*0.155*
IL-8	t4	5.1	(2.5)	2.6	(0.5)	*0.325*
	t5	6.0	(3.3)	2.5	(0.6)	*0.310*
	t6	1.7	(8.6)	11.7	(8.6)	*0.267*

Data are mean (±SE) measured as concentration difference from baseline (t1 - preoperatively) at t4 - postoperatively; t5 - 24 hours after reinsufflation; and t6 - 48 hours after reinsufflation.* *P value* of Student *t* test

REFERENCES

1. Hulscher JB, van Sandick JW, de Boer AG, Wijnhoven BP, Tijssen JG, Fockens P, Stalmeier PF, ten Kate FJ, van Dekken H, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the esophagus. N Engl J Med. 2002;347(21)1662-1669.
2. Atkins BZ, Shah AS, Hutcheson KA, Mangum JH, Pappas TN, Harpole DH, D'Amico TA. Reducing hospital morbidity and mortality following esophagectomy. Ann Thorac Surg. 2004;78(4)1170-1176.
3. Yamada T, Hisanaga M, Nakajima Y, Kanehiro H, Watanabe A, Ohyama T, Nishio K, Sho M, Nagao M, Harada A, Matsushima K, Nakano H. Serum interleukin-6, interleukin-8, hepatocyte growth factor, and nitric oxide changes during thoracic surgery. World J Surg. 1998;22(8)783-790.
4. Tsukada K, Miyazaki T, Katoh H, Masuda N, Ojima H, Fukuchi M, Manda R, Fukai Y, Nakajima M, Sohda M, Kuwano H. Interferon-gamma and granulocyte colony-stimulating factor in bronchoalveolar lavage fluid after oesophagectomy. Dig Liver Dis. 2004;36(9)572-576.
5. Cree RT, Warnell I, Staunton M, Shaw I, Bullock R, Griffin SM, Baudouin SV. Alveolar and plasma concentrations of interleukin-8 and vascular endothelial growth factor following oesophagectomy. Anaesthesia. 2004;59(9)867-871.
6. Tsukada K, Hasegawa T, Miyazaki T, Katoh H, Yoshikawa M, Masuda N, Kuwano H. Predictive value of interleukin-8 and granulocyte elastase in pulmonary complication after esophagectomy. Am J Surg. 2001;181(2)167-171.
7. Sakamoto K, Arakawa H, Mita S, Ishiko T, Ikei S, Egami H, Hisano S, Ogawa M. Elevation of circulating interleukin 6 after surgery: factors influencing the serum level. Cytokine. 1994;6(2)181-186.
8. Sato N, Koeda K, Kimura Y, Ikeda K, Ogawa M, Saito K, Endo S. Cytokine profile of serum and bronchoalveolar lavage fluids following thoracic esophageal cancer surgery. Eur Surg Res. 2001;33(4)279-284.
9. Abe T, Oka M, Tangoku A, Hayashi H, Yamamoto K, Yahara N, Morita K, Tabata T, Ohmoto Y. Interleukin-6 production in lung tissue after transthoracic esophagectomy. J Am Coll Surg. 2001;192(3)322-329.
10. Zingg U, Forberger J, Frey DM, Esterman AJ, Oertli D, Beck-Schimmer B, Zollinger A. Inflammatory response in ventilated left and collapsed right lungs, serum and pleural fluid, in transthoracic esophagectomy for cancer. Eur Cytokine Netw. 2010;21(1)50-57.
11. Mokart D, Capo C, Blache JL, Delpero JR, Houvenaeghel G, Martin C, Mege JL. Early postoperative compensatory anti-inflammatory response syndrome is associated with septic complications after major surgical trauma in patients with cancer. Br J Surg. 2002;89(11)1450-1456.
12. Sakao Y, Kajikawa O, Martin TR, Nakahara Y, Hadden WA, Harmon CL, Miller EJ. Association of IL-8 and MCP-1 with the development of reexpansion pulmonary edema in rabbits. Ann Thorac Surg. 2001;71(6)1825-1832.
13. Funakoshi T, Ishibe Y, Okazaki N, Miura K, Liu R, Nagai S, Minami Y. Effect of re-expansion after short-period lung collapse on pulmonary capillary permeability and pro-inflammatory cytokine gene expression in isolated rabbit lungs. Br J Anaesth. 2004;92(4)558-563.
14. Gama de Abreu M, Heintz M, Heller A, Szechenyi R, Albrecht DM, Koch T, . One-lung ventilation with high tidal volumes and zero positive end-expiratory pressure is injurious in the isolated rabbit lung model. Anesth Analg. 2003;96(1)220-228.
15. Jaber S, Michelet P, Chanques G. Role of non-invasive ventilation (NIV) in the perioperative period. Best Pract Res Clin Anaesthesiol. 2010;24(2)253-265.
16. Sato N, Koeda K, Ikeda K, Kimura Y, Aoki K, Iwaya T, Akiyama Y, Ishida K, Saito K, Endo S. Randomized study of the benefits of preoperative corticosteroid administration on the postoperative morbidity and cytokine response in patients undergoing surgery for esophageal cancer. Ann Surg. 2002;236(2)184-190.
17. van Hillegersberg R, Boone J, Draaisma WA, Broeders IA, Giezeman MJ, Borel Rinkes IH. First experience with robot-assisted thoracoscopic esophagolymphadenectomy for esophageal cancer. Surg Endosc. 2006;20(9)1435-1439.
18. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8)878-886.

19. de Jager W, Prakken BJ, Bijlsma JW, Kuis W, Rijkers GT. Improved multiplex immunoassay performance in human plasma and synovial fluid following removal of interfering heterophilic antibodies. J Immunol Methods. 2005;300(1-2)124-135.

20. de Jager W, Bourcier K, Rijkers GT, Prakken BJ, Seyfert-Margolis V. Prerequisites for cytokine measurements in clinical trials with multiplex immunoassays. BMC Immunol. 2009;1052-62.

21. Wrigge H, Uhlig U, Zinserling J, Behrends-Callsen E, Ottersbach G, Fischer M, Uhlig S, Putensen C. The effects of different ventilatory settings on pulmonary and systemic inflammatory responses during major surgery. Anesth Analg. 2004;98(3)775-781.

22. Lin E, Calvano SE, Lowry SF. Inflammatory cytokines and cell response in surgery. Surgery. 2000;127(2)117-126.

23. Scheepers JJ, Sietses C, Bos DG, Boelens PG, Teunissen CM, Ligthart-Melis GC, Cuesta MA, van Leeuwen PA. Immunological consequences of laparoscopic versus open transhiatal resection for malignancies of the distal esophagus and gastroesophageal junction. Dig Surg. 2008;25(2)140-147.

24. Schilling T, Kozian A, Huth C, Buhling F, Kretzschmar M, Welte T, Hachenberg T. The pulmonary immune effects of mechanical ventilation in patients undergoing thoracic surgery. Anesth Analg. 2005;101(4)957-965.

25. West JB, Mathieu-Costello O. Stress failure of pulmonary capillaries: role in lung and heart disease. Lancet. 1992;340(8822)762-767.

26. Hietbrink F, Koenderman L, Rijkers GT, Leenen LP. Trauma: the role of the innate immune system. World J Emerg Surg. 2006;115-25.

27. Millo JL, Schultz MJ, Williams C, Weverling GJ, Ringrose T, Mackinlay CI, van der PT, Garrard CS. Compartmentalisation of cytokines and cytokine inhibitors in ventilator-associated pneumonia. Intensive Care Med. 2004;30(1)68-74.

28. Schultz MJ, Millo J, Levi M, Hack CE, Weverling GJ, Garrard CS, van der PT. Local activation of coagulation and inhibition of fibrinolysis in the lung during ventilator associated pneumonia. Thorax. 2004;59(2)130-135.

29. Mutlu GM, Garey KW, Robbins RA, Danziger LH, Rubinstein I. Collection and analysis of exhaled breath condensate in humans. Am J Respir Crit Care Med. 2001;164(5)731-737.

30. Horvath I, Hunt J, Barnes PJ, Alving K, Antczak A, Baraldi E, Becher G, van Beurden WJ, Corradi M, Dekhuijzen R, Dweik RA, Dwyer T, Effros R, Erzurum S, Gaston B, Gessner C, Greening A, Ho LP, Hohlfeld J, Jobsis Q, Laskowski D, Loukides S, Marlin D, Montuschi P, Olin AC, Redington AE, Reinhold P, van Rensen EL, Rubinstein I, Silkoff P, Toren K, Vass G, Vogelberg C, Wirtz H. Exhaled breath condensate: methodological recommendations and unresolved questions. Eur Respir J. 2005;26(3)523-548.

31. Jackson AS, Sandrini A, Campbell C, Chow S, Thomas PS, Yates DH. Comparison of biomarkers in exhaled breath condensate and bronchoalveolar lavage. Am J Respir Crit Care Med. 2007;175(3)222-227.

32. Tyvold SS, Solligard E, Gunnes S, Lyng O, Johannisson A, Gronbech JE, Aadahl P. Bronchial microdialysis of cytokines in the epithelial lining fluid in experimental intestinal ischemia and reperfusion before onset of manifest lung injury. Shock. 2010;34(5)517-524.

33. Palanivelu C, Prakash A, Senthilkumar R, Senthilnathan P, Parthasarathi R, Rajan PS, Venkatachlam S. Minimally invasive esophagectomy: thoracoscopic mobilization of the esophagus and mediastinal lymphadenectomy in prone position--experience of 130 patients. J Am Coll Surg. 2006;203(1)7-16.

34. Jarral OA, Purkayastha S, Athanasiou T, Zacharakis E. Should thoracoscopic three-stage esophagectomy be performed in the prone or left lateral decubitus position? Interact Cardiovasc Thorac Surg. 2011;13(1)60-65.

CHAPTER 6

A new clinical scoring system to define pneumonia following esophagectomy for cancer

submitted

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ABSTRACT

Background

Pneumonia is a frequently observed and feared complication following esophagectomy. The lack of a uniform definition of pneumonia in the non-ventilated patient leads to large variations of pneumonia rates in literature. This study was designed to identify the diagnostic determinants of pneumonia treatment and to develop a scoring system for diagnosing pneumonia following esophagectomy at the hospital ward.

Methods

In a prospective cohort study of esophagectomy patients, clinical data were collected including temperature, leukocyte count, pulmonary radiography and sputum culture added with clinical data and known risk factors for pneumonia. Primary outcome was defined as the decision to treat suspected pneumonia. Multivariate Cox regression analysis with backward selection was used to identify predictors of pneumonia treatment.

Results

The majority of postoperative pneumonia treatments (88.2%) occurred at the hospital ward, where treatment was observed in 67 (36.2%) of 185 patients. Independent diagnostic determinants for pneumonia treatment were temperature (HR=1.283, P=0.073), leukocyte count (HR=1.040, P=0.078) and pulmonary radiography (HR>11.0, P=0.000). Sputum culture did not influence the decision to treat pneumonia. These findings were used to develop a scoring system which includes temperature, leukocyte count and pulmonary radiography.

Conclusion

The decision to treat pneumonia is based on temperature, leukocyte count and pulmonary radiography findings. The proposed clinical scoring system for pneumonia following esophagectomy at the hospital ward has the potential to aid clinical practice and improve comparability of future research in esophageal cancer surgery.

INTRODUCTION

Esophagectomy for cancer is accompanied with high postoperative morbidity rates.[1-3] Respiratory complications, dominated by pneumonia, are most common (20-60%), associated with an increased risk of mortality (up to 5-10%) and the principle cause of postoperative death.[1,4,5] Furthermore, postoperative complications after esophagectomy have been reported to correlate with recurrence of disease.[6-8]

Pneumonia in hospitalized patients can be divided into ventilator-associated pneumonia (VAP) and hospital acquired pneumonia (HAP).[9-11] Pneumonia is often difficult to diagnose. Criteria with high sensitivity and low specificity, such as fever, leukocytosis, infiltrative abnormalities on pulmonary radiographies and bacterial growing on sputum culture, are used to establish the diagnosis.[10]

VAP is defined as pneumonia in patients receiving mechanical ventilation for at least 24 hours and can be diagnosed with the Clinical Pulmonary Infection Score.[12-16] HAP is defined as pneumonia in patients with a first positive bacterial respiratory culture finding >2 days from admission who do not meet the VAP criteria.[11] However, no clinical scoring system is available for diagnosing HAP in non-ventilated patients. The use of different definitions in literature limits interpretation and comparison of postesophagectomy pneumonia rates across studies.[17]

A frequently used traditional definition of pneumonia in clinical studies is the presence of infiltrative findings on pulmonary radiography combined with a positive sputum culture.[2,18] Other authors prefer a general classification of complications adapted to the respiratory system (Modified Clavien Dindo Classification; MCDC).[17,19]

In this prospective observational cohort study the objective was to define the diagnostic determinants that affected the decision to treat pneumonia. Furthermore, we aimed at developing a new scoring system for the definition of pneumonia after esophagectomy in non-ventilated patients.

PATIENTS AND METHODS

INCLUSION

A prospective database was maintained from October 2003 including all esophageal resections in a tertiary referral center (University Medical Center Utrecht, the Netherlands) as approved of by the institutional review board with patient consent. Database entries included standard patient characteristics with medical history and prospectively collected per- and postoperative data. All patients operated up to March 2011 were selected. Patients were included when they had undergone esophagectomy with gastric conduit reconstruction for esophageal cancer.

PROCEDURE

Patients underwent either transhiatal esophagectomy (THE), transthoracic esophagectomy (TTE) or minimally invasive robot-assisted thoracolaparoscopic esophagectomy (RATE). Enteral continuity was restored with a gastric conduit with a cervical esophagogastrostomy. All patients received bilateral chest tubes and a feeding jejunostomy.

Postoperatively, all patients were transferred to the intensive care unit (ICU). The criteria for weaning from mechanical ventilation were hemodynamic stability without high dose positive inotropic or vasoconstrictive agents, core temperature >36˚C, peripheral temperature >31°C, SaO2 >94% with FiO2 ≤40% and PEEP ≤8 cm H_2O, physiological respiratory impulse (respiratory frequency 10-20 per minute) and adequate consciousness. After weaning, patients were discharged to the hospital ward. Adequate nutritional intake was ensured through the feeding jejunostomy. Oral feeding was discontinued for one week without signs of anastomotic leakage. The jejunostomy remained in place during hospital stay and was removed in the outpatient clinic only after re-establishment of sufficient oral intake.

PRIMARY OUTCOME

In the absence of a reliable gold standard for diagnosing pneumonia, primary outcome was defined as *the decision to treat suspected pneumonia* (MCDC grade II, see table 1). The decision to treat pneumonia was cross referenced with registration of antibiotic medication use in the electronic medical records from the pharmacology department. Unless contra-indicated, patients diagnosed with pneumonia were treated intravenously with ceftriaxon 2000mg/day according to hospital protocol. If sputum culture indicated resistance of microorganisms to specific antibiotics, the antibiotic regimen was adjusted accordingly.

DATA COLLECTION

With the use of electronic medical records, the following 4 diagnostic determinants were collected which were registered during patients' stay at the MCU or the hospital ward; temperature (degrees Celsius), leukocyte count (n x10^9/L), pulmonary radiography findings and sputum culture (analogous to the CPIS criteria for VAP12).

The diagnostic determinants were performed at the moment during which pneumonia was clinically suspected and before commencing antibiotic treatment. In cases where there had been no suspicion of pneumonia, data were collected on the fourth day at the hospital ward to ensure a sufficient time from ICU discharge. The time-to-treatment of pneumonia at the hospital ward was calculated from the day of discharge from the ICU department.

STATISTICAL ANALYSIS

The association of each individual risk factor and diagnostic determinant with the decision to treat patients for pneumonia was examined in univariate analysis using separate Cox

regression models for each variable. Subsequently, those factors with a *P value* ≤0.20 in univariate analysis were selected for multivariate analysis, together with variables which were considered clinically relevant based on literature reports. The selected risk factors and diagnostic determinants were then entered in two separate multivariate Cox models, respectively, from which relevant variables were selected using AIC based backwards selection. Finally, two models were obtained; one model for risk factors and one model for diagnostic determinants.

Table 1 Classification of surgical complications with clinical examples of the respiratory system (as proposed by Dindo *et al.*[19] and adapted by D'Journo *et al.*[17])

Grade	Definition	Respiratory system
I	Any deviation from the normal postoperative course without the need for pharmacological treatment or surgical, endoscopic, and radiological interventions Allowed therapeutic regimens are drugs as antiemetics, antipyretics, analgesics, diuretics, electrolytes, and physiotherapy	Secretion retention or atelectasis requiring physiotherapy
II	Any deviation from the normal postoperative course requiring pharmacological treatment with drugs other than such allowed for grade I complications Blood transfusions and total parenteral nutrition are also included	Pneumonia treated with antibiotics on the ward
III	Any deviation from the normal postoperative course requiring surgical, endoscopic, or radiological intervention	Suction during bronchoscopy
IIIa	Intervention not under general anesthesia	
IIIb	Intervention under general anesthesia	
IV	Life-threatening complication requiring IC/ICU management	
Iva	Single-organ dysfunction (including dialysis)	Respiratory failure requiring endotracheal or non-invasive ventilation
IVb	Multi-organ dysfunction	Respiratory failure with failure of another organ
V	Death of a patient	Death

RESULTS

Between October 2003 and March 2011, a consecutive series of 206 patients underwent esophagectomy with gastric conduit reconstruction. Temperature recordings, leukocyte counts, pulmonary radiographies and sputum culture results were all retrieved from 185 patients. Patients were excluded in case of missing data for temperature (n=7), leukocyte count (n=6) or pulmonary radiography (n=8).

PNEUMONIA

Pneumonia was suspected and treated accordingly in 70 of 185 (37.8%) patients (table 2). During postoperative ICU stay, 9 (4.9%) patients were treated for pneumonia. At the surgical ward, 67 (36.2%) patients were treated for pneumonia. The latter group was used for further analysis.

The median time between the day of ICU discharge until the day at which pneumonia treatment was started at the hospital ward was 4 (range 0-21) days. Overall, the median hospital length of stay was 21 (range 10-105) days. Patients who were treated for pneumonia had a median hospital stay of 22 (range 10-182) days, compared to 15 (range 3-98) days for patients who were not treated for pneumonia at the hospital ward (Mann-Whitney U test, p=0.000).

A total of 34 (18.4%) patients were readmitted to the ICU department (MCDC grade IV). In most cases, 33 of 34 (97%) patients, this was due to respiratory failure. Patients who were treated for pneumonia at the hospital ward were at increased risk of getting

Table 2 Postoperative outcomes

				n (%) (total n=185)
Treated for pneumonia (*MCDC grade II*)		During total hospital stay		70 (37.8)
		During postoperative ICU stay		9 (4.9)
		During hospital ward stay		67 (36.2)
Time-to-treatment of pneumonia (days)*			median (range)	4 (0-21)
Anastomotic leakage				35 (18.9)
Chylus leakage				26 (14.1)
Wound infection	Neck			5 (2.7)
	Thorax			1 (0.5)
	Abdomen			5 (2.7)
Recurrent nerve	pareses			18 (9.7)
	paralysis			4 (2.2)
Atrial fibrillation				23 (12.4)
Myocardial infarction				4 (2.2)
In hospital mortality (*MCDC grade V*)				5 (2.7)
Length of stay (days)	ICU postoperative		median (range)	1 (1-65)
	ICU total		median (range)	2 (1-65)
	Hospital total		median (range)	21 (10-105)
Length of postoperative mechanical ventilation (days)			median (range)	0 (0-64)

Values are n (%) unless indicated otherwise. * Time-to-treatment of pneumonia at the hospital ward was calculated from the day of discharge from the ICU department. Abbreviations: MCDC - Modified Clavien Dindo classification, ICU - Intensive care unit.

readmitted to the ICU department when compared to patients who were not treated for pneumonia (35.8% vs. 8.5%, OR 6.0, Chi-square test p=0.000).
The in-hospital mortality rate related to pneumonia and respiratory failure (MCDC grade V) was 2.7% in the study population. In patients treated for pneumonia at the hospital ward, the in-hospital mortality rate was 4.5% compared to 1.7% among patients without pneumonia (OR 2.7, Chi-square test P=0.262).

REGRESSION ANALYSIS OF RISK FACTORS FOR PNEUMONIA

Descriptive data on the distribution of pre- and perioperative risk factors for pneumonia are presented in table 3. In the univariate Cox regression models, age, gender, history of COPD, BMI, neoadjuvant therapy and the number of resected lymph nodes were significantly associated with an increased risk of pneumonia treatment, when employing a liberal criterion of a *P value* ≤0.20.
Results from the multivariate Cox model, with AIC based backward selection of relevant predictors, are presented in table 4. The overall univariate *P value* for operative approach (transhiatal vs. transthoracic) was P=0.208. This variable was included in the multivariate Cox regression model (before commencing with the backwards selection) based on clinical relevance and its' well known association with pulmonary complications in literature[2,20] In the multivariate Cox model, obtained after backwards selection, variables significantly associated with treatment of pneumonia included transthoracic operative approach (Hazard Ratio=2.393, P=0.007), age (HR=1.034, P=0.027), male gender (HR=2.379, P=0.011), history of COPD (HR=1.985, P=0.037) and total number of resected lymph nodes (HR=0.972, P=0.025). BMI and neoadjuvant therapy were rejected from the regression model.

REGRESSION ANALYSIS OF DIAGNOSTIC DETERMINANTS FOR TREATMENT OF PNEUMONIA

Associations between temperature, leukocyte count, pulmonary radiography and sputum culture with treatment of pneumonia are presented in table 5. All four diagnostic determinants showed significant associations in univariate Cox regression analysis (all overall *P values* <0.001) and were consequently entered into multivariate analysis.
Sputum culture was excluded from the multivariate Cox regression model through backward selection (table 6). Temperature and leukocyte count remained in the model (HR=1.283, P=0.073; HR=1.040, P=0.078, respectively). Abnormal pulmonary radiography was an independent predictor of pneumonia treatment (P=0.000) with HRs of 11.5 and 13.4 for diffused infiltrate and well-circumscribed infiltrate, respectively.
A scoring model was created based on the outcomes of the multivariate analysis. Sputum culture was not included since it was already excluded from the regression model. Temperature recording and leukocyte count were included because they were borderline significant and because they can easily be obtained in the clinical setting. Analogous to

the CPIS scoring system[12], each diagnostic determinant was assigned a score (table 7) of which the sum yields an overall score ranging from 0 to 5 points. Figure 1 shows the total scores grouped by the negative or positive decision to treat patients for pneumonia. The pivot point of pneumonia treatment in this scoring model was at 2 points. Of all 66

Table 3 Univariate Cox regression analysis of risk factors for pneumonia

			Treated for pneumonia			
		Total n=185	Yes n=67	No n=118		
Risk factor		n (%)	n(%)	n (%)	HR	*P value*†
Age [years]	mean (SD)	63.9 (9.0)	65.4 (9.3)	63.0 (8.8)	1.02	***0.118***
Gender [male]		141 (76.2)	56 (83.6)	85 (72.0)	1.80	***0.059***
BMI	mean (SD)	25.5 (4.2)	26.2 (4.0)	25.2 (4.3)	1.04	***0.146***
COPD		23 (12.4)	12 (17.9)	11 (9.3)	1.72	***0.109***
Cardiac disease		37 (20.0)	16 (23.9)	21 (17.8)	1.28	*0.397*
Diabetes		26 (14.1)	11 (16.4)	15 (12.7)	1.18	*0.622*
Tobacco use		95 (51.4)	34 (50.7)	61 (51.7)	1.01	*0.969*
Alcohol use		102 (55.1)	41 (61.2)	61 (51.7)	1.30	*0.286*
Neoadjuvant therapy		71 (38.4)	30 (44.8)	41 (34.7)	1.41	***0.169***
ASA score 1		47 (25.4)	15 (22.4)	32 (27.1)	1.00	*0.764*
2		106 (57.3)	40 (59.7)	66 (55.9)	1.22	
3-4		32 (17.3)	12 (17.9)	20 (16.9)	1.28	
Operative approach [transthoracic extended]		134 (72.4)	52 (77.6)	82 (69.5)	1.43	*0.208*
Operative blood loss [milliliters]	median (range)	360 (6490)	360 (3770)	375 (6490)	1.00	*0.399*
Operative time [minutes]	median (range)	402 (522)	417 (412)	386 (501)	1.00	*0.417*
pT* 0-1		45 (24.3)	17 (25.4)	28 (23.7)	1.00	*0.790*
2		16 (8.6)	4 (6.0)	12 (10.2)	0.70	
3-4		124 (67.0)	46 (68.7)	78 (66.1)	0.97	
Total number of LNN resected [n]	median (range)	19 (63)	19 (45)	19 (63)	0.98	***0.164***
Postoperative ICU stay [days]	median (range)	1 (131)	1 (130)	1 (40)	1.01	*0.538*
Postoperative mechanical ventilation [days]	median (range)	0 (119)	0 (119)	0 (35)	1.01	*0.449*
Pneumonia during postoperative ICU stay		9 (4.9)	6 (9.0)	3 (2.5)	1.62	*0.294*

Values are n (%) unless indicated otherwise. * according to the TNM staging system, 7th edition. † variables with a *P value* <0.20 were selected for multivariate analysis. Abbreviations: HR - Hazard ratio, COPD - Chronic obstructive pulmonary disease, BMI - Body mass index, ASA - American Society of Anesthesiologists, pT - depth of tumor infiltration, pN - lymph node involvement, ICU - Intensive care unit.

patients with a score of 2 or higher, 58 patients were treated for pneumonia. In a subset of 30 patients with a sum score of 2 points, who were treated for pneumonia, 25 patients scored at least 1 point for infiltrative signs on pulmonary radiography (24 patients with a diffused infiltrate, and 1 patient with a well circumscribed infiltrate). The remaining 5 patients scored 1 point for temperature and 1 point for leukocyte count. Out of 119 patients scoring 0 or 1 points, 9 patients were treated for pneumonia.

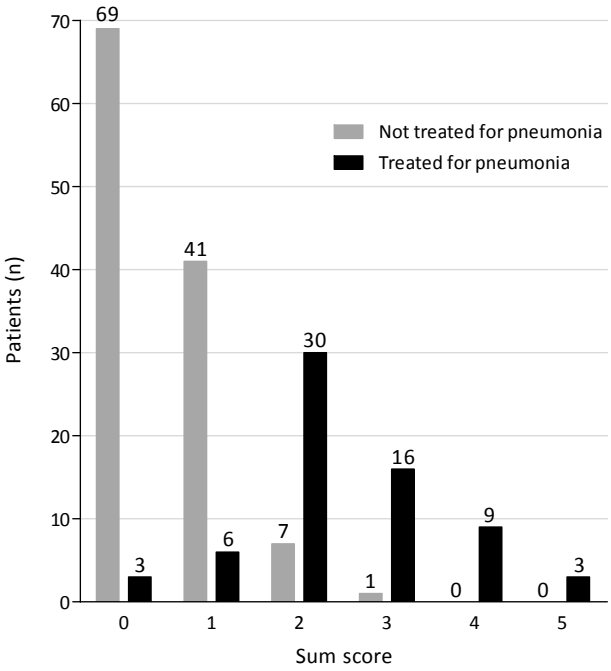

Figure 1 Treatment of pneumonia during hospital ward stay subdivided by the sum of scores

Table 4 Multivariate Cox regression analysis of risk factors for pneumonia

Risk factor	HR	P value
Operative approach [transthoracic extended]	2.393	***0.007***
Age	1.034	***0.027***
Gender [male]	2.379	***0.011***
History of COPD	1.985	***0.037***
Total number of LNN resected	0.972	***0.025***

Abbreviations: HR - Hazard ratio, COPD - Chronic obstructive pulmonary disease, LNN - lymph nodes.

Table 5 Univariate Cox regression analysis of diagnostic determinants for pneumonia

			Treated for pneumonia			
Diagnostic determinant		**Total n=185**	**Yes n=67**	**No n=118**	**HR**	***P value***
Temperature [°C]	median (range)	37.3 (4.6)	38.0 (4.6)	37.1 (3.0)	2.18	***0.000***
Leukocyte count [x10^9/L]	median (range)	11.3 (30.2)	14.4 (29.8)	9.7 (28.6)	1.10	***0.000***
Pulmonary radiography	No infiltrate	126 (68.1)	13 (10.3)	113 (89.7)	1.00	***0.000***
	Diffuse (or patchy) infiltrate	36 (19.5)	31 (86.1)	5 (13.9)	15.51	
	Well-circumscribed infiltrate	23 (12.4)	23 (100.0)	0 (0.0)	19.17	
Sputum culture	No sputum culture	41 (22.2)	11 (26.8)	30 (73.2)	1.00	***0.000***
	No PMO	77 (41.6)	19 (24.7)	58 (75.3)	0.89	
	PMO for pneumonia	59 (31.9)	29 (49.2)	30 (50.8)	1.88	
	PMO for pneumonia with corresponding gram stain	8 (4.3)	8 (100.0)	0 (0.0)	7.57	

Abbreviations: HR - Hazard ratio, PMO - pathogenic microorganism.

DISCUSSION

Esophagectomy is associated with a high risk of postoperative morbidity, mostly respiratory infections. Hence, studies on resection techniques in esophageal cancer surgery focus mainly on a reduction of morbidity, particularly respiratory complications and pneumonia. It is remarkable that these studies use different definitions of pneumonia[18,21] or fail to provide a definition altogether.[22-26] Consequently, the reported pneumonia rates vary widely. In this prospective series of patients who underwent esophageal resection with gastric conduit reconstruction, treatment of pneumonia (MCDC grade II) at the hospital ward was found in 36.2% of patients. Pneumonia was associated with respiratory failure and consequent readmission to the ICU (MCDC grade IV). When a different and frequently used definition of pneumonia is applied on our cohort (i.e. infiltrate on pulmonary radiography combined with a positive sputum culture[2,18]), the pneumonia rate decreases to 18.9%. It seems that a discrepancy exists between the diagnosis of pneumonia for clinical purposes and how pneumonia is potentially reported in literature. This illustrates the urgent need for a standardized approach towards pneumonia following esophagectomy.

Table 6 Multivariate Cox regression analysis of diagnostic determinants for pneumonia

Diagnostic determinant		HR	*P value*
Temperature		1.283	*0.073*
Leukocyte count		1.040	*0.078*
Pulmonary radiography	*No infiltrate*	1.000	***0.000***
	Diffused (or patchy) infiltrate	11.473	
	Well-circumscribed infiltrate	13.389	

Abbreviations: HR - Hazard ratio.

Table 7 Utrecht Pneumonia Scoring System for the decision to treat pneumonia at the hospital ward after esophagectomy *

Diagnostic determinant	Range	Score
Temperature [°C]	≥ 36.1 and ≤ 38.4	0
	≥ 38.5 and ≤ 38.9	1
	≥ 39.0 and ≤ 36.0	2
Leukocyte count [x10^9/L]	≥ 4.0 and ≤ 11.0	0
	< 4.0 or > 11.0	1
Pulmonary radiography	No infiltrate	0
	Diffused (or patchy) infiltrate	1
	Well-circumscribed infiltrate	2

* A sum score of 2 points or higher, of which at least 1 point is assigned due to infiltrative findings on pulmonary radiography, indicates treatment of pneumonia.

RISK FACTORS

In multivariate regression analysis of all relevant pre- and perioperative risk factors, a transthoracic operative approach was the strongest independent predictive factor followed by male gender and history of COPD. Increasing age was also associated with pneumonia treatment. The number of resected lymph nodes was negatively associated with postoperative pneumonia treatment (HR<1.0). Since lymph node harvest is generally higher during transthoracic surgery this was contrary to what we expected. Though significant, its' effect was minor.

The strong correlation of COPD with postoperative pneumonia treatment indicates that our results correspond with other studies, which report high predictive values of lung function and comorbidity for postoperative complications.[5,20] Operative approach (transhiatal vs. transthoracic) is also reported to be highly correlated to the development of pulmonary complications.[2,20] In accordance with other reports, ASA score and BMI were not associated with treatment of pneumonia in our series.[20,27]

Other authors have described nomograms and risk models to predict occurrence and/or severity of complications after esophagectomy.[5,20,28,29]

However, available prediction models suffer from low discriminative ability.[28,30] Pre- and perioperative risk factors play an important role, but the pathogenesis of postoperative complications is highly complex and dependent on a multitude of factors. Anesthesiological considerations, such as epidural analgesia and anesthetic management during surgery also influence postoperative outcomes.[31] Furthermore, genetic and immunological concepts may contribute in the development of complications as well.[32]

DIAGNOSTIC DETERMINANTS

Nomograms aid in risk stratification and patient selection for surgery, but do not facilitate the diagnostic process. Therefore, the second regression model focused on how diagnostic determinants for pneumonia had been used in clinical practice. Factors that play a role in the clinical decision-making process are general physical examination (chest auscultation, respiratory rate, coughing, presence and aspect of sputum, temperature) complemented with leukocyte count, arterial blood gas, pulmonary radiography and sputum culture.[10] The sensitivity of physical examination has been questioned in literature due to a high interobserver variability.[33] In this study it is impossible to assess its' contribution in the diagnostic process. C-reactive protein not only lacks specificity, but is highly responsive to major surgery and was therefore not included.[34] The determinants of pneumonia that could be objectively assessed are temperature recordings, leukocyte count, pulmonary radiography and sputum culture.

These four diagnostic variables were examined for their influence on the diagnostic process. Multivariate analysis showed a large effect of pulmonary radiography. It was the preferred or most important instrument for diagnosing pneumonia in clinical practice. Temperature and leukocyte also affected the decision to treat pneumonia. Although their effects appear minor, it must be noted that the risk increases exponentially with each incremental unit. This means that with an increase in leukocyte count of 20 units, the estimated risk (HR) of pneumonia treatment increases as $1.040^{20}=2.191$. Sputum culture was excluded during backward selection and did not independently correlate with the decision to treat pneumonia.

The findings correlate with the guidelines of the American Thoracic Society and the Infectious Diseases Society of America for the management of adults with HAP. The diagnosis of HAP is suspected if the patient has an infiltrate on pulmonary radiography which is new or progressive, along with clinical findings suggesting infection, such as fever, purulent sputum, leukocytosis, and decline in oxygenation.[10] The results of sputum culture, of which sensitivity is debatable10, become available after the decision to treat has already been taken. This is reflected in our findings which show that sputum culture is not independently associated with pneumonia treatment. It could be argued that pneumonia is overdiagnosed when sputum culture is not included in the decision process. Without a reliable gold standard it is impossible to adequately determine over- or

undertreatment rates. Though sputum culture does not influence the decision to treat pneumonia, it should always be performed to identify the responsible pathogen and to assess whether a switch of antibiotic medication is indicated.

SCORING MODEL FOR PNEUMONIA

To facilitate the diagnosis of VAP and improve comparability between clinical trials, the Clinical Pulmonary Infection Score (CPIS) is often used in clinical practice and literature.[12,13,35-40] Such a scoring system is not available for diagnosing HAP. Moreover, the majority of pneumonias in the studied cohort were diagnosed after discharge from the ICU. Hence, a standardized model for pneumonia at the hospital ward could be of great value in esophageal cancer research.

A clinical scoring model of the diagnostic determinants, analogous to the CPIS[12], was created based on multivariate Cox regression analysis. The model includes temperature, leukocyte count and pulmonary radiography, all easily obtainable from the non-ventilated patient. In contrast to the CPIS, no data was included on oxygenation (PaO_2/FiO_2 in mmHg) and tracheal secretions (number of required secretions) since these can only be assessed in mechanically ventilated patients. The presented scoring model accurately corresponds with the clinical practice of how pneumonia was diagnosed and treated accordingly in the studied series. It indicates that patients scoring 2 or higher were treated for pneumonia. The majority of patients scoring 2 points also had infiltrative signs on pulmonary radiography. Moreover, pulmonary radiography exercised the greatest effect (HR>11.0) in multivariate analysis. Based on these findings, we suggest a straightforward model which directs the decision to treat pneumonia after esophagectomy at the hospital ward. Patients with a sum score of 2 points or higher, of which at least 1 point is assigned due to infiltrative findings on pulmonary radiography, should be treated for pneumonia. Future studies should validate the utility of this model in the design, conduct, and evaluation of clinical research regarding pneumonia after esophagectomy.

CONCLUSION

In our series, a transthoracic operative approach, male gender and history of COPD were the strongest predictors for postoperative pneumonia treatment at the hospital ward. These risk factors are important for risk stratification and patient selection. However, the decision to treat pneumonia is composed by the outcomes of diagnostic determinants. In case of clinical suspicion of pneumonia after discharge from postoperative ICU stay, clinicians are guided by temperature, leukocyte count and pulmonary radiography findings. Sputum culture does not influence the decision to treat pneumonia, but is required to identify pathogens and appropriate antibiotic treatment.

The lack of a uniform definition leads to underreporting of pneumonia rates in literature.

We strongly advocate a standardized definition that includes the clinical decision to treat pneumonia. Pneumonia following esophagectomy is mostly diagnosed at the hospital ward and the decision to treat is based on temperature, leukocyte count and pulmonary radiography findings. After validation, the proposed scoring model has the potential to aid clinical practice and to improve comparability of future research in esophageal cancer surgery.

REFERENCES

1. Atkins BZ, Shah AS, Hutcheson KA, Mangum JH, Pappas TN, Harpole DH, Jr., D'Amico TA. Reducing hospital morbidity and mortality following esophagectomy. Ann Thorac Surg. 2004;78(4):1170-6.
2. Hulscher JB, van Sandick JW, de Boer AG, Wijnhoven BP, Tijssen JG, Fockens P, Stalmeier PF, Ten Kate FJ, van Dekken H, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the esophagus. N Engl J Med. 2002;347(21):1662-9.
3. Avendano CE, Flume PA, Silvestri GA, King LB, Reed CE. Pulmonary complications after esophagectomy. Ann Thorac Surg. 2002;73(3):922-6.
4. Dumont P, Wihlm JM, Hentz JG, Roeslin N, Lion R, Morand G. Respiratory complications after surgical treatment of esophageal cancer. A study of 309 patients according to the type of resection. Eur J Cardiothorac Surg. 1995;9(10):539-43.
5. Ferguson MK, Durkin AE. Preoperative prediction of the risk of pulmonary complications after esophagectomy for cancer. J Thorac Cardiovasc Surg. 2002;123(4):661-9.
6. Lerut T, Moons J, Coosemans W, van Sandick JW, De Leyn P, Decaluwe H, Decker G, Nafteux P. Postoperative complications after transthoracic esophagectomy for cancer of the esophagus and gastroesophageal junction are correlated with early cancer recurrence: role of systematic grading of complications using the modified Clavien classification. Ann Surg. 2009;250(5):798-807.
7. Takeuchi H, Saikawa Y, Oyama T, Ozawa S, Suda K, Wada N, Takahashi T, Nakamura R, Shigematsu N, Ando N, Kitajima M, Kitagawa Y. Factors influencing the long-term survival in patients with esophageal cancer who underwent esophagectomy after chemoradiotherapy. World J Surg. 2010;34(2):277-84.
8. Lagarde SM, de Boer JD, Ten Kate FJ, Busch OR, Obertop H, van Lanschot JJ. Postoperative complications after esophagectomy for adenocarcinoma of the esophagus are related to timing of death due to recurrence. Ann Surg. 2008;247(1):71-6.
9. Ewig S, Bauer T, Torres A. The pulmonary physician in critical care * 4: Nosocomial pneumonia. Thorax. 2002;57(4):366-71.
10. Guidelines for the management of adults with hospital-acquired, ventilator-associated, and healthcare-associated pneumonia. Am J Respir Crit Care Med. 2005 15;171(4):388-416.
11. Kollef MH, Shorr A, Tabak YP, Gupta V, Liu LZ, Johannes RS. Epidemiology and outcomes of health-care-associated pneumonia: results from a large US database of culture-positive pneumonia. Chest. 2005;128(6):3854-62.
12. Pugin J, Auckenthaler R, Mili N, Janssens JP, Lew PD, Suter PM. Diagnosis of ventilator-associated pneumonia by bacteriologic analysis of bronchoscopic and nonbronchoscopic "blind" bronchoalveolar lavage fluid. Am Rev Respir Dis. 1991;143(5 Pt 1):1121-9.
13. Singh N, Rogers P, Atwood CW, Wagener MM, Yu VL. Short-course empiric antibiotic therapy for patients with pulmonary infiltrates in the intensive care unit. A proposed solution for indiscriminate antibiotic prescription. Am J Respir Crit Care Med. 2000;162(2 Pt 1):505-11.
14. Kollef MH, Kollef KE. Antibiotic utilization and outcomes for patients with clinically suspected ventilator-associated pneumonia and negative quantitative BAL culture results. Chest. 2005;128(4):2706-13.
15. Rosbolt MB, Sterling ES, Fahy BG. The utility of the clinical pulmonary infection score. J Intensive Care Med. 2009;24(1):26-34.
16. Chastre J, Fagon JY. Ventilator-associated pneumonia. Am J Respir Crit Care Med. 2002;165(7):867-903.
17. D'Journo XB, Michelet P, Avaro JP, Trousse D, Giudicelli R, Fuentes P, Doddoli C, Thomas P. Respiratory complications after oesophagectomy for cancer. Rev Mal Respir. 2009;26:e41-51.
18. Biere SS, Maas KW, Bonavina L, Garcia JR, van Berge Henegouwen MI, Rosman C, Sosef MN, de Lange ES, Bonjer HJ, Cuesta MA, van der Peet DL. Traditional invasive vs. minimally invasive esophagectomy: a multi-center, randomized trial (TIME-trial). BMC Surg. 2011;11:2.

19. Dindo D, Demartines N, Clavien PA. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. Ann Surg. 2004;240(2):205-13.

20. Lagarde SM, Reitsma JB, Maris AK, van Berge Henegouwen MI, Busch OR, Obertop H, Zwinderman AH, van Lanschot JJ. Preoperative prediction of the occurrence and severity of complications after esophagectomy for cancer with use of a nomogram. Ann Thorac Surg. 2008;85(6):1938-45.

21. Briez N, Piessen G, Bonnetain F, Brigand C, Carrere N, Collet D, Doddoli C, Flamein R, Mabrut JY, Meunier B, Msika S, Perniceni T, Peschaud F, Prudhomme M, Triboulet JP, Mariette C. Open versus laparoscopically-assisted oesophagectomy for cancer: a multicentre randomised controlled phase III trial - the MIRO trial. BMC Cancer. 2011;11:310.

22. Doty JR, Salazar JD, Forastiere AA, Heath EI, Kleinberg L, Heitmiller RF. Postesophagectomy morbidity, mortality, and length of hospital stay after preoperative chemoradiation therapy. Ann Thorac Surg. 2002;74(1):227-31.

23. Walther B, Johansson J, Johnsson F, Von Holstein CS, Zilling T. Cervical or thoracic anastomosis after esophageal resection and gastric tube reconstruction: a prospective randomized trial comparing sutured neck anastomosis with stapled intrathoracic anastomosis. Ann Surg. 2003;238(6):803-12.

24. Okuyama M, Motoyama S, Suzuki H, Saito R, Maruyama K, Ogawa J. Hand-sewn cervical anastomosis versus stapled intrathoracic anastomosis after esophagectomy for middle or lower thoracic esophageal cancer: a prospective randomized controlled study. Surg Today. 2007;37(11):947-52.

25. Nederlof N, Tilanus HW, Tran TC, Hop WC, Wijnhoven BP, de JJ. End-to-end versus end-to-side esophagogastrostomy after esophageal cancer resection: a prospective randomized study. Ann Surg. 2011;254(2):226-33.

26. Kinjo Y, Kurita N, Nakamura F, Okabe H, Tanaka E, Kataoka Y, Itami A, Sakai Y, Fukuhara S. Effectiveness of combined thoracoscopic-laparoscopic esophagectomy: comparison of postoperative complications and midterm oncological outcomes in patients with esophageal cancer. Surg Endosc. 2012;26(2):381-90.

27. Grotenhuis BA, Wijnhoven BP, Hotte GJ, van der Stok EP, Tilanus HW, van Lanschot JJ. Prognostic value of body mass index on short-term and long-term outcome after resection of esophageal cancer. World J Surg. 2010;34(11):2621-7.

28. Steyerberg EW, Neville BA, Koppert LB, Lemmens VE, Tilanus HW, Coebergh JW, Weeks JC, Earle CC. Surgical mortality in patients with esophageal cancer: development and validation of a simple risk score. J Clin Oncol. 2006;24(26):4277-84.

29. Bartels H, Stein HJ, Siewert JR. Preoperative risk analysis and postoperative mortality of oesophagectomy for resectable oesophageal cancer. Br J Surg. 1998;85(6):840-4.

30. Grotenhuis BA, van Hagen P, Reitsma JB, Lagarde SM, Wijnhoven BP, van Berge Henegouwen MI, Tilanus HW, van Lanschot JJ. Validation of a nomogram predicting complications after esophagectomy for cancer. Ann Thorac Surg. 2010;90(3):920-5.

31. Liu S, Carpenter RL, Neal JM. Epidural anesthesia and analgesia. Their role in postoperative outcome. Anesthesiology. 1995;82(6):1474-506.

32. Sato N, Koeda K, Ikeda K, Kimura Y, Aoki K, Iwaya T, Akiyama Y, Ishida K, Saito K, Endo S. Randomized study of the benefits of preoperative corticosteroid administration on the postoperative morbidity and cytokine response in patients undergoing surgery for esophageal cancer. Ann Surg. 2002;236(2):184-90.

33. Wipf JE, Lipsky BA, Hirschmann JV, Boyko EJ, Takasugi J, Peugeot RL, Davis CL. Diagnosing pneumonia by physical examination: relevant or relic? Arch Intern Med. 1999;159(10):1082-7.

34. Dutta S, Fullarton GM, Forshaw MJ, Horgan PG, McMillan DC. Persistent elevation of C-reactive protein following esophagogastric cancer resection as a predictor of postoperative surgical site infectious complications. World J Surg. 2011;35(5):1017-25.

35. A'Court CH, Garrard CS, Crook D, Bowler I, Conlon C, Peto T, Anderson E. Microbiological lung surveillance in mechanically ventilated patients, using non-directed bronchial lavage and quantitative culture. Q J Med. 1993;86(10):635-48.

36. Fabregas N, Ewig S, Torres A, El-Ebiary M, Ramirez J, de La Bellacasa JP, Bauer T, Cabello H. Clinical diagnosis of ventilator associated pneumonia revisited: comparative validation using immediate post-mortem lung biopsies. Thorax. 1999;54(10):867-73.

37. Garrard CS, A'Court CD. The diagnosis of pneumonia in the critically ill. Chest. 1995;108(2 Suppl):17S-25S.

38. Fischer JE, Janousek M, Nadal D, Fanconi S. Diagnostic techniques for ventilator-associated pneumonia. Lancet. 1998;352(9133):1066-7.

39. Flanagan PG, Findlay GP, Magee JT, Ionescu A, Barnes RA, Smithies M. The diagnosis of ventilator-associated pneumonia using non-bronchoscopic, non-directed lung lavages. Intensive Care Med. 2000;26(1):20-30.

40. Schurink CA, Van Nieuwenhoven CA, Jacobs JA, Rozenberg-Arska M, Joore HC, Buskens E, Hoepelman AI, Bonten MJ. Clinical pulmonary infection score for ventilator-associated pneumonia: accuracy and inter-observer variability. Intensive Care Med. 2004;30(2):217-24.

PART TWO

PROGNOSIS

CHAPTER 7

How to define a positive circumferential resection margin in t3 adenocarcinoma of the esophagus

American Journal of Surgical Pathology. 2011;35(6):919-26

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ABSTRACT

Background

A positive circumferential resection margin (CRM+) is associated with poor survival following esophagectomy for cancer. The Royal College of Pathologists (RCP) defines a CRM+ when tumor is found <1 mm of the lateral margin whereas the College of American Pathologists (CAP) defines CRM+ when tumor cells are located at the lateral margin.

Methods and materials

This study evaluates the clinical prognostic significance of CRM+ on overall survival (OS) and disease free survival (DFS) in patients who underwent esophagectomy for T3 esophageal adenocarcinoma (EAC).

Results

Analysis included 132 patients. CRM+ was found in 26 cases (19.7%) corresponding to CAP criteria versus 89 cases (67.4%) corresponding to RCP criteria. Median OS using RCP criteria was 16.4 (95%CI 8.5-24.2) months for CRM+ patients versus 21.0 (95%CI 16.3-25.6) months in CRM- patients (p=.144). With CAP criteria, median OS in CRM+ and CRM- patients was 9.4 (95%CI, 7.6-11.2) months versus 21.6 (95%CI 18.9-24.3) months respectively (p=0.000). Median DFS using RCP criteria was 18.0 (95%CI 11.5-24.6) months for CRM- patients versus 11.0 (95%CI 8.1-14.0) months for CRM+ patients (p=.257). Applying CAP criteria, median disease free survival in CRM- and CRM+ patients was 16.3 (95%CI 10.6-22.0) months versus 7.0 (95%CI 6.3-7.8) months respectively (p=0.000). Effects of a positive CRM according to CAP criteria remained significant after multivariate testing (OS: HR 2.43, 95%CI 1.52-3.90; DFS: HR 2.09, 95%CI 1.32-3.30).

Conclusion

Only with the CAP criteria, CRM+ is an independent prognostic factor for survival and recurrence in patients with T3 adenocarcinoma of the esophagus. The circumferential margin should only be considered positive (R1) if tumor is found at the inked lateral margin of resection in accordance with the CAP criteria.

INTRODUCTION

Esophageal cancer is number eight on the list of most common cancers worldwide and sixth most common cause of death from cancer.[1] Parallel to the increased prevalence of obesity and reflux disease, esophageal adenocarcinoma (EAC) has become the most prevalent type of esophageal cancer, overtaking the rates of squamous cell carcinoma (SCC) in the Western World. In less than three decades, the incidence of EAC among Caucasian men increased with more than 450%.[2,3] Although improvements are made in both diagnostics and therapy, 5 year survival rates remain relatively poor. Survival is dependent mainly on disease stage. Definite staging is performed by histo-pathological examination, where tumor type, depth of invasion and lymph node involvement are identified.[4] Besides tumor stage, the residual tumor status (R-classification) is examined. The R-classification indicates the absence or presence of residual tumor in the resection margin after surgery.[5] R0 corresponds to resection for cure or complete remission; R1 to microscopic residual tumor; R2 to macroscopic residual tumor. For esophageal cancer the proximal, distal and circumferential resection margin can be involved. In rectal surgery, the R-classification has been demonstrated to be an important denominator for prognosis.[6,7] Residual tumor equals incomplete resection and hence an increased chance of tumor recurrence both locally and systemically. A similar influence on prognosis is seen in esophageal cancer.[8,9] Several studies have already demonstrated that a negative circumferential resection margin (CRM-) is associated with improved survival.[10-14] However, definition of a tumor positive circumferential resection margin (CRM+) remains unclear. Two major schools of pathologists differ in their assessment of the CRM. The Royal College of Pathologists (RCP) considers the CRM positive if tumor is found within 1 millimeter of the surgical margin whilst the College of American Pathologists (CAP) states that only tumor which is microscopically identified at the margin results in a positive CRM (figure 1).[15-17] Most studies investigating the association between CRM involvement and survival originated in the United Kingdom and merely assess the RCP criteria (table 1).[11-13,18,19] One of only few studies comparing RCP and CAP criteria, which was performed in the United States, demonstrated that a positive circumferential resection margin is of prognostic importance and suggested that the CAP criteria are clinically more meaningful.[10] The aim of this study is to evaluate the clinical prognostic significance of CRM involvement, according to CAP and RCP criteria, on both survival and recurrence of disease in patients with T3 adenocarcinoma of the esophagus.

METHODS AND MATERIALS

PATIENT SELECTION

All patients who underwent an esophageal resection with curative intent in the University

Medical Center Utrecht between 1988 and 2008 were collected in a database. Medical records were reviewed for patient characteristics, the use of neo-adjuvant therapy, surgical procedure, histological tumor-stage and both disease and survival status. Overall survival was defined as the time to death by any cause. Disease free survival was defined as the time to recurrence of both loco-regional and systemic disease. Disease- and survival-specific follow-up were retrieved by retrospective examination of medical records and consulting family physicians or the Dutch cancer registry (IKMN; Integraal Kanker Centrum Midden Nederland). All patients gave permission for the use of their medical data for research purposes at the time of pre-operative screening.

Table 1 Overview of published literature on circumferential resection margins in esophageal cancer surgery

	No. of patients	Histology	Neo-adjuvant therapy (%)*	T-stages	Review of path. slides?	R1 rate according to...		Difference in OS using CRM according to...	
						...CAP	*...RCP*	*...CAP†*	*...RCP†*
Sagar *et al.* (UK)[11]	50	all EC	0%	n/a	yes		40%	.	***yes***
Dexter *et al.* (UK)[12]	135	EAC, ESCC	n/a	1-3	no		47%	.	***yes***
Khan *et al.* (UK)[23]	329	EAC, ESCC	0%	1-3	no	12%	20%	.	no
	267	EAC, ESCC	0%	3				.	no
Griffiths *et al.* (UK)[13]	249	all EC	14%	1-3	no		32%	.	***yes***
	155	all EC	n/a	3			51%		
Sujendran *et al.* (UK)[18]	242	EAC, ESCC	62%	1-3	yes			.	.
	145	EAC, ESCC	61%	3			37%	.	***yes***
Deeter *et al.* (USA)[10]	135	all EC	44%	3	yes	12%	61%	***yes***	no
Saha *et al.* (UK)[19]	105	EAC	100%	1-4	yes		36%	.	***yes***
	72	EAC	100%	3-4			53%	.	***yes***
Scheepers *et al.* (NL)[14]	110	EAC, ESCC	28%	1-3	no	15%	38%	n/a‡	***yes***
	86	EAC, ESCC	36%	3		20%	49%	n/a‡	***yes***
Chao *et al.* (Taiwan/ China)[24]	151	ESCC	100%	3	yes	17%	51%	***yes***	no

* Neo-adjuvant therapy can include both chemotherapy and chemo-radiotherapy. † Only as reported in univariate analysis, a period (.) is indicated if the study did not analyze the effect of the specific CRM criteria on overall survival. ‡ Scheepers et al. did include both CAP and RCP criteria in their data, but did not perform a comparative analysis between the two definitions. Presented Kaplan-Meier curves suggest a significant difference for both CAP and RCP criteria. OS - overall survival; CAP - College of American Pathologists; RCP - Royal College of Pathologists; UK - United Kingdom; USA - United States of America; NL - the Netherlands; all EC - all types of esophageal carcinoma; EAC - esophageal adenocarcinoma; ESCC - esophageal squamous cell carcinoma; n/a - data not reported or not reproducible from article.

SURGICAL PROCEDURE

Operative approach was matched to patient physiology and tumor characteristics. Procedures included open transthoracic or transhiatal surgery. From 2003 a minimally invasive robot assisted thoracoscopic approach was also used.[20] The transthoracic approach was performed through right thoracotomy combined with a laparotomy and an intrathoracic or cervical anastomosis. Transhiatal surgery consisted of laparotomy or laparoscopy and a cervicotomy for the cervical anastomosis. Minimally invasive robot-assisted thoracoscopic esophagectomy was performed in conjunction with conventional laparoscopy and a cervical anastomosis. All thoracic approaches included a two field lymphadenectomy.

TISSUE HANDLING

Directly after resection, the surgical specimen was delivered to the pathology department. The specimen was opened longitudinally along the esophagus (from the proximal to the distal end) and along the gastric staple line. Thereafter, the specimen was pinned to a plate and fixated in formalin for at least 24 hours. After fixation took place, the circumferential, distal and proximal margins were inked. Tumors were enclosed completely in longitudinal slices of 3 millimeter. The slices where the tumor showed the closest proximity to the inked margin were embedded in paraffin blocks for standard diagnostic histological examination.

HISTOLOGICAL EXAMINATION

All original pathologic slides were retrieved and reassessed by a single experienced pathologist (FJWtK). In all specimens the minimal distance of the tumor to the nearest inked peri-esophageal dissection margin was measured microscopically in tenths of millimeters (figure 1). Furthermore, all specimens were reviewed for R classification (proximal and distal), lymph node (LN) involvement and the following tumor characteristics: histological type and grade, the presence of Barrett's mucosa, tumor location, tumor stage, vasoinvasive growth and perineural growth.

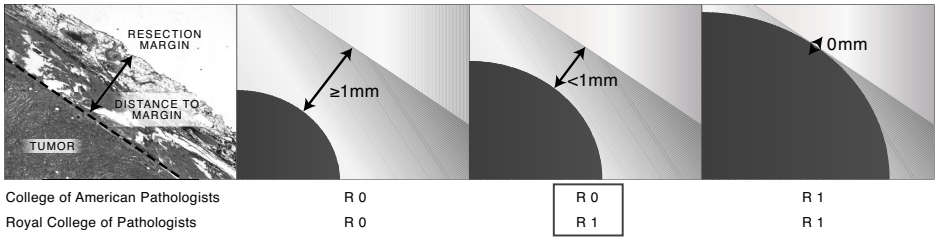

Figure 1 Definitions of the circumferential resection margin according to the criteria of the College of American Pathologists (CAP) and the Royal College of Pathologists (RCP)
Legend: The corresponding R-classification is denoted. Esophageal adenocarcinoma, H&E staining. R0 - no microscopic residual tumor; R1 - microscopic residual tumor; R2 - macroscopic residual tumor (not shown).

INCLUSION AND EXCLUSION CRITERIA

Since prognosis is highly influenced by disease stage and CRM involvement predominantly concerns advanced disease[13], only patients with T3 adenocarcinoma of the esophagus were included. Exclusion criteria were in-hospital mortality and the use of neoadjuvant chemo- or radiotherapy. These criteria were applied to yield a homogenous study population.

STATISTICAL ANALYSES

Data were analyzed using standard statistical software (SPSS, version 15.0; SPSS inc, Chicago, Illinois). Time to death and time to recurrence of disease were analyzed using survival analysis methods. Kaplan-Meier curves were used to estimate overall survival and time to recurrence. Log rank tests were applied to identify differences in survival or recurrence between groups. Univariate and multivariate Cox proportional hazard regression analyses were used to determine risk factors influencing survival or recurrence.
Investigated risk factors included the two different criteria of positive resection margin (CRM according to RCP and CRM according to CAP), the presence of Barrett's mucosa, vasoinvasive growth, perineural growth, the presence of tumor positive lymph nodes, lymph node ratio, the presence of more than four positive lymph nodes and perinodal extension. For each risk factor, the hazard ratio and associated 95% confidence interval from univariate analysis was reported. Multivariate analysis was conducted for those risk factors with significant *P values* (<0.05) from univariate analysis.

RESULTS

Two hundred sixty patients were operated on during the study period for an adenocarcinoma of the esophagus. Follow-up of 225 patients (86.5%) was completed. The final study population consisted of 167 patients with T3 EAC. Exclusions took place based on; in-hospital mortality (n=11), neoadjuvant therapy (n=13), and gastric tumor extending into the esophagus (n=3). After reassessment by the pathologist 8 cases were excluded for; T2 or T4 tumor stage instead of T3 (n=3), histological squamous cell carcinoma instead of adenocarcinoma (n=2) and missing or damaged pathology slides (n=3).
The final study population consisted of 132 patients; 112 (84.8%) men and 20 (15.2%) women. Median age was 62.6 years (range 33.8-82.9). Surgical approach included open transhiatal esophagectomy (n=91), laparoscopic transhiatal esophagectomy (n=1), transthoracic esophagectomy (n=19) and robot-assisted thoracolaparoscopic esophagectomy (n=21). The majority of tumors was located at the distal esophagus and gastroesophageal junction (n= 109, 82.6%). All patient and tumor characteristics are shown in table 2. Mean follow-up was 28.4 months ranging from 2.3 to 212.3 months. Positive circumferential margins were found in 89 cases (67.4%) corresponding to RCP

Table 2 Baseline characteristics (n=132)

		n (%)*
Gender	Male	112 (84.8)
	Female	20 (15.2)
Age (years)	Median (range)	62.6 (33.8 – 82.9)
Type of resection	Transthoracic (open)	19 (14.4)
	Transthoracic (scopic)	21 (15.9)
	Transhiatal (open)	91 (68.9)
	Transhiatal (scopic)	1 (0.8)
Localization	High/mid	2 (1.5)
	Distal	28 (21.2)
	GE junction	81 (61.4)
	Cardia	21 (15.9)
Differentiation grade	Well differentiated - G1	6 (4.5)
	Moderately differentiated - G2	66 (50.0)
	Poorly differentiated -G3	60 (45.5)
Tumor stage	IIA	15 (11.4)
	III	87 (65.9)
	IVA	26 (19.7)
	Unknown	4 (3.0)
Barrett's mucosa	no	81 (61.4)
	yes	51 (38.6)
Vasoinvasive growth	no	57 (43.2)
	yes	75 (56.8)
Perineural growth	no	77 (58.3)
	yes	55 (41.7)
LNN involvement	no	16 (12.1)
	yes	116 (87.9)
Perinodal extension†	no	39 (33.6)
	yes	77 (58.3)
LNN ratio >25%	no	72 (54.5)
	yes	60 (45.5)
No. of positive LNN>4	no	81 (61.4)
	yes	51 (38.6)
CRM status RCP	Negative - R0	43 (32.6)
	Positive - R1	89 (67.4)
CRM status CAP	Negative - R0	106 (80.3)
	Positive - R1	26 (19.7)

**numbers are indicated as: number of patients with percentage of patients, unless noted otherwise. † as a portion of the patients with lymph node involvement (n=116). LNN - lymph nodes*

criteria and in 26 cases (19.7%) corresponding to CAP criteria. The rate of CRM involvement did not differ between transthoracic and transhiatal surgery for both RCP (62.5% vs. 69.6%; Chi-square test; p=.426) and CAP criteria (15.0% vs. 21.7%; Chi-square test; p=.371).

Barrett's mucosa was present in 51 cases (38.6%) and 75 patients (56.8%) had vascular space involvement. Perineural growth was found in 55 patients (41.7%). Median harvest of lymph nodes was 15 (range 3-70). One hundred sixteen patients (87.9%) showed lymph node involvement with a median number of 4 tumorpositive lymph nodes (range 0-31). Lymph node ratio (number of positive nodes divided by the total number of resected lymph nodes) was less than or equal to 25% in 72 patients (54.5%) and 60 patients (45.5%) had more than 25% positive lymph nodes. Perinodal extension was observed in 66.4% of the patients with lymph node involvement.

OVERALL SURVIVAL

Figure 2 shows the overall survival grouped by resection margin status using both CRM definitions. Median overall survival was 19.5 (95% CI 16.7-22.3) months. Median overall survival using RCP criteria was 21.0 (95% CI 16.3-25.6) months for CRM-negative patients versus 16.4 (95% CI 8.5-24.2) months for CRM-positive patients (log rank p=.144). When analyzed according to the CAP criteria, median overall survival in CRM-negative and CRM-positive patients was 21.6 (95% CI 18.9-24.3) months versus 9.4 (95% CI 7.6-11.2) months respectively (log rank p=0.000). Univariate Cox proportional hazard model indicated a significant relationship between overall survival and CRM according to CAP, the presence of Barrett's mucosa, vascular space involvement, presence of positive lymph nodes, presence of 4 or more positive lymph nodes and a lymph node ratio of 25% (table 3). The CRM criteria according to RCP and perineural growth were not significantly related to overall survival. Multivariate analyses of the above mentioned significant prognostic factors confirmed positive resection margin using the CAP criteria as an independent predictor for overall survival (p=0.000, HR 2.43, 95% CI 1.52-3.90). Vasoinvasive growth also remained an independent prognostic factor (p=0.013, HR 1.73, 95% CI 1.12-2.66). The presence of Barrett's mucosa and the presence of positive lymph nodes did not prove significant after multivariate analysis.

DISEASE FREE SURVIVAL

Figure 3 shows the disease free survival grouped by resection margin status using both CRM criteria. Median disease free survival was 13.0 (95% CI 9.4-16.7) months. Median disease free survival using RCP criteria was 18.0 (95% CI 11.5-24.6) months for CRM-negative patients versus 11.0 (95% CI 8.1-14.0) months for CRM-positive patients (log rank p=.257). When analyzed according to the CAP criteria, median disease free survival in CRM-negative and CRM-positive patients was 16.3 (95% CI 10.6-22.0) months versus 7.0 (95% CI 6.3-7.8) months respectively (log rank p=0.000).

Univariate Cox proportional hazard model identified a significant relationship between disease free survival and CRM according to CAP, vascular space involvement, presence of positive lymph nodes, presence of 4 or more positive lymph nodes and a lymph node ratio of 25% (table 4). The CRM criteria according to RCP, the presence of Barrett's mucosa and perineural growth did not show a significant relationship with disease free survival.

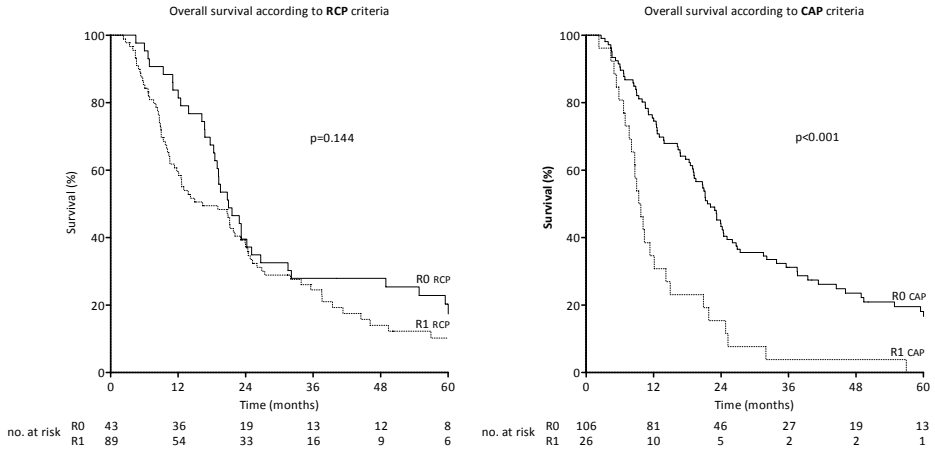

Figure 2 Kaplan-Meier curves of overall survival comparing patients with positive versus negative circumferential resection margin according to RCP and CAP criteria

Table 3 Univariate and multivariate analysis of the association between histopathologic parameters and overall survival

	Median OS in months		Unadjusted HR (95% CI)	*P value*	Adjusted HR (95% CI)*	*P value*
	negative	***positive***	***(univariate)***		***(multivariate)***	
CRM *CAP criteria*	21.6	9.4	2.734 (1.739-4.299)	***0.000***	2.433 (1.519-3.895)	***0.000***
CRM *RCP criteria*	21.0	16.4	1.343 (0.902-1.999)	*0.146*		
Barrett's mucosa	19.0	23.2	0.674 (0.456-0.998)	***0.049***	0.801 (0.528-1.216)	*0.298*
Vasoinvasion	25.2	16.2	2.103 (1.420-3.115)	***0.000***	1.729 (1.122-2.663)	***0.013***
Perineural growth	21.2	14.9	1.242 (0.848-1.818)	*0.266*		
Positive LNN	32.0	19.2	1.882 (1.023-3.461)	***0.042***	1.426 (0.710-2.864)	*0.319*
LNN ratio >25%	24.8	12.5	2.328 (1.588-3.413)	***0.000***	1.375 (0.849-2.226)	*0.196*
Positive LNN >4	23.9	11.3	2.142 (1.456-3.150)	***0.000***	1.336 (0.833-2.144)	*0.230*

*multivariate analysis is performed with variables proven significant in univariate analysis. OS - overall survival; CI - confidence interval; HR - hazard ratio; CRM - circumferential resection margin; CAP - College of American Pathologists; RCP - Royal College of Pathologists; LNN - lymph nodes

Multivariate analyses of the above mentioned prognostic factors confirmed positive resection margin using the CAP criteria as an independent predictor for disease free survival (p<0.05, HR 2.09, 95% CI 1.32-3.30). Vascular space involvement also remained an independent prognostic factor (p<0.05, HR 1.84, 95% CI 1.18-2.87).
The presence of Barrett's mucosa and the presence of positive lymph nodes did not prove significant after multivariate analysis.

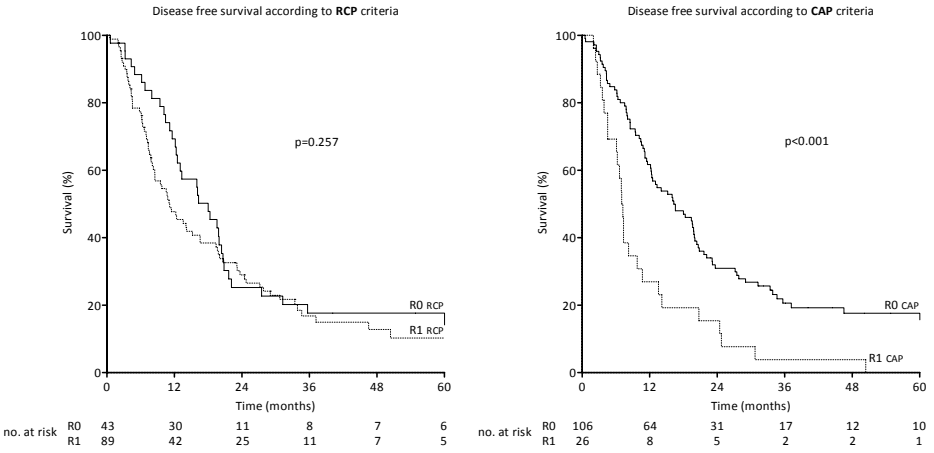

Figure 3 Kaplan-Meier curves of disease free survival comparing patients with positive versus negative circumferential resection margin according to RCP and CAP criteria

Table 4 Univariate and multivariate analysis of the association between histopathologic parameters and disease free survival

	Median DFS in months		Unadjusted HR (95% CI)	*P value*	Adjusted HR (95% CI)*	*P value*
	negative	***positive***	***(univariate)***		***(multivariate)***	
CRM CAP criteria	16.3	7.0	2.279 (1.458-3.562)	***0.000***	2.086 (1.320-3.296)	***0.002***
CRM RCP criteria	18.0	11.0	1.261 (0.844-1.885)	*0.258*		
Barrett's mucosa	13.0	15.2	0.909 (0.619-1.335)	***0.627***		
Vasoinvasion	21.7	9.8	2.280 (1.535-3.385)	***0.000***	1.844 (1.184-2.872)	***0.007***
Perineural growth	16.1	11.0	1.346 (0.921-1.967)	*0.125*		
Positive LNN	20.8	12.3	2.273 (1.180-4.378)	***0.014***	1.839 (0.886-3.817)	*0.102*
LNN ratio >25%	19.4	9.4	2.149 (1.468-3.147)	***0.000***	1.089 (0.671-1.767)	*0.730*
Positive LNN >4	19.4	8.2	2.413 (1.641-3.550)	***0.000***	1.540 (0.958-2.475)	*0.074*

*multivariate analysis is performed with variables proven significant in univariate analysis. DFS -disease free survival; CI - confidence interval; HR - hazard ratio; CRM - circumferential resection margin; CAP - College of American Pathologists; RCP - Royal College of Pathologists; LNN - lymph nodes

DISCUSSION

After surgical resection of esophageal cancer, a tumor positive resection margin (proximal, distal and circumferential) is related to reduced survival.[13,21,22] A negative circumferential resection margin is associated with improved survival and in some studies reported as the most important prognostic factor.[22] However, there is a lack of consensus regarding the criteria of a positive circumferential resection margin leading to the use of different definitions of tumor positive margins in the literature.

The present study covered a homogenous study population, focusing only on T3 tumors since CRM involvement is almost exclusively seen in these tumors. The presence of positive CRM in T1-2 carcinoma should be considered as technically inadequate surgical resection. Furthermore, tumor histology was intentionally limited to adenocarcinomas of the esophagus while many other studies included both EAC and SCC of the esophagus.[10-14,23]

In our study, reassessment of pathologic slides was performed by a single pathologist with great experience en special interest in esophageal pathology, guaranteeing consistency in interpretation and measurements. To eliminate the effect of neoadjuvant therapy on the resection margin, none of the included patients received chemo- or radiotherapy. However, it must be noted that in the era of neoadjuvant treatment for esophageal cancer a decrease in positive CRM is likely to be observed.[18,19]

This study is partly comparable to the paper by Deeter *et al*. In their study, only the CAP criteria showed to be an independent prognostic factor for survival in T3 esophageal cancer. No significant difference in survival between CRM negative and CRM positive patients was noted when using the RCP criteria.[10] However, Deeter *et al*. selected patients with both EAC and SCC. In addition, a substantial number of their patients received adjuvant chemo- or chemoradiotherapy (44%). It remains unclear from their data how this has influenced CRM involvement. They did not control for neo-adjuvant therapies or histologic tumor types in their multivariate analysis. In contrast, our study specifically examined a uniform cohort including only adenocarcinoma and excluding confounding influence of neo-adjuvant therapies.

This study demonstrates a significant difference in median survival between patients with positive and negative CRM measured according to the CAP criteria. No significant difference in median survival was seen in patients with positive tumor margins when using the RCP criteria. These results confirm conclusions from the previously discussed paper of Deeter *et al*,. and support earlier studies by Khan and Griffiths in less homogeneous patient series.[10,13,23] Moreover, the current study shows improved disease specific outcome (DFS) when applying the CAP criteria, which further demonstrates the significance from an oncological point of view.

Table 1 presents a summary of the available literature and illustrates the methodological heterogeneity regarding histology, tumor grade and the use of confounding neo-adjuvant therapy. In contrast to the study of Deeter *et al*., several studies with a specific focus on

circumferential resection margin indicate the RCP guideline for CRM as an independent prognostic factor.[11-14,18]. Griffiths *et al*. described a significant difference of survival using the RCP criteria among 249 patients with T1-3 esophageal carcinomas (median survival 37 months for CRM- versus 18 months for CRM+, p<0.01), but this effect was not reproducible in the subgroup of patients with T3 tumors.[13] Another large study by Khan *et al*. did not find a difference in survival with RCP criteria in both the overall T1-3 group of patients (n=329) nor in the subgroup of T3 patients (n=267).[23] None of these studies, mainly initiated from the United Kingdom, provided a comparison between the RCP and CAP criteria and only few performed multivariate analysis of their results.

In a recently published series of patients with squamous cell carcinoma of the esophagus, who received neo-adjuvant chemoradiotherapy, the CAP criteria also proved to be a significant discriminator for survival.[24] More importantly, the authors also included recurrence of disease in their analysis. Results showed equal rates of locoregional recurrence in patients with tumor <1 mm and tumor at 0 mm of the margin. However, it remains unclear whether the CAP criteria were of independent prognostic value. None of the other previously published studies have presented results on recurrence of disease, most probably due to incomplete retrospective follow-up data. Our results regarding DFS in patients with adenocarcinoma confirm the conception that recurrence of disease is dependent on a tumor free margin, but is not significantly altered when tumor cells are found within 1 mm from the resection margin.

The use of the RCP criteria is derived from studies of CRM involvement in rectal cancer where CRM is considered positive when tumor cells are found within 1 mm of the margin. It has been shown that this CRM definition is one of the most important predictive factors for recurrence and survival after surgery for rectal cancer.[6,7] However, caution should be taken when extrapolating these findings to esophageal cancer.

From an anatomical point of view, an explanation for the difference between the results for esophageal cancer versus rectal cancer could be found in the amount of connective tissue surrounding the respective organs. Both the esophagus and rectum are embedded in adventitia (in contrast to intraperitoneal organs which are covered by serosa). However, the rectum resides in more adventitia and is lined with the mesorectum. Instead, the esophagus lacks such an anatomical boundary. The esophagus is in very close proximity to vital mediastinal organs such as the trachea, heart and aorta. Hence, esophageal tumors encounter only a small anatomic barrier to local invasion. Furthermore, surgical en-bloc resection of the esophagus is limited by these structures. When rectal tumor cells are found in close range to the margin (i.e. within 1mm), it implies that the disease has already progressed through the wide manchet of mesorectum with subsequent poor survival.

Nodal metastatic burden (lymph node ratio <25% or >25%) was a significant determinant for OS and DFS in univariate analysis, but not an independent factor in multivariate analysis. Reports in the literature regarding lymph node ratio are conflicting. Some studies show a lymph node ratio to be of prognostic significance.[10,13,25] Others could not draw

this conclusion.[26] This study indicates that there is a significant relationship between a lymph node ratio >25% and survival. However, this could not be confirmed in multivariate analysis. The difficulty in assessing the prognostic value of lymph node ratio most probably resides in the applied surgical technique and the extent of lymphadenectomy. In our series, the number of resected nodes in transhiatal surgery (open and laparoscopic combined) was significantly lower than the number of nodes harvested in a transthoracic approach (mean 14.8 vs. 29.4 respectively, t-test $p<0.001$). Other studies confirm this difference.[27] However, a much less prominent difference is found when looking at the number of positive lymph nodes. Therefore, the average lymph node ratio after transhiatal surgery (this study, 37%) is higher than after transthoracic surgery (current study; 21%, t-test $p<0.01$). A lower lymph node ratio in a patient operated through a transthoracic route does not necessarily imply less advanced disease. Simply, more nodes have been resected when compared to the relatively limited lymphadenectomy in transhiatal surgery including only the lower mediastinal nodes. Therefore, the use of lymph node ratio as a prognostic marker should be cautiously applied when operative approach is not taken into consideration.

Although a discussion on the optimal surgical approach is beyond the scope of this study, it could be argued that operative approach might affect CRM involvement. Therefore, data was also analyzed for influence of operative approach on CRM. There was no difference in CRM involvement when comparing transhiatal and transthoracic surgery for both RCP and CAP criteria. In addition, there was no correlation between operative approach and survival. Consequently, it can be concluded that the observed survival data of the studied cohort is affected by CRM involvement regardless of operative approach.

Only one of the examined studies included the effect of vasoinvasion in their analysis.[19] Corresponding to the results of Saha *et al.*[19], our results show that OS and DFS are affected by vasoinvasive growth of tumor cells in multivariate analysis. As expected, the invasion of tumor cells into the vasculature is highly correlated to haematogenic spreading of disease.

In search of a meaningful prognostic tool, there clearly is a lack of agreement with respect to CRM involvement. For reliable prognostication of survival and recurrence, comparability of clinical studies and surgical quality monitoring it is important to internationally comply with one definition of a positive surgical resection margin for esophageal adenocarcinoma. The presented results show a significant relationship with decreased prognosis when tumor cells are found at the circumferential margin of the resected specimen. However, no influence on prognosis was found when tumor reaches within 1 millimeter of the margin. Positive CRM according to CAP criteria showed to be the strongest independent prognostic factor for overall and disease free survival and has to be considered as the clinical standard for patients with T3 adenocarcinoma of the esophagus.

REFERENCES

1. Parkin DM, Bray F, Ferlay J, Pisani P. Global Cancer Statistics, 2002. CA Cancer J Clin. 2005;5574-108.
2. Hongo M, Nagasaki Y, Shoji T. Epidemiology of esophageal cancer: Orient to Occident. Effects of chronology, geography and ethnicity. J Gastroenterol Hepatol. 2009;24(5)729-735.
3. Cook MB, Chow WH, Devesa SS. Oesophageal cancer incidence in the United States by race, sex, and histologic type, 1977-2005. Br J Cancer. 2009;101(5)855-859.
4. Sarwar SCM, Luktich JD, Landerneau RJ, Abbas G. Esophageal Cancer: An update. Int J Surg. 2010;8417-422.
5. Hermanek P, Wittekind C. The pathologist and the residual tumor (R) classification. Pathol Res Pract. 1994;190(2)115-123.
6. Wibe A, Rendedal PR, Svensson E, Norstein J, Eide TJ, Myrvold HE, Soreide O. Prognostic significance of the circumferential resection margin following total mesorectal excision for rectal cancer. Br J Surg. 2002;89(3)327-334.
7. Adam IJ, Mohamdee MO, Martin IG, Scott N, Finan PJ, Johnston D, Dixon MF, Quirke P. Role of circumferential margin involvement in the local recurrence of rectal cancer. Lancet. 1994;344(8924)707-711.
8. Law S, Arcilla C, Chu KM, Wong J. The significance of histologically infiltrated resection margin after esophagectomy for esophageal cancer. Am J Surg. 1998;176(3)286-290.
9. Mulligan ED, Dunne B, Griffin M, Keeling N, Reynolds JV. Margin involvement and outcome in oesophageal carcinoma: a 10-year experience in a specialist unit. Eur J Surg Oncol. 2004;30(3)313-317.
10. Deeter M, Dorer R, Kuppusamy MK, Koehler RP, Low DE. Assessment of criteria and clinical significance of circumferential resection margins in esophageal cancer. Arch Surg. 2009;144(7)618-624.
11. Sagar PM, Johnston D, McMahon MJ, Dixon MF, Quirke P. Significance of circumferential resection margin involvement after oesophagectomy for cancer. Br J Surg. 1993;80(11)1386-1388.
12. Dexter SP, Sue-Ling H, McMahon MJ, Quirke P, Mapstone N, Martin IG. Circumferential resection margin involvement: an independent predictor of survival following surgery for oesophageal cancer. Gut. 2001;48(5)667-670.
13. Griffiths EA, Brummell Z, Gorthi G, Pritchard SA, Welch IM. The prognostic value of circumferential resection margin involvement in oesophageal malignancy. Eur J Surg Oncol. 2006;32(4)413-419.
14. Scheepers JJ, van der Peet DL, Veenhof AA, Cuesta MA. Influence of circumferential resection margin on prognosis in distal esophageal and gastroesophageal cancer approached through the transhiatal route. Dis Esophagus. 2009;22(1)42-48.
15. Dataset for the histopathological reporting of oesophageal carcinoma, 2nd edition. Document G006. 2007.
16. Edge SB, Byrd DR, Compton CC, et al. AJCC Cancer Staging Manual. 2010.
17. Surgical Pathology Cancer Case Summary (Checklist): Esophagus. 2005.
18. Sujendran V, Wheeler J, Baron R, Warren BF, Maynard N. Effect of neoadjuvant chemotherapy on circumferential margin positivity and its impact on prognosis in patients with resectable oesophageal cancer. Br J Surg. 2008;95(2)191-194.
19. Saha AK, Sutton C, Rotimi O, Dexter S, Sue-Ling H, Sarela AI. Neoadjuvant chemotherapy and surgery for esophageal adenocarcinoma: prognostic value of circumferential resection margin and stratification of N1 category. Ann Surg Oncol. 2009;16(5)1364-1370.
20. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8)878-886.
21. Holscher AH, Bollschweiler E, Bumm R, Bartels H, Hofler H, Siewert JR. Prognostic factors of resected adenocarcinoma of the esophagus. Surgery. 1995;118(5)845-855.
22. Saha S, Dehn TC. Ratio of invaded to removed lymph nodes as a prognostic factor in adenocarcinoma of the distal esophagus and esophagogastric junction. Dis Esophagus. 2001;14(1)32-36.

23. Khan OA, Fitzgerald JJ, Soomro I, Beggs FD, Morgan WE, Duffy JP. Prognostic significance of circumferential resection margin involvement following oesophagectomy for cancer. Br J Cancer. 2003;88(10)1549-1552.

24. Chao YK, Yeh CJ, Chang HK, Tseng CK, Chu YY, Hsieh MJ, Wu YC, Liu HP. Impact of Circumferential Resection Margin Distance on Locoregional Recurrence and Survival after Chemoradiotherapy in Esophageal Squamous Cell Carcinoma. Ann Surg Oncol. 2011;18(2)529-534.

25. Mariette C, Piessen G, Briez N, Triboulet JP. The number of metastatic lymph nodes and the ratio between metastatic and examined lymph nodes are independent prognostic factors in esophageal cancer regardless of neoadjuvant chemoradiation or lymphadenectomy extent. Ann Surg. 2008;247(2)365-371.

26. Rice TW, Blackstone EH, Rybicki LA, Adelstein DJ, Murthy SC, DeCamp MM, Goldblum JR. Refining esophageal cancer staging. J Thorac Cardiovasc Surg. 2003;125(5)1103-1113.

27. Veeramachaneni NK, Zoole JB, Decker PA, Putnam JB, Jr., Meyers BF, American College of Surgeons Oncology Group. Lymph node analysis in esophageal resection: American College of Surgeons Oncology Group Z0060 trial. Ann Thorac Surg. 2008;86(2)418-421.

CHAPTER 8

Cyclooxygenase isoenzyme-2 and vascular endothelial growth factor are associated with poor prognosis in esophageal adenocarcinoma

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ABSTRACT

Background

Cyclooxygenase isoenzyme-2 (COX-2) and vascular endothelial growth factor (VEGF) contribute to angiogenesis and are over-expressed in various malignancies. The aim of the study was to evaluate expression, prognostic value and correlation between COX-2 and VEGF expression in esophageal adenocarcinoma (EAC).

Methods

Surgical specimens of 154 patients with EAC were used to construct a tissue micro array (TMA). TMA sections were immunohistochemically stained for COX-2 and VEGF and scored on intensity of staining.

Results

Estimated 5 year cancer specific survival was 37%. High COX-2 and VEGF expression was observed in 39 (26.5%) and in 77 (53.8%) tumors, respectively. Both markers were associated with poor cancer specific survival (p=.022 and p=.004, respectively, log rank). No significant correlation was found between VEGF and COX-2 expression (r=063; p=.455). In multivariate analysis, high COX-2 expression (HR 1.65; 95% CI 1.04-2.61; p=.034) was associated with overall survival. In patients with T3 tumors, COX-2 expression was an independent prognostic factor for cancer specific survival (HR 1.81 95% CI 1.10-2.95; p=.019).

Conclusions

This is the first study to evaluate the prognostic value and correlation of COX-2 and VEGF expression in a large and homogenous population of patients with EAC. No correlation between COX-2 and VEGF expression was found. Both markers were expressed in EAC and were associated with poor prognosis. The findings support the use of COX-2 and VEGF inhibitors in future clinical studies.

INTRODUCTION

Worldwide, esophageal cancer is the 7th leading cause of cancer death.[1] Advancements in diagnostics, surgical techniques and the application of neoadjuvant chemotherapy have improved survival rates. Studies report 5 year survival rates of up to 40% for patients with resectable disease.[2-4] Further therapeutic improvements are warranted and the focus is now turning to targeted therapy. This has led to an increasing interest in the prognostic and therapeutic value of biological tumor markers that are known to play a key role in carcinogenesis and progression. Markers which can be selectively blocked with pharmaceuticals are of particular interest.[5-7] Two of these biomarkers, cyclooxygenase-2 (COX-2) and vascular endothelial growth factor (VEGF) may play a significant role in esophageal adenocarcinoma (EAC). COX-2 is the rate-limiting enzyme involved in the conversion of arachidon acid to eisosanoids such as prostaglandin (PG), prostacyclin and tromboxanes. Its expression is induced by several stimuli including growth factors, inflammation and cytokines. Over-expression of COX-2 is reported in many human malignancies such as colon[8], gastric[9], pancreas[10], lung[11], breast carcinoma[12] and also in esophageal cancer[13,14]. The VEGF gene family belongs to the platelet-derived growth factor (PDGF) gene and encodes for VEGF-A, B, C, D, E and placenta growth factor (PlGF). VEGF-A, referred to as VEGF, is the most potent endothelial cell specific mitogen that induces angiogenesis, which is critical to both tumor growth and systemic spreading of tumor cells.[15] High VEGF expression is reported in several malignancies, including esophageal cancer.[13,16-19] A relationship between COX-2 and VEGF has been described earlier.[20,21] COX-2 generated PGs contributed to angiogenesis through the induction of VEGF. Moreover, it was demonstrated that COX-2 inhibition (e.g. with the use of diclofenac, rofecoxib or celecoxib) resulted in both COX-2 and VEGF protein down-regulation. These findings suggest that COX-2 and VEGF expression are interlinked.[22,23] The role of COX-2 and VEGF and their interaction in EAC is yet unclear. The aim of this study was to evaluate expression and prognostic significance of COX-2 and VEGF in adenocarcinoma of the esophagus. Furthermore, the correlation between COX-2 and VEGF protein expression in EAC was evaluated.

MATERIALS AND METHODS

STUDY POPULATION

All patients who underwent esophagectomy for cancer between August 1988 and November 2009 at the University Medical Center Utrecht were collected in a database. Patients with histologically proven EAC were included. Patients with esophageal squamous cell carcinoma (ESCC), neoadjuvant treatment, pathological T4 disease, distant metastases, tumor positive resection margins (R1) according to the College of American Pathologists

(CAP) criteria[24], and patients who died in-hospital or within 30 days from operation were excluded from the study. All tumor resection specimens were reviewed by an experienced pathologist (FJWtK). Tumors were staged according to the TNM staging system (7th edition).[25] A follow-up study was performed in which patients were followed up until death or up to July 2011. Follow-up data were collected with the use of chart examination, general practitioners archives and the Dutch Cancer Registry. Primary outcome was the percentage of COX-2 and VEGF expression. Secondary outcomes were the correlation between COX-2 and VEGF expression as well as cancer specific survival (CSS; defined as the time between surgery and death due to cancer) and overall survival (OS; defined as the time between surgery and death). The study was performed in accordance with the local ethical guidelines concerning informed consent using patient's material after surgical resection.

TMA CONSTRUCTION

Formalin-fixed, paraffin-embedded tumor blocks were used for construction of a tissue micro array (TMA) as described earlier by Boone *et al.*[13] Briefly, for each tumor specimen, three representative regions were marked on the pathological hematoxylin & eosin (H&E) stained slide. These regions, referred to as tumor cores, were punched out and placed into the TMA paraffin block.

IMMUNOHISTOCHEMISTRY

From the TMA, 4µm sectioned slides were deparaffinized in xylene (15 minutes) and dehydrated in serial ethanol dilutions (15 minutes). Between all steps, tumor slides were rinsed with Tris-HCL buffered saline pH 7.4 (TBS). The endogenous peroxide activity was blocked by hydrogen peroxidase (0.3%) methanol solution for 20 minutes. Antigen retrieval was achieved by boiling the slides for 10 minutes in 0.01 mol/L sodium citrate (pH 6.0). Then TMA slides were incubated during 60 minutes in blocking solution (contents: 0.1 mol/L Tris-HCL pH 7.4; 1 mol/L MgCl2; Tween-20; 10% BSA; Goat serum and H2O) to block unspecific binding sites. Subsequently, TMA slides were incubated with COX-2 monoclonal antibody (Cayman Chemical, Catalog#160112, Clone CX229, dilution 1:100), overnight at 4 °C. The next day slides were rinsed with TBST (Tris-HCL buffered saline pH 7.4 with Tween-20) and post-antibody blocking solution (not diluted) was added for 15 minutes. Subsequently, slides were incubated with polyclonal Goat-anti-Mouse/Rabbit/Rat IgG (not diluted) during 30 minutes. Peroxidase staining was visualized with 3-amino-9-ethylcarbazole (AEC) solution and slides were counter stained with hematoxylin (10 seconds). For VEGF staining, tumor slides were deparaffinized in xylene (15 minutes) and dehydrated in serial ethanol dilutions (15 minutes). Between all steps, slides were rinsed with phosphate buffered saline pH 7.4 (PBS). The endogenous peroxide activity was blocked by hydrogen peroxide (3%) for 15 minutes. Antigen retrieval was achieved by boiling the slides for 20 minutes in sodium citrate (pH 6.0). After a cooling off period (20

minutes), the slides were incubated with polyclonal $VEGF_{165}$ antibody (R&D Systems, Catalog#AF293NA, dilution 1:50) for 60 minutes at room temperature. Subsequently, TMA slides were incubated with biotinylated secondary Rabbit-anti-Goat antibody (1:50) for 30 minutes. Slides were treated with Strep Avidin-Biotin complex for 30 minutes and peroxidase staining was visualized using 3,3'-diaminobenzide for 10 minutes. The sections were counterstained with hematoxylin (10 seconds). For positive controls, colon and stomach carcinoma (known to express high COX-2 protein) were included and a Grawitz tumor as positive control for VEGF staining. Non-cancerous esophageal squamous cell epithelia were used for internal control. Negative controls were achieved by omitting the primary antibody.

IMMUNOHISTOCHEMICAL SCORING

Immunohistochemical scoring was performed by FJWtK. Cores were considered lost if less than 10% of the tissue contained tumor (i.e. sampling error), less than 10% of tissue was present (i.e. absent core) or when 2 out of 3 cores were lost. The scoring of VEGF and COX-2 expression were based on the intensity of staining, which ranged from 1 (no staining), 1 (weak), 2 (moderate) and 3 (strong staining). The lowest observed staining score for COX-2 was 1. At least 1 tumor core had to have a score of 3 to be considered as high COX-2 staining. The highest scoring intensity observed in VEGF stained cores was 2. For VEGF, the median score of the sum of the two highest cores was calculated and used as a cut off value. Tumors which scored <1 were defined as low VEGF expression and tumors which scored ≥ 1 as high VEGF staining.

STATISTICAL ANALYSIS

Association between clinical parameters and COX-2 and VEGF staining were evaluated using cross tabulation (Pearson's Chi-square test). Survival rates were estimated by using the Kaplan Meier function (log rank test) to compare the OS and CSS among patients with high versus low COX-2 and VEGF expression. Correlation between COX-2 and VEGF expression (dichotomous values) was evaluated using the Pearson's correlation. The following parameters were evaluated in univariate analysis: T-stage (T1 or T2 vs. T3), lymph node metastases (no vs. yes), differentiation grade (good and moderate vs. poor), COX-2 (low vs. high), lymph node ratio (≤ 25% vs. >25%), vasoinvasion (no vs. yes), perineural growth (no vs. yes), VEGF (low vs. high), median age (< 64 vs. ≥ 64 years), gender and perinodal extension (no vs. yes). Variables that were significant in univariate analysis were included in multivariate analysis (Cox proportional hazards regression analysis). A *P value* of <.050 was considered statistically significant. All analyses were performed using standard statistical software (SPSS version 15.0; SPSS inc, Chicago, Illinois).

RESULTS

Between 1988 and 2009, 290 patients underwent esophageal resection for EAC at the Department of Surgery of the University Medical Centre Utrecht. Patients with tumor positive resection margins (R1; n= 26), with T4 disease (n=7), patients who were pretreated (n= 49) and patients whose clinical and/or pathological data was incomplete or missing (n= 54) were excluded.

In a total of 154 patients, the female to male ratio was 1:4.5 with a median age of 64.0 years (range, 33.8-81.3). Lymph node metastases were observed in 104 (67.5%) patients of which 62 (59.6%) patients had perinodal extension (i.e. extra capsular location of tumor cells). A lymph node ratio (i.e. the number of positive nodes divided by the total number of resected nodes) of >25%24 was observed in 48 (31.2%) patients.

Follow-up was complete in 144 (93.5%) patients with a median follow-up of 26.4 months (range 2.7 to 260.3). Estimated 5 year CSS was 37% (median CSS 30.4 months). All patient and tumor characteristics are summarized in table 1.

Table 1 Baseline characteristics (n= 154)

	n (%)*
Gender	
Male	126 (81.8)
Female	28 (18.2)
Age (y)	
Median (range)	64.0 (33.8-81.3)
Type of resection	
Transthoracic (open)	24 (15.6)
Transthoracic (scopic)	24 (15.6)
Transhiatal (open)	105 (68.2)
Unknown	1 (0.6)
Tumor stage†	
I AB	25 (16.2)
II AB	24 (15.6)
III AB	69 (44.8)
III C	32 (20.8)
Unknown	4 (2.6)
Tumor extend (T stage)	
T1	17 (11.0)
T2	35 (22.7)
T3	102 (66.2)
LNN involvement	
Yes	104 (67.5)
No	46 (29.9)
Not present‡	4 (2.6)
LNN ratio > 25%	
Yes	48 (31.2)
No	102 (66.2)
Unknown‡	4 (2.6)
Perinodal extension§	
Yes	62 (59.6)
No	42 (40.4)

* Data are n (%), unless noted otherwise. † Tumors were staged according to the TNM classification (anatomical stage groups, 7th edition). ‡ In 4 cases resection specimen did not contain lymph nodes. Median number of resected lymph nodes was 13 (range 0-70). § As a proportion of patients with lymph node involvement (n= 104) . LNN - lymph nodes.

COX-2 EXPRESSION

Tumor cores of 147 (95.5%) patients were assessable for COX-2 scoring. Overall COX-2 staining was seen in all tumor cores (100%). Cytoplasmic COX-2 staining was high in 39 (26.5%) tumors and low in 108 (73.5%) tumors (figure 1A-B). COX-2 staining was positively associated with lymph node metastases (p=.015, table 2). Median CSS was 39.5 months in patients with low COX-2 expression (95% CI 20.00-58.93) versus 21.0

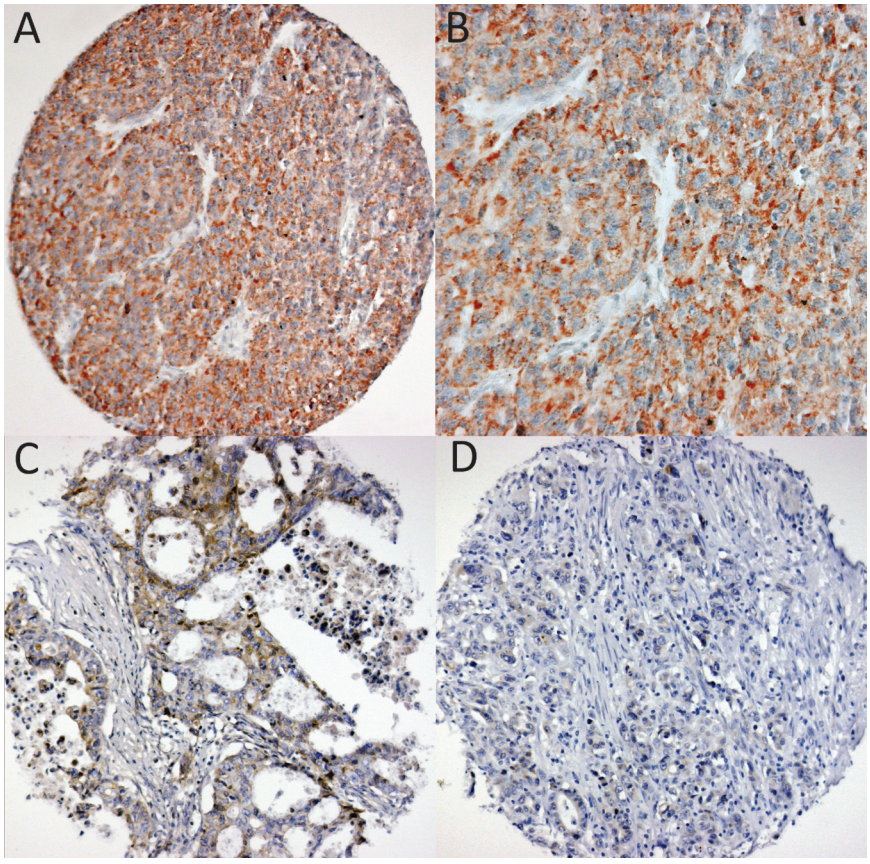

Figure 1 Representative examples of COX-2 and VEGF staining
Legend: A: Tumor core showing strong (3+) cytoplasmic and granular COX-2 staining. Stromal tumor cells did not or only slightly stain positive for COX-2. Original magnification (200 x). B: Magnification (400 x) of tumor core shown in A. C: Tumor core showing strong cytoplasmic VEGF staining (2+). Original magnification (200 x). D: Tumor cells showing weak cytoplasmic VEGF staining (1+). Original magnification (200 x).

months (95% CI 17.96-24.00) in patients with high COX-2 expression (p=.022; log rank, figure 2). High COX-2 expression was significantly and inversely associated with CSS (HR 1.70; 95% CI 1.07-2.69; p=.023, table 4). COX-2 expression was not associated with recurrence of disease (p=.173, table 2)

VEGF EXPRESSION

VEGF evaluation was assessable in tumor cores of 143 (92.9%) patients. Overall staining was seen in 90 (62.9%) tumors. High cytoplasmic expression was seen in 77 (53.8%) and low staining in 66 (46.2%) of patients (figure 1C-D). VEGF expression positively correlated with tumor stage (p=.000), presence of lymph node metastases (p=.032) and a lymph node ratio of >25% (p=.001, table 3). In patients with lymph node metastases,

Table 2 Correlation of COX-2 expression and clinical and pathological parameters

		COX-2 expression		
	Total (n)	**Low (%)**	**High (%)**	***P value†***
Total*	147	73.5	26.5	-
Gender				
Male	121	76.0	24.0	*0.129*
Female	26	61.5	38.5	
Median age				
< 64 years	72	75.0	25.0	*0.680*
≥ 64 years	75	72.0	28.0	
PATHOLOGICAL DATA				
T-stage				
T1 or T2	46	82.6	17.4	*0.090*
T3	101	69.3	30.7	
Tumor differentiation				
G1 or G2	55	72.7	27.3	*1.000*
G3	88	72.7	27.3	
Vaso invasion				
Yes	69	75.4	24.6	*0.625*
No	78	71.8	28.2	
Perineural growth				
Yes	43	65.1	34.9	*0.149*
No	103	76.7	23.3	
LNN metastases				
Yes	102	68.6	31.4	***0.015***
No	42	88.1	11.9	
LNN ratio				
≤ 25%	96	78.1	21.9	*0.138*
> 25%	48	66.7	33.3	
FOLLOW-UP DATA				
Recurrence and /or metastases				
yes	88	69.3	30.7	*0.173*
no	50	80.0	20.0	
Localregional recurrence				
yes ‡	31	74.2	25.8	*0.541*
no	50	80.0	20.0	
Distant metastases				
yes ‡	75	68.0	32.0	*0.140*
no	50	80.0	20.0	

* Tumor cores from 147 patients were assessable for COX-2 scoring (147 of 154: 95.5%). † Pearson Chi-square test. ‡ A proportion of patients (n= 18) experienced both locoregional and distant metastases. LNN - lymph nodes.

VEGF was significantly associated with perinodal nodal extension (p=.024, data not shown). Patients with high VEGF expression more frequently developed recurrence of disease (p=.004, table 3). Patients with low VEGF expression had a median CSS of 59.5 months (95% CI 0.00-153.03) whereas patients with high VEGF expression had a median CSS of 21.6 months (95% CI 16.18-27.02; p=.004; log rank, figure 3). High VEGF expression was significantly associated with poor CSS (HR 1.90; 95% CI 1.22-2.96; p=.005, table 4).

CORRELATION BETWEEN COX-2 AND VEGF

Tumor cores of 143 (92.9%) patients were assessable for combined evaluation of VEGF and COX-2 staining. There was no significant correlation between VEGF and COX-2 expression (r=.063; p=.455).

MULTIVARIATE ANALYSIS

In multivariate analysis, high COX-2 expression was not associated with poor CSS (HR 1.55; 95% CI 0.95-2.53; p=.081, table 4) For OS, high COX-2 expression was an independent prognostic factor (HR 1.65; 95% CI 1.04-2.61; p=.034, table 5). VEGF expression was not independently associated with CSS (HR 0.88; 95% CI 0.54-1.42; p=.591, table 4) nor with OS (HR 1.05; 95% CI 0.67-1.64; p=.848, table 5).

In the subset of patients with advanced disease (i.e. T3 tumors), COX-2 was an independent prognostic marker for CSS (HR 1.81; 95% CI 1.10-2.95; p=.019, table 6) as well as for OS (HR 1.90; 95% CI 1.18-3.05; p=.008, data not shown).

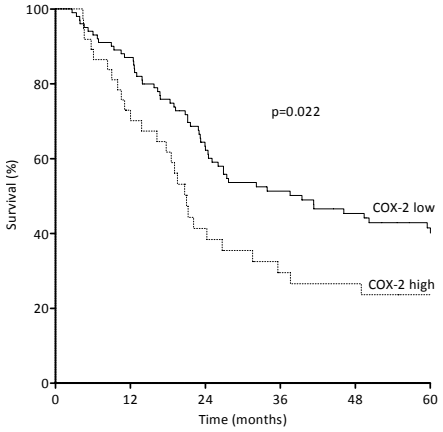

Figure 2 Cancer specific survival according to high and low COX-2 staining
Legend: Log rank test was used to compare differences between survival curves.

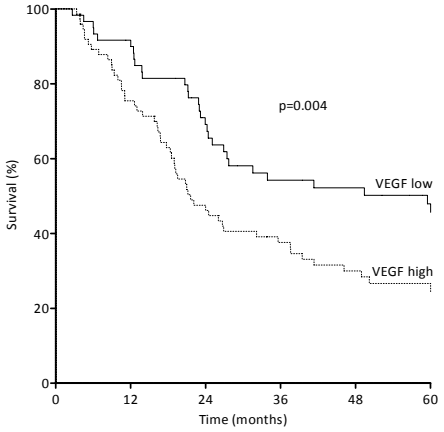

Figure 3 Cancer specific survival according to high and low VEGF staining
Legend: Log rank test was used to compare differences between survival curves.

Table 3 Correlation of VEGF expression and clinical and pathological parameters

		VEGF expression		
	Total (n)	**Low (%)**	**High (%)**	***P value†***
Total*	143	46.2	53.8	-
Gender				
Male	117	44.4	55.6	0.384
Female	26	53.8	46.2	
Median age				
< 64 years	71	49.3	50.7	0.454
≥ 64 years	72	43.1	56.9	
PATHOLOGICAL DATA				
T-stage				
T1 or T2	44	72.7	27.3	***0.000***
T3	99	34.3	65.7	
Tumor differentiation				
G1 or G2	52	53.8	46.2	0.090
G3	87	39.1	60.9	
Vaso invasion				
Yes	68	38.2	61.8	0.071
No	75	53.3	46.7	
Perineural growth				
Yes	41	43.9	56.1	0.695
No	101	47.5	52.5	
LNN metastases				
Yes	100	40.0	60.0	***0.032***
No	40	60.0	40.0	
LNN ratio				
≤ 25%	93	55.9	44.1	***0.001***
> 25%	47	25.5	74.5	
FOLLOW-UP DATA				
Recurrence and /or metastases				
Yes	87	35.6	64.4	***0.004***
No	47	61.7	38.3	
Localregional recurrence				
Yes ‡	30	26.7	73.3	***0.003***
No	47	61.7	38.3	
Distant metastases				
Yes ‡	75	37.3	62.7	***0.009***
No	47	61.7	38.3	

* Tumor cores from 143 patients were assessable for VEGF scoring (143 of 154: 92.9%). † Pearson Chi-square test. ‡ A proportion of patients (n= 18) experienced both locoregional and distant metastases. LNN - lymph nodes.

Table 4 Univariate and multivariate analysis of associations between histopathologic parameters and cancer specific survival (CSS)

	Median CSS in months		Unadjusted HR (95% CI)	*P value*	Adjusted HR (95% CI)*	*P value*
	Negative	Positive	(Univariate)		(Multivariate)	
Perinodal extension ‡	26.9	20.7	1.864 (1.160-2.995)	***0.010***		
Gender (male/ female)	32.2	26.7	1.050 (0.592-1.860)	*0.868*		
Median age (<64/ ≥64 Y)	24.5	37.6	1.001 (0.657-1.525)	*0.998*		
T stage (T1,T2/ T3)	†	22.9	10.282 (4.441-23.807)	***0.000***	3.833 (1.404-10.468)	***0.009***
Positive lymph nodes	†	23.2	6.595 (3.166-13.740)	***0.000***	2.338 (0.979-5.587)	0.056
G grade (G1,G2/ G3) §	127.9	21.2	2.952 (1.809-4.817)	***0.000***	2.103 (1.214-3.644)	***0.008***
LNN ratio (≤ 0.25/ > 25)	64.3	18.5	3.832 (2.194-5.213)	***0.000***	1.595 (0.943-2.698)	0.082
COX-2 (low/ high)	39.4	21.0	1.698 (1.074-2.685)	***0.023***	1.546 (0.947-2.525)	0.081
Vasoinvasion	†	21.6	3.283 (2.107-5.116)	***0.000***	1.538 (0.901-2.623)	0.114
Perineural growth	49.4	21.2	2.532 (1.633-3.927)	***0.000***	1.142 (0.705-1.851)	0.589
VEGF (low/ high)	59.5	21.6	1.897 (1.216-2.959)	***0.005***	0.877 (0.544-1.415)	0.591

* Multivariate analysis was carried out with variables proven significant in univariate analysis. † Median survival was not determined, since expected cumulative survival within the study period did not reach 50%. ‡ As a proportion of patients with positive lymph nodes (n= 100). Perinodal extension was not included in multivariate analysis. § G grade indicates tumor differentiation. LNN - lymph nodes; CI - confidence interval.

Table 5 Univariate and multivariate analysis of associations between histopathologic parameters and overall survival (OS)

	Median CSS in months		Unadjusted HR (95% CI)	*P value*	Adjusted HR (95% CI)*	*P value*
	Negative	Positive	(Univariate)		(Multivariate)	
Perinodal extension †	26.7	19.2	1.736 (1.108-2.722)	***0.016***		
Gender (male/ female)	24.0	29.0	1.120 (0.674-1.860)	*0.661*		
Median age (<64/ ≥64 Y)	24.4	27.4	1.319 (0.899-1.934)	*0.157*		
T stage (T1,T2/ T3)	196.1	21.2	5.537 (3.148-9.736)	***0.000***	2.974 (1.390-6.364)	***0.005***
G grade (G1,G2/ G3) ‡	64.3	19.5	2.864 (1.846-4.443)	***0.000***	2.218 (1.342-3.665)	***0.002***
COX-2 (low/ high)	29.1	19.5	1.732 (1.138-2.636)	***0.010***	1.645 (1.037-2.607)	***0.034***
Vasoinvasion	61.6	21.2	2.635 (1.771-3.922)	***0.000***	1.458 (0.897-2.367)	0.128
Positive lymph nodes	193.1	22.9	3.585 (2.130-6.034)	***0.000***	1.393 (0.730-2.657)	0.314
LNN ratio (≤ 0.25/ > 25)	48.9	18.3	2.592 (1.735-3.873)	***0.000***	1.355 (0.827-2.221)	0.228
Perineural growth	39.4	21.0	2.333 (1.536-3.543)	***0.000***	1.139 (0.714-1.815)	0.585
VEGF (low/ high)	41.3	21.0	1.945 (1.295-2.920)	***0.001***	1.045 (0.667-1.636)	0.848

* Multivariate analysis was carried out with variables proven significant in univariate analysis. †As a proportion of patients with positive lymph nodes (n= 100). Perinodal extension was not included in multivariate analysis. ‡ G grade indicates tumor differentiation. LNN - lymph nodes; CI - confidence interval.

Table 6 Univariate and Multivariate analysis of associations between histopathologic parameters and cancer specific survival (CSS) in T3 tumors (n=100)

	Median CSS in months		Unadjusted HR (95% CI)	*P value*	Adjusted HR (95% CI)*	*P value*
	Negative	Positive	(Univariate)		(Multivariate)	
Gender (male/ female)	23.2	16.4	1.461 (0.818-2.608)	0.200		
Perineural growth	23.2	21.2	1.387 (0.887-2.169)	0.151		
Perinodal extension †	24.0	20.7	1.261 (0.775-2.052)	0.350		
VEGF (low/ high)	23.9	21.0	1.056 (0.665-1.677)	0.819		
Median age (<64/ ≥64 Y)	22.1	24.0	1.012 (0.653-1.567)	0.959		
G grade (G1,G2/ G3) ‡	37.6	19.1	2.335 (1.373-3.971)	***0.002***	2.017 (1.159-3.511)	***0.013***
COX-2 (low/ high)	24.4	19.1	1.769 (1.099-2.847)	***0.019***	1.805 (1.103-2.954)	***0.019***
Positive lymph nodes	41.3	22.9	2.200 (1.005-4.816)	***0.048***	1.822 (0.789-4.209)	0.160
Vasoinvasion	26.9	20.7	2.020 (1.278-3.195)	***0.003***	1.640 (0.963-2.791)	0.068
LNN ratio (≤ 0.25/ > 25)	26.7	18.5	2.226 (1.420-3.488)	***0.000***	1.453 (0.859-2.458)	0.163

* Multivariate analysis was carried out with variables proven significant in univariate analysis. † As a proportion of patients with positive lymph nodes (n= 88). ‡ G grade indicates tumor differentiation. LNN - lymph nodes; CI - confidence interval.

DISCUSSION

This is the first study that evaluated the prognostic value and interaction of COX-2 and VEGF expression in a large and homogenous population of patients with EAC. Patients who received neo-adjuvant treatment or patients with T4 disease, distant metastases at time of operation and tumor positive resection margins were excluded.

THE PROGNOSTIC VALUE OF COX-2 AND VEGF OVER-EXPRESSION

The results showed high COX-2 and VEGF expression in 39 (26.5%) and in 77 (53.8%) of patients. Both were significantly associated with poor cancer specific survival (CSS) (p=.022 and p=.004, respectively, log rank). In addition, COX-2 was independently associated with poor overall survival (OS). In patients with advanced disease (T3), COX-2 was also an independent prognostic marker for CSS. This suggests that COX-2 is particularly of prognostic significance for patients with advanced disease.

Other studies have also identified COX-2 expression as an independent prognostic factor in EAC[14,26,27] and in ESCC[28]. Some authors have reported an independent prognostic significance of VEGF over-expression in ESCC[29-31], whereas other studies could not find any association between VEGF expression and survival in EAC[32] and ESCC[33-35]. In EAC, only one study has showed the independent prognostic value of VEGF over-expression.[18] Some important limitations of these studies must be mentioned. Buskens *et al.*, Takatori *et al.*, Saad *et al.* and Ogata *et al.* included patients who had

distant metastases at time of resection, which was seen in 19% (28/145), 33% (75/228), 40% (30/75) and in 24% (22/92) of cases, respectively.[14,18,28,31] In addition, the series of Buskens *et al*. included 33 of 145 (23%) patients with a positive resection margin (R1).[14] Patients with distant metastases and R1 margins have poor prognosis irrespective of COX-2 expression. The inclusion of such cases greatly affects the survival rate of the studied population, making it difficult to interpret the correlation between protein expression and prognosis. Furthermore, many of these studies analyzed OS instead of CSS.[14,26,27]

CORRELATION BETWEEN COX-2 AND VEGF EXPRESSION

COX-2 and VEGF both play an important role in carcinogenesis, tumor progression and angiogenesis. In the studied cohort, we did not observe a correlation between VEGF and COX-2 expression (r=.063; p=.455). Other studies with varying population size, patient selection and methodology reported otherwise and suggest that the two markers are interlinked.[23,36,37] Vallböhmer *et al*. (n=75) and von Rahden *et al*. (n=123) evaluated the quantity of VEGF and COX-2 messenger RNA in EAC using quantitative reverse transcription polymerase chain reaction (qRT PCR). Both authors reported a significant correlation between COX-2 and VEGF (r=.460; p< .001 and r=.764; p< .001, respectively).[23,36] However, Vallböhmer *et al*. investigated a mixed population (16 patients with ESCC, 15 with Barrett's esophagus and 44 with EAC) and found that COX-2 and VEGF expressions were only correlated when the overall study population was analyzed.[36] In another study of 40 patients with ESCC, a significant correlation between high COX-2 (65.4%) and VEGF (50%) expression (p<.005) was reported using immunohistochemistry.[37] The study included patients who received neoadjuvant therapy before surgery. Pathological complete response (i.e. the absence of visible tumor cells) was seen in 35% of patients. The evaluation of tumor specific protein expression in specimens without visible tumor cells is questionable. However, these specimens were considered as negative staining for both COX-2 and VEGF. This increased the proportion of VEGF and COX-2 co-expression (i.e. negative staining).

RELATIONSHIP OF COX-2 AND VEGF EXPRESSION AND CLINICAL AND PATHOLOGICAL PARAMETERS

Analysis of (linear) relationships between COX-2 expression and prognostic parameters showed a significant correlation between high COX-2 expression and the presence of lymph node metastases. This is consistent with previous studies.[27,37,38] High VEGF expression was significantly correlated with depth of tumor invasion (T-stage), presence of lymph node metastases, lymph node ratio, and perinodal growth. In addition, expression was associated with recurrence of disease. This is also consistent with previous studies which describe a correlation between VEGF and T-stage[18,29,35,39,40], lymph node metastases[18,39-43], and recurrence of disease[18,29].

COX-2 and VEGF expression were both associated with lymphatic tumor metastases. A possible explanation might be found in the involvement of HIF-1α, which is an important transcription factor regulating transcription of genes that are involved in metastatic spreading of disease. COX-2 and VEGF are target genes of HIF-1α and are both responsible for vasodilation, increased vessel permeability and tumor cell invasion. We hypothesize that COX-2 and VEGF both lead to vasodilatation of lymphatic vessels leading to increased microvascular permeability. The leaky state of microvessels causes extravasation of tissue metalloproteinase, which is also induced by COX-2 and VEGF.[44,45] This causes degradation of the extracellular matrix, further facilitating invasion of cancer cells into the lymphatic vessels. Furthermore, VEGF induced metalloproteinases may lead to degradation of basement membrane and therefore to higher T-stage (i.e. depth of tumor invasion). Again, this is hypothetical and requires investigation in future studying.

EVALUATION OF COX-2 AND VEGF EXPRESSION USING IMMUNOHISTOCHEMISTRY (IHC)

Reported rates of high VEGF and COX-2 protein expression in esophageal cancer vary widely from 25%-91% for COX-2[13,14,46] and 24%-80% for VEGF expression[13,18,30,35,40,42]. This variation is mostly due to differences in patient selection (e.g. inclusion of patients with both ESCC and EAC, with distant metastases or R1 resection margins) and methodology (e.g. the use of different cutoff values, reagents or scoring methods).

It might be argued that evaluation of messenger RNA expression with qRT-PCR and Northern blot techniques provides a more objective analysis of COX-2 and VEGF expression. However, immunhositochemistry (IHC) is the most frequently used and most reliable method today for analyzing protein expression from formalin-fixed and paraffin embedded material. IHC is still the gold standard to evaluate tumor marker expression for diagnostic purposes. Moreover, a major advantage of IHC above other techniques is that it enables precise location of the (increased) signal within the tumor cell. With techniques that evaluate messenger RNA expression the histology of the tumor is no longer available. These techniques may overestimate COX-2 and VEGF expression, because non-cancerous tissues such as inflammatory cells and vascular endothelial cells (that also express high levels COX-2 and VEGF) are included in the sample.

TARGETED THERAPIES WITH COX-2 AND VEGF INHIBITORS

The results showed that COX-2 and VEGF expression were both associated with poor survival. Therefore, specific inhibition of these pathways may affect prognosis. To date, several trials have been conducted with VEGF inhibitors such as monoclonal antibody bevacizumab or tyrosine kinase inhibitors (TKI's) like sorafenib.[47-49] Other trials included COX-2 specific inhibitors such as celecoxib.[22,50,51] Two phase I-II trials, applying chemoradiotherapy (cisplatin, fluorouracil) added with celecoxib, report clinical complete response 7 of 13 patients (54%) and pathological complete response in 5 of 22 patients

(22%), respectively.[50,51] Furthermore, Tuynman *et al.* reported significant COX-2 down-regulation in patients who underwent neoadjuvant celecoxib treatment (p< .010).[22] Although concerns exist about long-term usage of COX-2 inhibitors with regard to its potential cardiovascular risk[52], in all studies celecoxib was accompanied with acceptable toxicities without cardiovascular events.

A phase II study combining chemotherapy (irinotecan and cisplatin) with bevacizumab treatment in 34 patients resulted in an overall response rate (i.e. complete and partial response) of 65% (complete pathologic response in 6%).[47] Another phase II study, which combined doxetaxel and cisplatin with sorafenib, yielded 41% partial response (complete response in 0%).[48] A phase II study with doxetaxel, cisplatin and fluorouracil added with bevacizumab (n=39) showed a response rate of 67% (complete response in 5%).[49] These results support the use of VEGF and COX-2 inhibitors in future studies. Combining the inhibitors could possibly show synergistic effects, ultimately benefitting patients' prognosis.

FUTURE PERSPECTIVES

At present, all patients receive neoadjuvant treatment. Therefore, it is not possible to validate the prognostic value of COX-2 and VEGF expression in a prospective cohort. However, it would be of great value to prospectively evaluate COX-2 and VEGF expression in pre-treatment biopsies. Using this method, correlation between COX-2 and VEGF expression and response to neoadjuvant treatment can be evaluated. When patients are not expected to benefit from adjuvant therapy, surgery could be rescheduled accordingly to prevent further progression of disease.

In conclusion, the results show high COX-2 in a quarter of EAC patients and high VEGF expression in over half of EAC patients. Both markers were associated with poor prognosis. Moreover, COX-2 was an independent prognostic marker for survival. Although a relationship between COX-2 and VEGF expression could not be identified, the findings support the use of COX-2 and VEGF inhibitors in future clinical studies.

REFERENCES

1. Jemal A, Siegel R, Xu J, Ward E. Cancer statistics. 2010. CA Cancer J.Clin. 2010;60:277-300.
2. Cunningham D, Allum WH, Stenning SP, Thompson JN, van de Velde CJ, Nicolson M, Scarffe JH, Lofts FJ, Falk SJ, Iveson TJ, Smith DB, Langley RE, Verma M, Weeden S, Chua YJ, MAGIC Trial Participants. Perioperative chemotherapy versus surgery alone for resectable gastroesophageal cancer. N Engl J Med. 2006;355:11-20.
3. Omloo JM, Lagarde SM, Hulscher JB, Reitsma JB, Fockens P, van Dekken H, ten Kate FJW, Obertop H, Tilanus HW, Van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the mid/distal esophagus: five-year survival of a randomized clinical trial. Ann Surg. 2007;246:992-1000.
4. van Meerten E, Muller K, Tilanus HW, Siersema PD, Eijkenboom WM, van Dekken H, Tran TC, van der Gaast A. Neoadjuvant concurrent chemoradiation with weekly paclitaxel and carboplatin for patients with oesophageal cancer: a phase II study. Br J Cancer. 2006;94:1389-1394.
5. Aklilu M, Ilson DH. Targeted agents and esophageal cancer--the next step? Semin Radiat Oncol. 2007;17:62-69.
6. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell. 2011;144:646-674.
7. Tew WP, Kelsen DP, Ilson DH. Targeted therapies for esophageal cancer. Oncologist. 2005;10:590-601.
8. Ogino S, Kirkner GJ, Nosho K, Irahara N, Kure S, Shima K, Hazra A, Chan AT, Dehari R, Giovannucci EL, Fuchs CS. Cyclooxygenase-2 expression is an independent predictor of poor prognosis in colon cancer. Clin Cancer Res. 2008;14:8221-8227.
9. Lim HY, Joo HJ, Choi JH, Yi JW, Yang MS, Cho DY, Kim HS, Nam DK, Lee KB, Kim HC. Increased expression of cyclooxygenase-2 protein in human gastric carcinoma. Clin Cancer Res. 2000;6:519-525.
10. Matsubayashi H, Infante JR, Winter J, Klein AP, Schulick R, Hruban R, Visvanathan K, Goggins M. Tumor COX-2 expression and prognosis of patients with resectable pancreatic cancer. Cancer Biol Ther. 2007;6:1569-1575.
11. Wu YC, Su LJ, Wang HW, Jeff Lin CF, Hsu WH, Chou TY, Huang CY, Lu CL, Hsueh CT. Co-overexpression of cyclooxygenase-2 and microsomal prostaglandin E synthase-1 adversely affects the postoperative survival in non-small cell lung cancer. J Thorac Oncol. 2010;5:1167-1174.
12. Barisik NO, Keser SH, Gul AE, Sensu S, Kandemir NO, Kucuk HF, Gumus M, Karadayi N. The value of COX-2 expression in the prognostic parameters of invasive ductal carcinoma of the breast. Med Oncol. 2010;28:703-708.
13. Boone J, van Hillegersberg R, Offerhaus GJ, van Diest PJ, Borel Rinkes IHM, ten Kate FJW. Targets for molecular therapy in esophageal squamous cell carcinoma: an immunohistochemical analysis. Dis Esophagus. 2009;22:496-504.
14. Buskens CJ, Van Rees BP, Sivula A, Reitsma JB, Haglund C, Bosma PJ, Offerhaus GJ, Van Lanschot JJ, Ristimaki A. Prognostic significance of elevated cyclooxygenase 2 expression in patients with adenocarcinoma of the esophagus. Gastroenterology. 2002;122:1800-1807.
15. Kleespies A, Bruns CJ, Jauch KW. Clinical significance of VEGF-A, -C and -D expression in esophageal malignancies. Onkologie. 2005;28:281-288.
16. Donnem T, Al-Shibli K, Andersen S, Al-Saad S, Busund LT, Bremnes RM. Combination of low vascular endothelial growth factor A (VEGF-A)/VEGF receptor 2 expression and high lymphocyte infiltration is a strong and independent favorable prognostic factor in patients with nonsmall cell lung cancer. Cancer. 2010;116:4318-4325.
17. Patard JJ, Rioux-Leclercq N, Masson D, Zerrouki S, Jouan F, Collet N, Dubourg C, Lobel B, Denis M, Fergelot P. Absence of VHL gene alteration and high VEGF expression are associated with tumour aggressiveness and poor survival of renal-cell carcinoma. Br J Cancer. 2009;101:1417-1424.
18. Saad RS, El-Gohary Y, Memari E, Liu YL, Silverman JF. Endoglin (CD105) and vascular endothelial growth factor as prognostic markers in esophageal adenocarcinoma. Hum Pathol. 2005;36:955-961.
19. Wu Y, Jin M, Xu H, Shimin Z, He S, Wang L, Zhang Y. Clinicopathologic significance of HIF-1alpha, CXCR4, and VEGF expression in colon cancer. Clin Dev Immunol. 2010;2010:

20. Masferrer JL, Leahy KM, Koki AT, Zweifel BS, Settle SL, Woerner BM, Edwards DA, Flickinger AG, Moore RJ, Seibert K. Antiangiogenic and antitumor activities of cyclooxygenase-2 inhibitors. Cancer Res. 2000;60:1306-1311.

21. Gately S. The contributions of cyclooxygenase-2 to tumor angiogenesis. Cancer Metastasis Rev. 2000;19:19-27.

22. Tuynman JB, Buskens CJ, Kemper K, ten Kate FJW, Offerhaus GJ, Richel DJ, Van Lanschot JJ. Neoadjuvant selective COX-2 inhibition down-regulates important oncogenic pathways in patients with esophageal adenocarcinoma. Ann Surg. 2005;242:840-9.

23. von Rahden BH, Stein HJ, Puhringer F, Koch I, Langer R, Piontek G, Siewert JR, Hofler H, Sarbia M. Coexpression of cyclooxygenases (COX-1, COX-2) and vascular endothelial growth factors (VEGF-A, VEGF-C) in esophageal adenocarcinoma. Cancer Res. 2005;65:5038-5044.

24. Verhage RJ, Zandvoort HJ, ten Kate FJ, van Hillegersberg R. How to define a positive circumferential resection margin in T3 adenocarcinoma of the esophagus. Am J Surg Pathol. 2011;35:919-926.

25. Edge SB, Byrd DR, Compton CC, Fritz AG, Greene FL, Trotti A. AJCC cancer staging manual (7th edition). 2010.

26. Heeren P, Plukker J, van Dullemen H, Nap R, Hollema H. Prognostic role of cyclooxygenase-2 expression in esophageal carcinoma. Cancer Lett. 2005;225:283-289.

27. Bhandari P, Bateman AC, Mehta RL, Stacey BS, Johnson P, Cree IA, Di NF, Patel P. Prognostic significance of cyclooxygenase-2 (COX-2) expression in patients with surgically resectable adenocarcinoma of the oesophagus. BMC Cancer. 2006;6:134.

28. Takatori H, Natsugoe S, Okumura H, Matsumoto M, Uchikado Y, Setoyama T, Sasaki K, Tamotsu K, Owaki T, Ishigami S, Aikou T. Cyclooxygenase-2 expression is related to prognosis in patients with esophageal squamous cell carcinoma. Eur J Surg Oncol. 2008;34:397-402.

29. Inoue K, Ozeki Y, Suganuma T, Sugiura Y, Tanaka S. Vascular endothelial growth factor expression in primary esophageal squamous cell carcinoma. Association with angiogenesis and tumor progression. Cancer. 1997;79:206-213.

30. Shih CH, Ozawa S, Ando N, Ueda M, Kitajima M. Vascular endothelial growth factor expression predicts outcome and lymph node metastasis in squamous cell carcinoma of the esophagus. Clin Cancer Res. 2000;6:1161-1168.

31. Ogata Y, Fujita H, Yamana H, Sueyoshi S, Shirouzu K. Expression of vascular endothelial growth factor as a prognostic factor in node-positive squamous cell carcinoma in the thoracic esophagus: long-term follow-up study. World J Surg. 2003;27:584-589.

32. Couvelard A, Paraf F, Gratio V, Scoazec JY, Henin D, Degott C, Flejou JF. Angiogenesis in the neoplastic sequence of Barrett's oesophagus. Correlation with VEGF expression. J Pathol. 2000;192:14-18.

33. Ahn MJ, Jang SJ, Park YW, Choi JH, Oh HS, Lee CB, Paik HK, Park CK. Clinical prognostic values of vascular endothelial growth factor, microvessel density,and p53 expression in esophageal carcinomas. J Korean Med Sci. 2002;17:201-207.

34. Rosa AR, Schirmer CC, Gurski RR, Meurer L, Edelweiss MI, Kruel CD. Prognostic value of p53 protein expression and vascular endothelial growth factor expression in resected squamous cell carcinoma of the esophagus. Dis Esophagus. 2003;16:112-118.

35. Lu CL, Ji Y, Ge D, Guo J, Ding JY. The expression of CXCR4 and its relationship with matrix metalloproteinase-9/ vascular endothelial growth factor in esophageal squamous cell cancer. Dis Esophagus. 2011;24:283-290.

36. Vallbohmer D, Peters JH, Kuramochi H, Oh D, Yang D, Shimizu D, DeMeester SR, Hagen JA, Chandrasoma PT, Danenberg KD, Danenberg PV, DeMeester TR. Molecular determinants in targeted therapy for esophageal adenocarcinoma. Arch Surg. 2006;141:476-481.

37. Yoshikawa R, Fujiwara Y, Koishi K, Kojima S, Matsumoto T, Yanagi H, Yamamura T, Hashimoto-Tamaoki T, Nishigami T, Tsujimura T. Cyclooxygenase-2 expression after preoperative chemoradiotherapy correlates with more frequent esophageal cancer recurrence. World J Gastroenterol. 2007;13:2283-2288.

38. Mobius C, Stein HJ, Spiess C, Becker I, Feith M, Theisen J, Gais P, Jutting U, Siewert JR. COX2 expression, angiogenesis, proliferation and survival in Barrett's cancer. Eur J Surg Oncol. 2005;31:755-759.

39. Kitadai Y, Haruma K, Tokutomi T, Tanaka S, Sumii K, Carvalho M, Kuwabara M, Yoshida K, Hirai T, Kajiyama G, Tahara E. Significance of vessel count and vascular endothelial growth factor in human esophageal carcinomas. Clin Cancer Res. 1998;4:2195-2200.

40. Uchida S, Shimada Y, Watanabe G, Tanaka H, Shibagaki I, Miyahara T, Ishigami S, Imamura M. In oesophageal squamous cell carcinoma vascular endothelial growth factor is associated with p53 mutation, advanced stage and poor prognosis. Br J Cancer. 1998;77:1704-1709.

41. Sato F, Shimada Y, Watanabe G, Uchida S, Makino T, Imamura M. Expression of vascular endothelial growth factor, matrix metalloproteinase-9 and E-cadherin in the process of lymph node metastasis in oesophageal cancer. Br J Cancer. 1999;80:1366-1372.

42. Mukherjee T, Kumar A, Mathur M, Chattopadhyay TK, Ralhan R. Ets-1 and VEGF expression correlates with tumor angiogenesis, lymph node metastasis, and patient survival in esophageal squamous cell carcinoma. J Cancer Res Clin Oncol. 2003;129:430-436.

43. Millikan KW, Mall JW, Myers JA, Hollinger EF, Doolas A, Saclarides TJ. Do angiogenesis and growth factor expression predict prognosis of esophageal cancer? Am Surg. 2000;66:401-405.

44. Pufe T, Harde V, Petersen W, Goldring MB, Tillmann B, Mentlein R. Vascular endothelial growth factor (VEGF) induces matrix metalloproteinase expression in immortalized chondrocytes. J Pathol. 2004;202:367-374.

45. Sivula A, Talvensaari-Mattila A, Lundin J, Joensuu H, Haglund C, Ristimaki A, Turpeenniemi-Hujanen T. Association of cyclooxygenase-2 and matrix metalloproteinase-2 expression in human breast cancer. Breast Cancer Res Treat. 2005;89:215-220.

46. Sivula A, Buskens CJ, Van Rees BP, Haglund C, Offerhaus GJ, Van Lanschot JJ, Ristimaki A. Prognostic role of cyclooxygenase-2 in neoadjuvant-treated patients with squamous cell carcinoma of the esophagus. Int J Cancer. 2005;116:903-908.

47. Shah MA, Ramanathan RK, Ilson DH, Levnor A, D'Adamo D, O'Reilly E, Tse A, Trocola R, Schwartz L, Capanu M, Schwartz GK, Kelsen DP. Multicenter phase II study of irinotecan, cisplatin, and bevacizumab in patients with metastatic gastric or gastroesophageal junction adenocarcinoma. J Clin Oncol. 2006;24:5201-5206.

48. Sun W, Powell M, O'Dwyer PJ, Catalano P, Ansari RH, Benson AB3. Phase II study of sorafenib in combination with docetaxel and cisplatin in the treatment of metastatic or advanced gastric and gastroesophageal junction adenocarcinoma: ECOG 5203. J Clin Oncol. 2010;28:2947-2951.

49. Shah MA, Jhawer M, Ilson DH, Lefkowitz RA, Robinson E, Capanu M, Kelsen DP. Phase II study of modified docetaxel, cisplatin, and fluorouracil with bevacizumab in patients with metastatic gastroesophageal adenocarcinoma. J Clin Oncol. 2011;29:868-874.

50. Dawson SJ, Michael M, Biagi J, Foo KF, Jefford M, Ngan SY, Leong T, Hui A, Milner AD, Thomas RJ, Zalcberg JR. A phase I/II trial of celecoxib with chemotherapy and radiotherapy in the treatment of patients with locally advanced oesophageal cancer. Invest New Drugs. 2007;25:123-129.

51. Govindan R, McLeod H, Mantravadi P, Fineberg N, Helft P, Kesler K, Hanna N, Stoner C, Ansari R, Fox E. Cisplatin, fluorouracil, celecoxib, and RT in resectable esophageal cancer: preliminary results. Oncology. 2004;18:18-21.

52. Solomon SD, McMurray JJ, Pfeffer MA, Wittes J, Fowler R, Finn P, Anderson WF, Zauber A, Hawk E, Bertagnolli M. Cardiovascular risk associated with celecoxib in a clinical trial for colorectal adenoma prevention. N Engl J Med. 2005;352:1071-1080.

PART THREE

SUMMARY

CHAPTER 9

Summary

The incidence of esophageal cancer is still rising steadily. In the Western World, this is evidently due to the increasing incidence of esophageal adenocarcinoma. Surgery, nowadays often supplemented with neoadjuvant treatment strategies such as chemo- and/or radiotherapy, constitutes the mainstay of esophageal cancer therapy. Despite advances in perioperative care, the development of less invasive surgical techniques and the improvement of chemo(radio)therapeutic treatment regimens, the 5-year survival rates after potentially curable esophageal cancer rarely exceeds 40%. Regardless of surgical approach or the application of other treatment modalities, treatment of esophageal cancer is associated with high morbidity and mortality rates. It is essential to further improve medical and surgical therapy strategies. This will not only benefit short term peri-operative outcomes, but also affect long term survival.
The studies presented in this thesis focus on various features of esophageal cancer surgery. These include perioperative morbidity (short term outcomes) and factors that influence prognostication of survival (long term outcomes).

PART ONE | PERIOPERATIVE MORBIDITY

A systematic review of studies comparing open esophagectomy with minimally invasive esophagectomy was performed in **Chapter 2**. A literature search yielded 10 eligible papers of comparative studies. The studies were subjected to a critical appraisal and the short term outcomes were pooled and compared between open and minimally invasive techniques for esophagectomy. The review confirms that minimally invasive esophagectomy is feasible and safe. The findings indicate that there is a tendency towards better short term outcomes with respect to postoperative morbidity after minimally invasive esophagectomy when compared to conventional open surgery. While there were no randomized trials available for evaluation, the findings of this systematic review are based on case-controlled studies. Well designed trials are needed to assess the true value of minimally invasive surgery for esophageal cancer.
Chapter 3 describes the thromboembolic toxicity profile in a consecutive series of patients who underwent esophagectomy for esophageal adenocarcinoma. The incidence and severity of thromboembolic events (TEE) was compared between two groups; patients who were treated with surgery alone versus patients who underwent esophagectomy supplemented with neoadjuvant perioperative chemotherapy. The chemotherapy regimen consisted of epirubicin, cisplatin, and capecitabine (ECC). The study showed that patients who were treated with perioperative ECC chemotherapy experienced a significantly higher risk of developing clinical or subclinical thrombosis during their treatment period compared to patients who were treated with surgery alone. In multivariate analysis, ECC chemotherapy was identified as an independent risk factor for the development of a TEE. All TEEs that were diagnosed in the preoperative period were treated with antithrombotic therapy. A preoperative TEE did not increase the risk of a postoperative TEE, nor did it

affect the duration of postoperative hospital stay. Although clinical consequences were manageable and no grade 5 TEE toxicity (i.e. death) was observed, the study stresses the importance of adequate preoperative diagnostics which is not only focused on tumor response to therapy, but also at chemotherapy related thromboembolic toxicity. Untreated subclinical development of thrombosis in the preoperative period could seriously affect postoperative outcome of esophagectomy in patients who are treated with neoadjuvant ECC therapy.

The experimental study described in **chapter 4** was designed to evaluate how anastomotic strength is affected by adding a fibrin-thrombin coated patch to an esophagogastric anastomosis an animal model. In 54 rats, a hand sewn end-to-side anastomosis was created between the distal esophagus and the stomach. After completion of the anastomosis, subjects were randomized into two groups: a control group (without additional sealing) and a group in which a sealant was added to the anastomosis. The animals were sacrificed on different postoperative days (0, 3, 5 and 7 days). The experiments showed that application of a fibrin-thrombin coated patch increases the strength of the anastomoses. Since anastomotic leakage usually occurs within the first postoperative week, the findings indicate that sealing of the anastomosis has the potential to prevent leakage after esophagectomy in humans. It is hypothesized that reinforcement of the anastomosis with a sealant increases anastomotic strength and creates an environment which improves the anastomotic healing process due to mechanical support and the induction of cellular regenerative response. This could ultimately lead to a reduction of postoperative anastomotic leakages and consequent morbidity.

A relationship has been described between postoperative septic events and elevated immunological response to surgery. Particularly thoracic surgery and prolonged single lung ventilation contribute to an increased release of pro- and anti-inflammatory mediators. The randomized controlled trial, described in **chapter 5**, is the first trial of its kind that has evaluated both local and systemic immunological response to single lung ventilation in combination with continuous positive airway pressure (CPAP) on the collapsed lung. The study has shown that application of CPAP on the right collapsed lung during thoracoscopic esophagectomy reduces the local inflammatory response. A systemic response to CPAP was not noticeable in serum, which might explain the absence of a clinical effect. The pneumonia rates were comparable between the two groups. However, the study was designed to test immunological effects only. To detect a significant or clinically relevant influence of CPAP on clinical endpoints, such as postoperative pneumonia, a much larger number of patients is required.

Pneumonia is a common postoperative complication after esophagectomy and is often used as a clinical endpoint in literature. The etiology of postesophagectomy pneumonia is multi-factorial. Several risk factors, such as comorbidity and operative characteristics, are involved. The majority of postoperative pneumonias are diagnosed at the surgical ward where diagnosis is often difficult. **Chapter 6** evaluated the effect of four different

diagnostic determinants on the decision to treat pneumonia in a large series of patients who underwent esophagectomy. A multivariate analysis showed that clinicians are guided predominantly by pulmonary radiography findings and, to a lesser extent, by temperature recordings and leukocyte count. Sputum culture did not affect the decision to treat pneumonia. Based on these findings a simple scoring system was developed that, after appropriate validation, has the potential to aid diagnosis of pneumonia after esophagectomy at the hospital ward. Moreover, the proposed model could facilitate comparability of research in esophageal cancer surgery in which underreporting of pneumonia rates seems to occur due to the use of varying definitions for pneumonia.

PART TWO | PROGNOSIS

An important prognostic factor after esophageal cancer surgery is the circumferential resection margin (CRM). The finding of a negative margin at pathological examination improves chances of long term survival. However, there is a lack of consensus regarding the criteria of positive/negative CRM which has led to the use of different definitions in literature and clinical practice. In **Chapter 7**, the prognostic significance of two commonly used definitions is reviewed in a series of 167 patients. All patients underwent resection of a T3 adenocarcinoma with curative intent without having received neoadjuvant therapy. The pathologic tumor slides were reviewed and microscopic resection margins were assessed following the criteria as prescribed by the Royal College of Pathologists (RCP) and the College of American Pathologists (CAP). Patients were grouped into *CRM positive or CRM negative* according to both criteria. Overall survival and disease free survival were compared for each of the two criteria. In this uniform study population, the observation of tumor cells within 1 millimeter of the margin (following the RCP criteria) did not distinguish survival from patients with tumor cells further away from the margin. The results showed that survival was only significantly reduced when tumor cells were found at the circumferential margin of the resected specimen. A positive CRM according to the CAP criteria showed to be the strongest independent prognostic factor for both overall and disease free survival. It was concluded that the CAP criteria have to be considered the clinical standard for determination of the CRM in patients with T3 adenocarcinoma of the esophagus.

Chapter 8 examined the expression profile and prognostic significance of two important biomarkers in esophageal adenocarcinoma. COX-2 and VEGF play an important role in carcinogenesis, tumor progression and angiogenesis. COX-2 and VEGF expression was evaluated using immunohistochemistry in a Tissue Micro Array (TMA). Of each tumor, three representative tissue regions were carefully selected using the pathological slides of the resected specimen. With the TMA technique, a biopsy core (diameter 0.6mm) is punched out from each representative tissue region of the corresponding donor paraffin block which contains the original tumor. The biopsy cores are then transferred to a paraffin

TMA block. Sections can be sliced from the TMA block, which can be used for immunohistochemical staining. The technology facilitates high-throughput staining and all tissue cores can be simultaneously analyzed under identical laboratory and evaluation conditions. With this technique it was possible to determine expression of COX-2 and VEGF within the tumors of 154 patients. The results showed that COX-2 was over-expressed in 26.5% of patients with esophageal adenocarcinoma. Over-expression of VEGF was observed in 53.8% of tumors. The expression profiles were correlated with follow-up data. The study showed that both biomarkers were significantly associated with poor prognosis. Moreover, COX-2 was an independent prognostic marker for survival. Specific inhibition of the pathways in which COX-2 and/or VEGF are involved may affect treatment outcomes and prognosis. These findings support the use of COX-2 and VEGF inhibitors in future clinical studies.

CHAPTER 10

Future perspectives

Despite the advancements in currently available therapies and a high standard of care, treatment of esophageal cancer still carries poor prognosis and significant morbidity. As described in this thesis, improvements can still be achieved in various fields of esophageal cancer treatment.

The mainstay of treatment is surgery complemented with neodjuvant chemo(radio) therapy. Though aimed at improving long term survival, neoadjuvant chemotherapy also exposes patients to thromboembolic toxicity and associated morbidity. The rate of thromboembolic events among patients receiving neoadjuvant chemotherapy for esophageal cancer is alarmingly high. This finding is confirmed in a recent study by Moore *et al.*[1] In a large retrospective series of patients treated with cisplatin-based chemotherapy for various cancer types, the incidence of thrombosis was 18%. Events occurred mainly during early treatment course. The authors conclude that this rate is unacceptable, but do not describe the influence of preoperative thromboembolic events on surgical outcomes. In our series, all patients were adequately treated and preoperatively occurring events did not affect surgical outcomes. However, our findings clearly point out that careful monitoring for development of thrombotic disease is warranted. Moreover, prophylactic antithrombotic therapy should be considered when cisplatin-based regimens are used in the neoadjuvant setting of esophageal cancer treatment. In agreement with Moore *et al.*, prospective studies evaluating the use of prophylactic therapy are necessary.

Another dilemma presents itself in the choice of the optimal surgical approach for each individual patient. The surgeon is forced to balance conflicting aims. Morbidity and mortality can be minimized by opting for a limited transhiatal resection. But this strategy might come at the price of suboptimal oncological clearance with subsequent negative effect on long term survival. The alternative, more extended, approach entails a transthoracic procedure with a two-field lymphadenectomy. However, this approach is associated with increased morbidity and mortality rates. Irrespective of operative approach, it must be acknowledged that esophageal cancer surgery has reached a certain plateau. New surgical techniques will probably generate marginal improvements in long term survival of esophageal cancer. Survival rates will be affected mostly through the development of new neoadjuvant treatment strategies.

Research in esophageal cancer surgery will have to focus mainly on a further reduction of postoperative morbidity rates and on improving the quality of life after esophagectomy. Minimally invasive techniques that have been developed over the past two decades are gaining popularity among esophageal cancer surgeons.[2] It is thought that minimally invasive surgery for esophageal cancer is associated with less postoperative morbidity whilst preserving an oncologically safe resection. Promising reports of minimally invasive esophagectomy have been published.[3,4,5,6] These studies report reduced postoperative pain, reduced pulmonary complication rates, and shortened hospital and intensive care stay. However, its' superiority over conventional open surgery has not yet been confirmed

in well designed trials. The results of such trials are expected in the near future.[7-9]
Particular focus in esophageal cancer surgery should be directed at reducing frequently observed and feared complications, such as anastomotic leakage and pneumonia. Both are associated with significant morbidity and prolonged hospital stay.
The experimental study presented in this thesis showed an increased anastomotic strength during early postoperative recovery when a sealant was applied on the esophagogastric anastomosis. This finding gives way to a phase II clinical trial to evaluate this technique. Besides feasibility and safety, the short term clinical outcomes of such a study are anastomotic leakage and leakage related morbidity. There is no data available on the long term effects of esophagogastric sealing. Careful monitoring of possible stenosis should therefore be included in the design of such studies.
Many studies will focus on respiratory complications after esophagectomy, particularly pneumonia. However, current literature lacks a standardized definition of pneumonia. To facilitate comparability of studies, a uniform definition is urgently needed. The scoring system, which is proposed in this thesis, will have to be evaluated in a prospective cohort. Once validated, it has the potential to become a powerful tool in clinical research of esophageal cancer surgery.
One strategy to reduce respiratory complications after esophagectomy may lie in diminishing the use of single lung ventilation during thoracoscopic dissection. The reported trial showed a reduced local immune response when applying continuous positive airway pressure on the collapsed lung during single lung ventilation. This finding supports the idea of omitting complete single lung ventilation in thoracic esophagectomy. Thoracoscopic esophagectomy without the use of single lung ventilation has been reported in the literature.[10] This approach is performed with the patient in prone position. An important downside of the prone position is the limited access to the thorax when conversion to open surgery is necessary. Repositioning of the patient to the lateral decubitus position is required in case of hemorrhage, but this takes valuable time. Hence, the development of a surgical technique that uses double lung ventilation with patient positioning that does not limit access to the thorax in case of conversion deserves attention.
Improvements in therapy may also be expected from biomarkers which are potential candidates for targeted therapy. COX-2 and VEGF are highly expressed in esophageal cancer. Other studies have also reported on the prognostic value of both markers mainly in mixed study populations with adenocarcinoma and squamous cell carcinoma. It has also been suggested that COX-2 and VEGF are linked. In this thesis, the expression of COX-2 and VEGF is studied in a homogenous cohort of patients with esophageal adenocarcinoma, but a correlation between the two markers could not be confirmed. As esophageal adenocarcinoma and squamous cell carcinoma are two different disease entities, it is necessary to separate the two histological tumor types in future research into biomarker expression.
In light of the current focus on neoadjuvant chemo(radio)therapy, it would also be valuable

to assess biomarker expression in preoperatively collected biopsy material. Combined with data on response to therapy, this might enable patient selection for neoadjuvant therapy. Patients who are not likely to respond to therapy will not have to be exposed to toxicity and will not have to experience an unnecessary delay of surgery. However, the predictive accuracy of currently available biomarkers is far from sufficient. New studies will have to focus on the correlation between preoperatively identified biomarkers and response assessment. In addition, the current standards for pathological examination of surgical resection specimens may have to be re-evaluated in the future. Depending on the extent of response, the use of neoadjuvant therapies influences the assessability of the resection margins as well as their prognostic significance.

This thesis has aimed to provide new insights in the field of esophageal cancer surgery. As a small, but important contributor, it takes part in the ongoing efforts of around the world to improve outcomes for patients with this devastating disease. Future research will hopefully continue to provide new strategies that will further reduce therapy associated morbidity and enable an individualized approach towards esophageal cancer therapy.

REFERENCES

1. Moore RA, Adel N, Riedel E, Bhutani M, Feldman DR, Tabbara NE, Soff G, Parameswaran R, Hassoun H. High incidence of thromboembolic events in patients treated with cisplatin-based chemotherapy: a large retrospective analysis. J Clin Oncol. 2011;29(25)3466-3473.
2. Boone J, Livestro DP, Elias SG, Borel Rinkes IH, van Hillegersberg R. International survey on esophageal cancer: part I surgical techniques. Dis Esophagus. 2009;22(3)195-202.
3. Luketich JD, Alvelo-Rivera M, Buenaventura PO, Christie NA, McCaughan JS, Litle VR, Schauer PR, Close JM, Fernando HC. Minimally invasive esophagectomy: outcomes in 222 patients. Ann Surg. 2003;238(4)486-494.
4. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8)878-886.
5. Biere SS, Cuesta MA, van der Peet DL. Minimally invasive versus open esophagectomy for cancer: a systematic review and meta-analysis. Minerva Chir. 2009;64(2)121-133.
6. Verhage RJ, Hazebroek EJ, Boone J, van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64(2)135-146.
7. Biere SS, Maas KW, Bonavina L, Garcia JR, van Berge Henegouwen MI, Rosman C, Sosef MN, de Lange ES, Bonjer HJ, Cuesta MA, van der Peet DL. Traditional invasive vs. minimally invasive esophagectomy: a multi-center, randomized trial (TIME-trial). BMC Surg. 2011;11:2.
8. Briez N, Piessen G, Bonnetain F, Brigand C, Carrere N, Collet D, Doddoli C, Flamein R, Mabrut JY, Meunier B, Msika S, Perniceni T, Peschaud F, Prudhomme M, Triboulet JP, Mariette C. Open versus laparoscopically-assisted oesophagectomy for cancer: a multicentre randomised controlled phase III trial - the MIRO trial. BMC Cancer. 2011;11:310.
9. van der Sluis PC, Ruurda JP, van der Horst S, Besselink MG, Verhage RJ, Joore JCA, Kroese AC, van Leeuwen MS, Lolkema MP, Reerink O, Steenhagen E, Vleggaar FP, Schipper ME, Siersema PD, ten Kate FJ, Koffijberg H, Borel Rinkes IB, van Hillegersberg R. ROBOT trial: Robot-assisted minimally invasive thoraco-laparoscopic esophagectomy vs. transthoracic esophagectomy. unpublished trial protocol. 2011.
10. Palanivelu C, Prakash A, Senthilkumar R, Senthilnathan P, Parthasarathi R, Rajan PS, Venkatachlam S. Minimally invasive esophagectomy: thoracoscopic mobilization of the esophagus and mediastinal lymphadenectomy in prone position--experience of 130 patients. J Am Coll Surg. 2006;203(1)7-16.

CHAPTER 11

Summary in Dutch

Nederlandse samenvatting

De incidentie van slokdarmkanker neemt gestaag toe. In de Westerse Wereld is dit voornamelijk toe te wijden aan de stijgende incidentie van het adenocarcinoom van de slokdarm. De verwachting is dat deze stijging in de toekomst door zal zetten. Het chirurgisch verwijderen van de slokdarm, al dan niet aangevuld met chemo(radio)therapie, biedt vooralsnog de enige curatieve oplossing voor het resectabel slokdarmcarcinoom. Ondanks vooruitgangen in peri-operatieve zorg, de ontwikkeling van minder invasieve chirurgische technieken en verbetering van chemo(radio)therapeutische behandelingsschema's, stijgt de vijf jaar overleving van potentieel te genezen slokdarmkanker nauwelijks boven de 40% uit. Daar wordt aan toegevoegd dat de behandeling van slokdarmkanker vaak gepaard gaat met een gecompliceerd beloop en relatief hoge mortaliteit. Verbeteringen in de behandeling en de prognose van patiënten met deze ingrijpende ziekte zijn dus van groot belang. Postoperatieve complicaties en vernieuwende inzichten in lange termijn overleving staan daarom centraal in dit proefschrift.

DEEL ÉÉN | PERIOPERATIVE MORBIDITEIT

Hoofdstuk 2 beschrijft een systematisch uitgevoerde literatuurstudie naar minimaal invasieve behandeling van slokdarmkanker met behulp van een kijkoperatie. Een tiental vergelijkende onderzoeken naar minimaal invasieve slokdarmresectie en traditionele open slokdarmresectie werden geëvalueerd. Deze studie liet zien dat minimaal invasieve slokdarmresectie veilig uitgevoerd kan worden en dat er een trend te zien is naar een verbetering van korte termijn uitkomsten bij gebruik van minimaal invasieve technieken, zoals een vermindering van het aantal complicaties en een verkorting van opnameduur. Om dergelijke bevindingen te bevestigen zijn goed opgezette en gerandomiseerde studies nodig.

Voorafgaand aan de operatie wordt in veel gevallen chemotherapie gegeven waarvan bewezen is dat het een betere kans op overleving geeft na slokdarmresectie. Uit het onderzoek beschreven in **hoofdstuk 3** is gebleken dat er een verhoogd risico bestaat op tromboembolische complicaties (het ontstaan van stolsels in de bloedvaten) bij perioperatief gebruik van de chemotherapeutica epirubicine, cisplatinum en capecitabine volgens het ECC schema. Bij een derde van de behandelde patiënten werden trombotische afwijkingen ontdekt. Op basis van de literatuur lijkt cisplatinum hiervoor verantwoordelijk te zijn. In een deel van de patienten gaven deze bloedstolsels nog geen klachten, maar als gevolg van deze stolsels kunnen patiënten een trombosebeen of, in ernstigere gevallen, longembolieën ontwikkelen. De resultaten uit dit onderzoek geven aan dat er mogelijk een indicatie bestaat voor preventieve antistollingstherapie bij patiënten die preoperatief behandeld worden met cisplatinum om de vorming van trombose tegen te gaan en zodoende ernstige complicaties te voorkomen.

Om de tumor uiteindelijk in zijn geheel weg te nemen worden het bovenste deel van de

maag en het grootste deel van de slokdarm operatief verwijderd. Van de maag wordt dan een nieuwe slokdarm geconstrueerd (buismaag) welke doorgaans in de hals verbonden wordt aan het resterende bovenste deel van de slokdarm. Zodoende wordt de continuïteit van het maag-darmkanaal hersteld. Een belangrijke en gevreesde complicatie na slokdarmchirurgie betreft lekkage van de verbinding tussen de buismaag en het resterende deel van de slokdarm (naadlekkage). Ondanks verbeteringen in chirurgische technieken komen postoperatieve naadlekkages bij ruim 15% van de geopereerde patiënten voor. Naadlekkage geeft een verhoogd risico op het ontstaan van andere complicaties en geven op lange termijn een verhoogde kans op littekenvorming en vernauwing (stenose) van de naad. Snelle genezing van deze kwetsbare naad is dus van essentieel belang. In de experimentele dierstudie van **hoofdstuk 4** is gekeken naar het effect van een matje met humane stollingsfactoren op de sterkte en genezing van de naad. Toevoeging van dit matje blijkt de verbinding tussen slokdarm en maagweefsel te versterken tijdens de vroege postoperatieve fase. Tevens waren er aanwijzingen voor een toegenomen activiteit van cellen die verantwoordelijk zijn voor het remodellerings- en genezingsproces. Deze bevindingen geven aanleiding tot klinisch onderzoek dat uit zal moeten wijzen of hiermee ook het aantal naadlekkages na slokdarmresectie verminderd kan worden.

Longontsteking (pneumonie) is een andere veel voorkomende complicatie na slokdarmresectie. Postoperatieve pneumonie leidt tot een verlenging van de ziekenhuisopname en is geassocieerd met een verhoogd risico op postoperatieve sterfte. Beschreven oorzaken voor het ontstaan van pneumonie na slokdarmresectie zijn langdurige beademing op één long (voor optimaal zicht op de slokdarm in de borstholte) en manipulatie van de gecollabeerde long tijdens de operatie. In **hoofdstuk 5** wordt een gerandomiseerde klinische studie beschreven naar het effect van een continue positieve ademwegdruk (CPAP) op de gecollabeerde long. Met deze techniek wordt de gecollabeerde long in lichte mate opgeblazen zonder hem in zijn geheel te beademen. Dit heeft als doel longschade ten gevolge van kunstmatige beademing te verminderen. Longschade vertaalt zich in een reactie van het immuunsysteem. Uit deze studie is gebleken dat het niveau van immuunrespons in de longen lager is wanneer deze techniek wordt toegepast. Mogelijk kan dit het aantal postoperatieve longontstekingen reduceren. Nader onderzoek zal dit beoogde effect uit moeten wijzen.

Het optreden van longontsteking na slokdarmresectie vindt voornamelijk plaats op de verpleegafdeling tijdens de postoperatieve herstelfase. Het diagnosticeren van een pneumonie op de afdeling is lastig en in de literatuur worden verschillende definities gebruikt voor het beschrijven van pneumonieën. **Hoofdstuk 6** beschrijft het gebruik van vier veelgebruikte diagnostische determinanten die de arts op de afdeling ter beschikking staan: lichaamstemperatuur, het aantal witte bloedcellen (leukocyten), een longfoto (thoraxfoto) en een kweek van opgehoest slijm (sputumkweek). Uit een prospectieve analyse van 185 patiënten na slokdarmresectie blijkt dat de thoraxfoto de meest voorspellende waarde heeft voor de behandeling met antibiotica voor pneumonie,

aangevuld met de gegevens van een verhoogde temperatuur en leukocyten aantal. De sputumkweek wordt vaak wel uitgevoerd en is van essentieel belang voor het bepalen van het verantwoordelijke micro-organisme. Het blijkt echter niet van invloed te zijn op de beslissing om te behandelen voor pneumonie. Toch wordt deze bepaling in onderzoek veelvuldig gebruikt voor het definiëren van pneumonie als primaire uitkomstmaat. Het gebruik van een positieve sputumkweek als vereiste voor de diagnose pneumonie leidt tot een onderrapportage van het aantal pneumonieën in de literatuur. Daarom is op basis van de uitkomsten van deze studie een scoremodel ontwikkeld dat mogelijk kan helpen met het diagnosticeren van pneumonie na slokdarmresectie op de verpleegafdeling. Op voorwaarde dat dit scoremodel in de praktijk gevalideerd wordt, kan het model de vergelijkbaarheid van toekomstige klinische studies faciliteren doordat het een eenduidige definitie biedt van pneumonie.

DEEL TWEE | PROGNOSE

Naast optimalisatie van het postoperatief herstel is het voor patiënten van belang dat zij een pragmatische en geïndividualiseerde inschatting krijgen van hun prognose. Belangrijke informatie die bepalend is voor de prognose komt voort uit pathologisch onderzoek van de verwijderde tumor onder de microscoop. Een van de belangrijkste prognostische parameters betreft de beoordeling of de tumor in zijn geheel (radicaal) verwijderd is. Hiertoe wordt zowel macroscopisch als microscopisch gekeken naar de snijranden van de verwijderde slokdarm (resectiepreparaat) om te beoordelen of er tumorcellen aanwezig zijn in de snijrand (resectiemarge). Het College of American Pathologists (CAP) uit de Verenigde Staten en het Royal College of Pathologists (RCP) uit het Verenigd Koninkrijk hanteren verschillende criteria voor deze microscopische beoordeling van de resectiemarge. Volgens de RCP criteria is de verwijdering van de tumor *niet* radicaal wanneer er tumorcellen gevonden worden binnen 1 millimeter afstand van de resectiemarge. De CAP criteria bepalen daarentegen dat de tumor pas irradicaal verwijderd is wanneer de tumorcellen zich in de resectiemarge bevinden (binnen 1 millimeter van de marge is dus wel radicaal). De definites worden door elkaar gebruikt in de literatuur. **Hoofdstuk 7** evalueert deze twee definities van radicaliteit in een grote groep patiënten die geopereerd zijn voor een adenocarcinoom van de slokdarm. Uit het onderzoek is gebleken dat er bij het gebruik van de RCP criteria geen verschil is in lange termijn overleving tussen patiënten met een positieve en negatieve resectiemarge. De beoordeling van de resectiemarge volgens de CAP criteria gaf wel een significant verschil in overleving aan en geeft derhalve een betere voorspelling van de prognose. Voor toekomstig onderzoek is het daarom aan te raden om de CAP criteria te hanteren.

Hoofdstuk 8 behandelt de prognostische waarde van twee moleculaire eigenschappen van het slokdarm adenocarcinoom. De moleculaire markers COX-2 en VEGF zijn betrokken bij het ontstaan van kanker en kunnen dus verhoogd aanwezig zijn in tumorcellen. Deze

bio-markers kunnen worden aangekleurd door middel van immunohistochemische kleuringen en het resultaat van deze kleuringen kan beoordeeld worden onder de microscoop. De mate van aankleuring is representatief voor de expressie van deze tumormarkers. In het onderzoek is gekeken naar de aanwezigheid en de prognostische waarde van deze markers. Een verhoogde VEGF en COX-2 expressie werd gezien in, respectievelijk, de helft en een kwart van 154 patiënten met een adenocarcinoom. Een verhoogde expressie was gecorreleerd aan een slechtere langetermijn overleving voor beide biomarkers. Deze resultaten bevestigen het mogelijke nut van VEGF en COX-2 in toekomstig onderzoek naar de ontwikkeling van nieuwe kankerspecifieke therapieën.

ADDENDA

A

What are the indications and limits of video-assisted lymphadenectomy in the surgical treatment of esophageal cancer?

Annals of the New York Academy of Sciences. 2011;1232:258-60

Roy JJ Verhage
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From:
Komanduri S, Deprez PH, Atasoy A, Hofmann G, Pokieser P, Ba-Ssalamah A, Collard JM, Wijnhoven BP, Verhage RJ, Brücher B, Schuhmacher C, Feith M, Stein H.
Barrett's esophagus: treatments of adenocarcinomas I.
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Rapidly increasing incidence of esophageal adenocarcinoma has boosted research in this rapidly evolving field of cancer medicine. However, survival remains relatively poor. A probable explanation for this low survival is that lymph node involvement in esophageal cancer is common and already occurs in early stage disease. Lymph node metastases are found in less than 2% of T1a tumors, but for T1b disease this number goes up to 20%. For T3 tumors, the number of patients with positive lymph nodes is even 80%. These figures support endoscopic resection for T1a stage disease, but warrant surgical treatment with extensive lymphadenectomy for higher stage disease to accomplish oncological clearance.

Current diagnostic strategies are lacking enough predictive value for identifying patients with and without lymph node involvement. Endoscopic ultrasound for the detection of N-stage has a sensitivity of 80% and specificity of 70% for regional lymph nodes.[1] Computed tomography is an unreliable tool to detect small lymph node metastases, which are frequently observed during pathological assessment of tumor specimens.

Surgical treatment should therefore be directed towards a radical tumor resection as well as proper lymphadenectomy. In our own series, the robot assisted thoracolaparoscopic approach yielded a median of 29 lymph nodes per patient (figure 1a).[2] From 47 patients, 35 had a distal tumor in the lower third of the esophagus or at the gastro-esophageal junction. One third of the positive lymph nodes in these patients were found proximally in the mediastinum (figure 1b). These lymph nodes would not have been resected when a transhiatal procedure was performed. This further illustrates the need for extensive lymphadenectomy, even when distal tumors are concerned.

The limits in diagnostics and the high rate and location of lymph node involvement warrant lymph node dissection for esophageal cancer. Several surgical strategies exist, which can

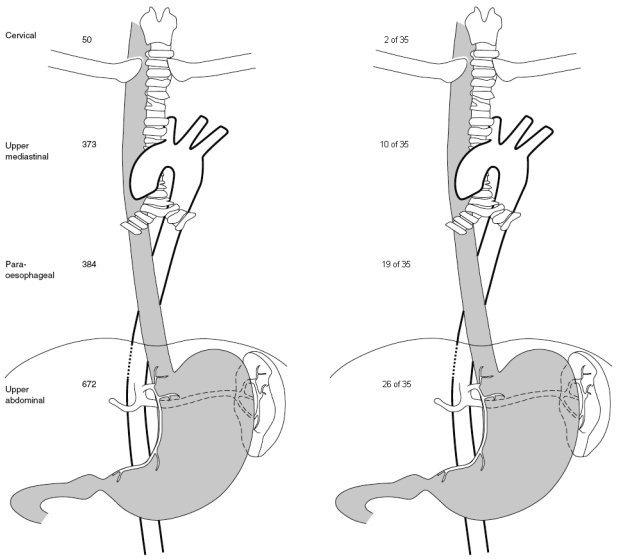

Figure 1 Lymph node distribution
Legend: (a) Distribution of 1479 lymph nodes dissected in 47 patients who underwent robot assisted thoracolaparoscopic esophagectomy. (b) Location of lymph node metastases in 35 patients with a distal or gastro-esophageal junction tumor.
(from Boone J *et al.* 2009, with permission)

be grouped in the open transhiatal approach and the transthoracic approach and their minimally invasive counterparts, laparoscopic and thoracolaparoscopic esophagectomy. The largest trial to date showed improved survival in patients who underwent transthoracic esophagectomy when compared to transhiatal surgery. Long term results showed significantly improved 5 year survival for patients with 1-8 positive lymph nodes when operated on through the transthoracic approach.[3] These results support that more extensive surgical lymphadenectomy improves oncological outcome.

However, transthoracic surgery is associated with higher morbidity than transhiatal surgery. To limit surgical trauma and morbidity, whilst at the same time achieving oncological clearance, thoracoscopic techniques offer an alternative. Data from randomized trials is not available yet, but review of current literature supports that minimally invasive esophagectomy reduces trauma and morbidity (table 1).[4] Furthermore, minimally invasive techniques improve the surgeon's vision of the operative field. Several other studies have also shown that lymph node harvest in video-assisted lymphadenectomy is at least comparable to open surgery.[2,5]

For the surgeon, the advantage of minimally invasive esophagectomy with extended lymphadenectomy lies in the improved magnified vision through angles that cannot be reached at open surgery. For the patient, the advantages include reduced blood loss, reduced postoperative pain, shorter recovery, less postoperative morbidity and improved cosmetics.

Some important limits to minimally invasive techniques for esophagectomy must be mentioned. This type of surgery requires a surgeon with advanced endoscopic skills. The learning curve for developing these skills is far from steep. Much training is necessary before a surgeon is able to perform at such an advanced level. However, with the advent

Table 1 Average short-term postoperative outcomes for open transthoracic esophagectomy (TTE) versus thoraco-laparoscopic esophagectomy (TLSE)

		TTE (n=203)	TLSE (n=78)
Operating time total	minutes	331	384
EBL	milliliters	630	317
ICU stay	days	4.0	4.4
Hospital stay	days	18.2	14.9
Lymph node harvest	n	16.1	17.4
Pulmonary complications	%	26.5	15.3
Complications	%	67.3	57.1
Perioperative mortality	%	3.8	1.3

Values are weighted means (adapted from Verhage et al. 2009, with extra data added from Smithers et al. 2007). EBL - estimated blood loss; ICU - intenstive care unit; TTE - transthoracic esophagectomy; TLSE - thoraco-laparoscopic esophagectomy; n - number.

of robot assisted surgery, esophagectomy has become less demanding for the surgeon. Ergonomics are significantly improved and the surgeon is provided with a three dimensional vision, further improving the view of the operative field. Additionally, the surgeon is able to control articulating instruments with great precision devoid of tremor. Robot assisted surgery overcomes many limits of conventional endoscopic surgery and is a suitable technique for performing complex procedures such as en bloc esophagectomy with extensive lymphadenectomy.[2]

In conclusion, lymphadenectomy in patients with esophageal cancer is warranted for achievement of complete oncological clearance. Such extensive surgery can be facilitated by minimally invasive or video-assisted techniques in the hands of an experienced endoscopic surgeon. Further improvements in this field might be achieved with robot assisted surgery.

REFERENCES

1. van Vliet EP, Heijenbrok-Kal MH, Hunink MG, Kuipers EJ, Siersema PD. Staging investigations for oesophageal cancer: a meta-analysis. Br J Cancer. 2008;98:547–57.
2. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8):878-86.
3. Omloo JM, Lagarde SM, Hulscher JB, Reitsma JB, Fockens P, van Dekken H, Ten Kate FJ, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the mid/distal esophagus: five-year survival of a randomized clinical trial. Ann Surg. 2007;246(6):992-1000.
4. Verhage RJ, Hazebroek EJ, Boone J, Van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64(2):135-46.
5. Puntambekar SP, Agarwal GA, Joshi SN, Rayate NV, Sathe RM, Patil AM. Thoracolaparoscopy in the lateral position for esophageal cancer: the experience of a single institution with 112 consecutive patients. Surg Endosc. 2010;24:2407–14.

B

What is the role of robotic surgery in the treatment of esophageal cancer?

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Radical surgical resection of the esophagus and surrounding lymph nodes offers the best chance for cure in patients with locoregional disease.[1] Optimal treatment for esophageal cancer, therefore, consists of transthoracic en bloc esophagectomy with an extensive mediastinal lymph node dissection (LND). This approach through thoracotomy is accompanied by significant morbidity, mainly consisting of cardiopulmonary complications. To reduce surgical trauma and morbidity of open transthoracic esophagectomy, minimally invasive esophagectomy (MIE) techniques have been introduced.

An international survey showed that thoracic esophagectomy with a 2-field LND is the most commonly applied extent of LND.[2] The survey also revealed that 40% of the surgeon responders routinely use minimally invasive techniques for esophagectomy.

With regard to MIE, review of the literature shows a substantial decrease in blood loss, complication rate and hospital stay.[3] However, conventional scopic techniques have important limitations, such as a 2-dimensional view, a disturbed eye-hand-coordination and a decrease in degrees of freedom due to large, rigid instruments.

Robotic systems have been developed to overcome the limitations of standard minimally invasive procedures.[4] The DaVinci® robotic system (Intuitive, Sunnyvale, California, USA) provides a three-dimensional, tenfold magnified view of the operating field. It filters the tremor of the surgeon, restores the natural eye–hand coordination axis as a result of the ergonomically designed surgeon's console, and offers more degrees of freedom through its articulating scopic surgical instruments. During esophagectomy, the robotic platform enables the surgeon to perform an accurate mediastinal dissection of the esophagus with surrounding lymph nodes in a confined surgical field.

In our tertiary referral center the robot assisted thoracolaparoscopic approach is routinely used for patients with resectable cancer of the esophagus. The patient is positioned in the left lateral decubitus position, tilted 45° towards the prone position (figure 1).

The robotic system is placed at the dorsocranial side of the patient. Three robotic and two assistant's instrument ports are placed. After the pulmonary ligament has been divided, the parietal pleura is dissected at the anterior side of the esophagus from the diaphragm up to the azygos arch. The azygos arch is ligated and dissection of the parietal pleura continues above the arch for a right paratracheal lymph node dissection.

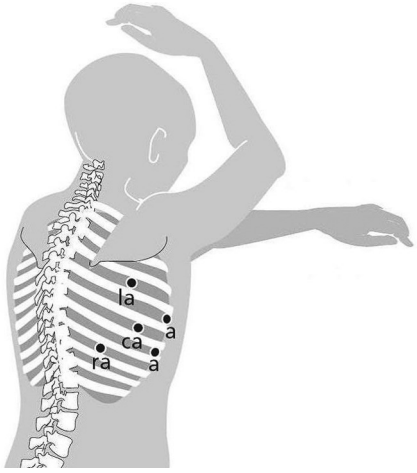

Figure 1 Trocar arrangement during robot assisted thoracoscopic phase
Legend: la, left robotic arm; a, assistant port; ca, robotic camera arm; ra, right robotic arm. (from Boone *et al.* 2009, with permission)

Subsequently, the parietal pleura is dissected at the posterior side of the esophagus cranially to caudally along the azygos vein, including the thoracic duct to avoid postoperative chyle leakage. At the level of the diaphragm, the thoracic duct is clipped with a 10-mm endoscopic clipping device (Endoclip™ II; Covidien, Mansfield, Massachusetts, USA). A Penrose drain is then placed around the esophagus to facilitate esophageal mobilization. In this way, the esophagus can be resected en *bloc* with the surrounding mediastinal lymph nodes and the thoracic duct from the diaphragm up to the thoracic inlet. For LND the robotic system provides an excellent view at angles which cannot be reached during open surgery. LND includes the right-sided paratracheal (lymph node station 2R), tracheobronchial (lymph node station 4), aortopulmonary window (station 5), carinal (station 7) and peri-esophageal (station 8) lymph nodes. The abdominal phase of the operation is performed with conventional laparoscopy (figure 2), dissecting the greater and lesser curvature of the stomach, crux and celiac trunk. LND includes lymph nodes surrounding the left gastric artery and the lesser omental lymph nodes. The resected specimen is removed through a 7-cm transverse transabdominal incision. Linear staplers (GIA 80, 3.8 mm; Covidien) are used to create a gastric conduit 3–4 cm wide with oversewing of the staple line. Through a left-sided vertical incision along the sternocleidoid muscle, a handsewn end-to-side anastomosis is created between the gastric tube and the cervical esophagus using 3/0 polydioxanone single-layer running sutures. No formal cervical LND is carried out unless lymph node metastases are suspected macroscopically. Our first series reported 47 patients who underwent robot assisted thoracolaparoscopic esophagectomy (RTE).[5] Conversion to thoracotomy was necessary in 7 patients. Median operating time was 450 minutes (360–550). Median blood loss during thoracoscopy was 250ml (0–800) and 625ml (150–5300) for the entire procedure. A learning curve was observed, illustrated by a significant decrease in total blood loss was between the first 23 and second 24 patients (median 900 vs. 450 ml respectively; $P < 0.001$) and a reduction of operating time (median 7.5 vs. 7.0 h; $P = 0.024$). Patients were ventilated for a median of 1 day (0–126). Median ICU stay was 3 days (0–136) and hospital stay 18 days (10–182). Though not significant, the first 23 patients had a higher pulmonary complication rate than the last 24 (13 of 23 vs. 8 of 24; P=0.147).

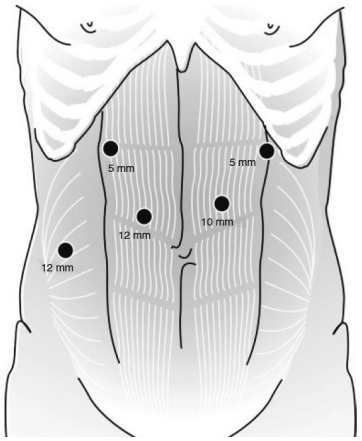

Figure 2 Trocar arrangement during conventional laparoscopic phase
Legend: The camera is inserted through the 10-mm trocar port and two 5-mm trocars are used as working ports. The liver retractor is inserted through the 12-mm right pararectal trocar port and the harmonic scalpel introduced through the 12-mm paraumbilical port. (from Boone et al. 2009, with permission)

A median of 29 (range 8–68) lymph nodes was dissected and R0 resection was achieved in 36 patients. Twenty-three patients had stage IVa disease. After a median follow-up of 35 months, median disease-free survival was 15 months (95%CI 12-18).
Robot-assisted thoracoscopic esophagectomy in conjunction with conventional laparoscopy has shown to be technically feasible providing oncological clearance whilst minimizing blood loss. Despite their short history in the field of esophagectomy, MIE and robot assisted esophagectomy offer promising results with outcomes which are at least comparable to conventional open surgery. Future research will focus on long term outcomes and the comparison of MIE with open surgery.

REFERENCES

1. Mariette C, Piessen G, Triboulet JP. Therapeutic strategies in oesophageal carcinoma: role of surgery and other modalities. Lancet Oncol. 2007; 8: 545-53.
2. Boone J, Livestro DP, Elias SG, Borel Rinkes IHM, van Hillegersberg R. International survey on esophageal cancer: part I surgical techniques. Dis Esoph. 2009;22(3)195–202.
3. Verhage RJ, Hazebroek EJ, Boone J, Van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64(2):135-46.
4. Ruurda JP, van Vroonhoven TJ, Broeders IA. Robot-assisted surgical systems: a new era in laparoscopic surgery. Ann R Coll Surg Engl. 2002;84:223-6.
5. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8):878-86.

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Robot-Assisted Thoracolaparoscopic Esophagectomy; The Netherlands

Thoracic Robotic Surgery: Atlas and Guide.

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ABSTRACT

For locally advanced esophageal cancer, radical surgical resection is the mainstay of treatment. Lymph node metastases occur along the entire tract of the esophagus in an early stage. Optimal surgical treatment includes a two field thoraco-abdominal en bloc esophagectomy with an extensive mediastinal and truncal lymph node dissection.

To reduce surgical trauma and morbidity of traditional open esophagectomy, techniques for minimally invasive esophagectomy have been introduced. However, conventional scopic surgery is limited by 2-dimensional vision, reduced dexterity and limited degrees of freedom. Robotic systems have been developed to overcome such limitations. The robotic platform enables the surgeon to perform complex minimally invasive surgical procedures. Advantages are reduced blood loss and fast postoperative recovery.

This chapter describes the indications and preoperative considerations for robot-assisted thoracolaparoscopic esophagectomy. Furthermore, anesthesiological management is discussed, addressing important intraoperative issues such as single lung ventilation and fluid management.

The three-stage operative procedure is described in detail. The thoracoscopic phase is performed using the robotic DaVinci Si system (Intuitive Surgical Inc., Sunnyvale CA, USA). The laparoscopic phase is performed with conventional laparoscopy. A gastric conduit is created extracorporally and an esophagogastric anastomosis is formed at the level of the neck through a left sided cervical incision.

Additionally, the clinical care of patients after esophagectomy is discussed with a specific focus on anastomotic leakage and chylous leakage.

INTRODUCTION

Esophageal cancer is the 8th most common type of malignancy and the 6th most common cause of cancer mortality in the World.[1] In 2002, approximately 462,000 patients were newly diagnosed with esophageal cancer.[1] The two most common histologic subtypes are esophageal squamous cell carcinomca (ESCC), arising from dysplastic squamous epithelium of the esophagus and esophageal adenocarcinoma (EAC), originating from dysplasia in columnar-lined esophagus with intestinal metaplasia (i.e. Barrett's esophagus).[2,3] The incidence of esophageal cancer has rapidly increased over the past decades, particularly due to a rise in EAC.[4] Worldwide the incidence of ESCC is highest.[1] Radical surgical resection is the mainstay of treatment for patients diagnosed with locally advanced esophageal cancer, offering the best chance of cure.[5] Symptoms, such as dysphagia and retrosternal discomfort, arise only when the tumor is large enough to obstruct the esophageal lumen. Therefore, patients are frequently diagnosed at an advanced stage of disease. Consequently, less than half of patients are eligible for surgery due to tumor ingrowth into adjacent structures or due to the presence of distant metastases. As the esophagus has a unique longitudinal lymphatic drainage system in the submucosal layer, lymph node metastases of esophageal cancer can occur along the entire tract of the esophagus from the cervical to the abdominal part. Optimal treatment for esophageal cancer, therefore, consists of transthoracic en bloc esophagectomy (TTE) with an extensive mediastinal lymph node dissection (LND). This approach through thoracotomy is accompanied by significant morbidity, mainly consisting of cardiopulmonary complications.

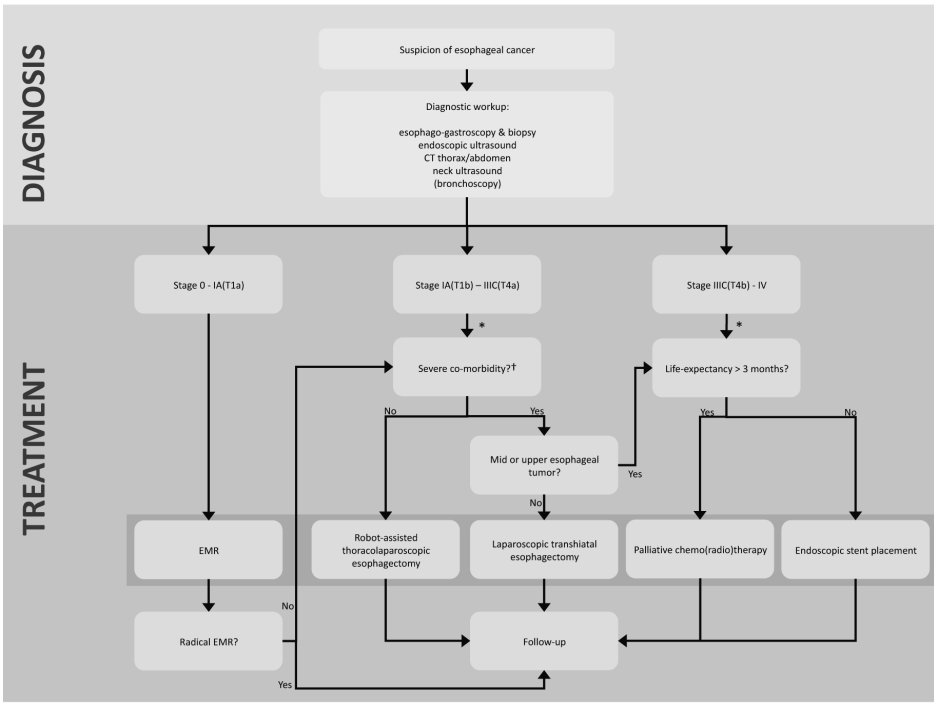
To reduce surgical trauma and morbidity of open transthoracic esophagectomy, less invasive surgical techniques such as transhiatal esophagectomy (THE) and minimally invasive esophagectomy (MIE) have been introduced. A randomized controlled trial by Hulscher *et al.* comparing the transthoracic versus transhiatal esophagectomy has shown the latter to have a lower complication rate.[6] However in the transhiatal approach a limited lymph node dissection is performed, with no dissection of the upper mediastinal lymph nodes.[7,8] Approximately 30% of lymph node metastases in patients with cancer of the distal esophagus or gastro-esophageal junction (GEJ) are located in the upper mediastinum.[9] The transhiatal approach does not include these nodes leading to a trend towards a better survival for transthoracic over transhiatal esophagectomy.[10,11] Other studies have mixed results, not clearly demonstrating superiority of the transthoracic approach.[12-14]

Recent analyses of the MIE to date have shown a decreased operative blood loss, complication rate and hospital stay.[15-17] However, conventional endoscopic surgery has important limitations, such as a 2-dimensional view, a cumbersome hand-eye-coordination and limited degrees of freedom due to the rod-like, inflexible instruments. Robotic systems have been developed to overcome these limitations.[18,19] During esophagectomy, the robotic platform enables the surgeon to perform an accurate mediastinal dissection.

It allows en *bloc* resection of the esophagus with its surrounding mediastinal fat and lymphatic tissue, which often harbour metastatic disease. The available space for this dissection is often limited. In this particular aspect, the robotic approach excels in comparison with open thoracotomy or other MIE techniques. Robot-assisted thoracoscopic esophagectomy (RTE) in conjunction with conventional laparoscopy has shown to be technically feasible.[9,20] Moreover, it provides sufficient oncological resection and is associated with low blood loss.[9,21]

INDICATIONS

Appropriate patient selection is essential to a successful esophageal surgery program. Thirty to forty percent of esophageal cancer patients are eligible and/or can be prepared to undergo an esophagectomy addressing their comorbidities and considering their tumor stage (flowchart Treatment). The risk of postoperative complications is associated with advanced age and comorbidity. Additionally, prolonged single-lung ventilation during the thoracic phase may further increase the risk of pulmonary complications.



Flowchart Workup for diagnosis and treatment of esophageal carcinoma.
Legend: * The application of (neo-)adjuvant chemo(radio)therapy is omitted, but should be considered and discussed for each individual case by a multidisciplinary team. †ASA score > 3, preoperative pulmonary evaluation; FEV1 60-80% → pulmonary training, FEV1 <60% → transhiatal procedure

Although the minimally invasive approach may offer a potentially curative surgical resection to a greater percentage of patients, careful patient selection is critical. The presence and degree of comorbidities such as cardiovascular and pulmonary diseases and diabetes must be assessed in each individual case.
Furthermore, if a patient has an esophageal carcinoma of the cardia or distal esophagus (lower third) a laparoscopically-assisted transhiatal may be considered, rather than a robotic transthoracic approach. Patients with distal early stage (T1) tumors and no evidence of distant adenopathy and no to minimal local adenopathy on endoscopic ultrasonography (EUS), and/or in whom endoscopic mucosal resection (EMR) did not achieve a complete resection, are also candidates for laparoscopic transhiatal esophagectomy.

PREOPERATIVE CONSIDERATIONS

Routine preoperative diagnostic investigations (flowchart Diagnosis) include esophagogastroscopy with as necessary tumor biopsy, endoscopic ultrasound (EUS), computed tomography (CT) of the chest and abdomen, ultrasonography of the neck with fine-needle aspiration of suspicious of cervical nodes, electrocardiography and lung function testing. Bronchoscopy is performed if airway involvement is suspected and [18F] fluorodeoxyglucose (FDG) positron emission tomography (PET) with CT fusion when metastases beyond the surgical field or organ metastases are suspected on CT. Any FDG-avid lesions are biopsied to confirm the presence of metastasis.
The proximal and distal borders of the tumor in the esophagus circumferential involvement, degree of obstruction, skip lesions and their location and the health of the remaining mucosa is determined by gastroscopy or EUS. Tumors are considered upper esophageal when they were located between 18 and 24 cm from the incisor teeth, mid oesophageal when located between 24 and 32 cm, and lower esophageal (including tumors of the GEJ) when located between 32 and 40 cm.[22]

ANESTHESIA MANAGEMENT

Perioperative management involves a multi-specialty team consisting of surgical, anesthetic, critical care, physiotherapy, dietetic and nursing specialists. Standardized perioperative clinical protocols provide a framework for patient management aiming to improve efficiency and outcome. However, there is little evidence from randomized clinical trials to guide management of anaesthesia for esophagectomy. The section below highlights the anaesthesia for robot-assisted thoracoscopic esophagectomy (RTE) conducted in the University Medical Center Utrecht.

PREOPERATIVE

All patients planning to undergo RTE are seen by an anaesthesiologist in the Preoperative Clinic. The physical status of the patient is assessed and preoperative testing is guided by institutional guidelines. Patients with the presence of and increased degree of perioperative complications (e.g. cardiovascular complications) will be referred for additional specialty care, as necessary, and treatment as directed by the anaesthesiologist.

PEROPERATIVE

Thoracic epidural analgesia (TEA) and its effects during the postoperative period have been studied extensively. TEA most likely decreases the risk of postoperative respiratory failure and results in improved pain control.[23,24] Furthermore, TEA may increase the blood supply to the esophago-gastric anastomosis area after esophagectomy.[25] Although there are no specific publications on the effects of TEA during minimally invasive esophagectomy, the advantages of TEA in the postoperative course of open esophagectomy can probably be extrapolated to thoracoscopic esophagectomy.

Normally, the epidural catheter is placed between the fifth and the eight thoracic vertebrae. After insertion of the catheter, a test dose of lidocaine with epinephrine is administered to exclude subarachnoid or intravasal placement of the catheter. To avoid the risk of sympathicolysis, no additional boluses of local anaesthetics are given. Usually epidural sufentanil is used intraoperatively and a continuous infusion of bupivacaine and morphine is applied postoperatively. To enable selective deflation of the right lung during the thoracoscopic phase, patients are intubated with a left-sided double-lumen tube. Patients receive two large-bore peripheral cannulae, a central venous line in the right internal jugular vein, an arterial line, a urinary catheter and a nasogastric tube. Furthermore ASA standards for basic anaesthetic monitoring are applied.[26] Antibiotic prophylaxis is provided by i.v. administration of 2000 mg cefazolin and 500 mg metronidazole. Thirty minutes before incision, 10 mg/kg methylprednisolone is administered to minimize postoperative pulmonary complications.[27]

Patients receive either propofol or volatile anaesthesia at the discretion of the attending anaesthesiologist. During the thoracoscopic phase of the operation patients are positioned in the left lateral decubitus position, and selective ventilation of the left lung is instituted. Continuous intravenous muscle relaxation is used to facilitate dissection of the esophagus along the trachea, azygos vein, aorta and pulmonary veins as sudden, unexpected movements of the patient could have detrimental effects. The patient must be protected against inadvertent contact from the motions of the robotic arms. After the instruments are connected to the arms of the robot and are placed inside the patient, the body position cannot be modified unless the instruments are disengaged and removed from the body cavity. When the robotic system is in place, access to the patient in case of emergency is limited. Therefore, the surgical team should be capable of rapidly removing the robot if required.

MANAGEMENT OF ONE-LUNG VENTILATION IN RTE

The management of one-lung ventilation (OLV) remains challenging. Common problems include hypoxemia, failure to isolate the lungs properly and the potential for causing acute lung injury. To install OLV, a left-sided double-lumen tube (DLT) is used. Positioning of the DLT is most reliably achieved with a fiberoptic bronchoscope. It has been shown that left DLTs, when positioned only by inspection and auscultation, were in fact malpositioned in more than 33% of the cases.[28] After positioning the patient from supine to lateral, the position of the DLT is checked again routinely. Cuff pressure is measured to prevent high intracuff pressures and possible mucosal damage. Patients with a difficult airway present an extra challenge. Airways that are difficult for placement of a single-lumen tube (SLT) are even more difficult for placing a DLT because of its size and shape. Oral fiberoptic intubation with a DLT has been described in both awake and anesthetized patients.[29] Alternatives include the use of a bronchial blocker as well as the use of a tube exchanger. The latter may be used for inserting a DLT or changing a SLT for a DLT. As mentioned earlier, the development of hypoxemia is a problem. During OLV both lungs are perfused. Perfusion of the nonventilated lung inevitably leads to transpulmonary shunting, impairment of oxygenation and possible hypoxemia. Another important problem is the risk of acute lung injury, caused by volume or pressure induced stress of the ventilated lung. To decrease the incidence of hypoxemia and the risk of acute lung injury, a good ventilation strategy is important. In our institution, during OLV a protective lung ventilation (PLV) protocol is applied. This consists of a pressure-controlled ventilation strategy with a maximum pressure of 20 cm H2O. Tidal volume is reduced to 6 ml/kg predicted body weight. Furthermore, 5 cm H2O PEEP is routinely used. Although hypoxemia is a constant threat, the lowest possible fraction of inspired oxygen (FiO2) is delivered to prevent oxidative damage and postoperative ALI.[30]

In case of hypoxemia, the first treatment is an increase in FiO2. If no improvement occurs, the surgeon is informed and the nonventilated lung is expanded with 100% oxygen. Our clinical experience suggests that dislocation of the DLT, atelectasis or bronchial occlusion of the ventilated lung with blood or secretions are the most occurring causes of hypoxemia. Therefore, immediate fiberoptic bronchoscopy is performed to rule out or even correct dislocation of the DLT and occluded bronchi. Once these are ruled out, a recruitment maneuver is performed to open possible atelectasis.

When hypoxemia persists, the administration of oxygen with or without CPAP to the nonventilated lung is a valuable option. Clear communication with the surgeon is necessary in these circumstances as both manoeuvres may have a negative impact on the surgical exposure during thorascopy. When applying CPAP, the nonventilated lung is first reinflated as CPAP alone does not inflate an atelectatic lung. At the end of the thoracoscopic phase, the nonventilated lung is reinflated under direct vision and extensive recruitment manoeuvres are performed after which two-lung ventilation is restarted and 10 cm H2O PEEP is added. There is no more need for lung separation during the rest of the operation

and usually the DLT is exchanged for a SLT. The risks and benefits of changing the DLT should be carefully considered. After large fluids shifts and an extended surgical procedure, swelling in the upper airways occurs relatively often. Exchanging the DLT for an SLT should be done under direct vision if possible. If adequate exposure is not possible, an airway exchanger may be used.

FLUID MANAGEMENT

Much has been written about intraoperative fluid administration. Several publications suggest that a restrictive fluid management reduces length of hospital stay, cost and complication rates. In our institution fluid strategy during RTE is aimed at a mildly positive fluid balance of approximately 500–1000ml at the end of the procedure. Additional information is obtained from the use of FloTrac/Vigileo which calculates continuous cardiac output and stroke volume variation from arterial pressure waveform characteristics. These values can be used to predict the effect of extra boluses of fluid. Although not validated for thoracoscopic procedures, it has been validated for laparoscopic operations in pigs.[31] Therefore, FloTrac/Vigileo can be of assistance in the laparoscopic phase of the operation especially to predict fluid responsiveness and distinguish those patients with low cardiac output from hypovolemia and the patients that are in need of inotropic support. The use of central venous oxygen saturation may have additional value in particular in patients with decreased cardiac function. However, at the moment no large scale randomized trials are available.

PERIOPERATIVE COMPLICATIONS

The most common complications encountered perioperatively include arrhythmias, most often seen as the result of manipulation of the heart during the thoracoscopic phase of the operation. Usually these arrhythmias are self-limited after interruption of the surgical manipulation. Another complication regularly seen is the development of a pneumomedastinum as a result of the opening of the hiatus during the laparoscopic phase of the operation. Hemodynamics may show the characteristics of a tension pneumothorax. Again the surgeon should be informed immediately and asked to lower the pressure of the pneumoperitoneum. If indicated, thoracic drains are inserted to relieve the pneumomediastinum.

POSTOPERATIVE CARE

Postoperatively all patients remain under general anaesthesia and are intubated until they are transferred to the intensive care unit. Extubation is aimed for the same day. Although immediate extubation in the operating room has been described and considered safe[32], we consider it appropriate to ventilate patients postoperatively until chest X-ray is obtained. When the X-ray shows no significant atelectasis, weaning from ventilation is started. As stated earlier, thoracic epidural analgesia improves pain control and decreases pulmonary

complications. In order to ensure analgesia is satisfactory enough to enable mobilization and physiotherapy, each patient is visited on a daily basis by a pain service as long as the epidural catheter remains in place.

PROCEDURES

ROBOT-ASSISTED THORACOSCOPIC DISSECTION

Instruments

- Hook
- Cadiere
- Needle driver
- Long tip forceps
- optional: Large Hem-o-lok® Clip Applier

Positioning

The patient is positioned in the left lateral decubitus position, tilted 45° towards the prone position. The operating table is flexed, lowering the legs and upper thorax (the patient is positioned with the xyphoid above the pivoting point of the table). This extends the thorax and widens intercostal space for introducing trocars. The bedside cart of the robotic system (DaVinci Si system, Intuitive Surgical Inc., Sunnyvale CA, USA) is brought into the operative field from the dorsocranial side of the patient (figure 1). Before incision, the right lung is desufflated. A 10-mm camera port is placed at the sixth intercostal space, posterior to the posterior axillary line. Two 8-mm ports are placed just anterior to the scapular rim in the fourth intercostal space and more posterior in the eighth intercostal space.

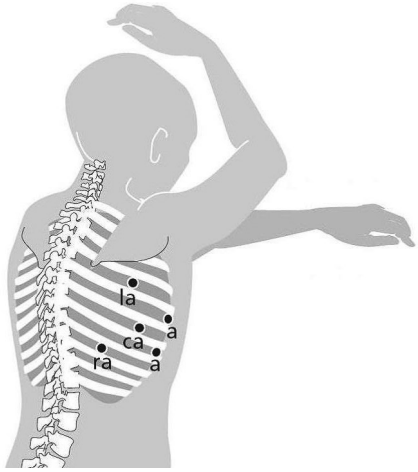
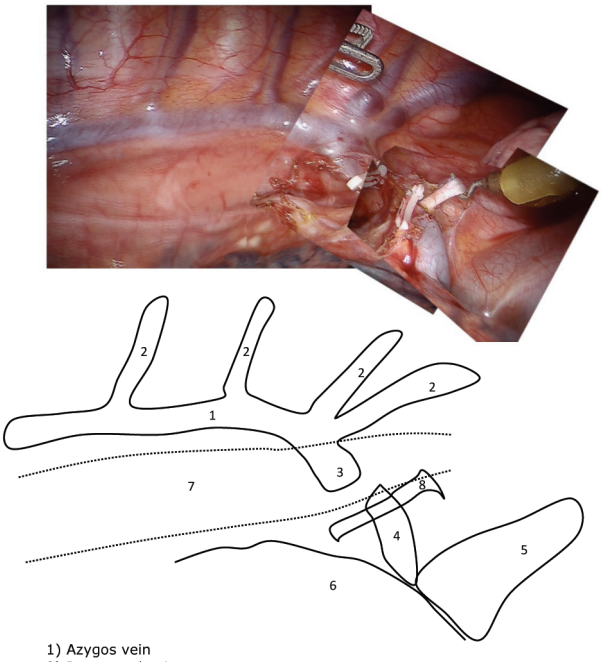


Figure 1 Thoracoscopic phase of robot-assisted thoracolaparoscopic esophagectomy
Legend: An overview showing the setup of the operating room with the robot, the patient in the left lateral decubitus position (45˚ tilted towards prone) and port placement. ra - right robotic arm; la - left robotic arm; ca - camera arm; a - assisting port. (from Boone *et al.*[9], with permission)



1) Azygos vein
2) Intercostal veins
3) Azygos arch (proximal lump with hem-o-lok, after transection)
4) Azygos arch (distal lump with hem-o-lok, after transection)
5) Superior caval vein
6) Right lung (desufflated)
7) Esophageal tract
8) Right vagal nerve

Figure 2 Thoracoscopic view of the upper mediastinum showing the pleural dissection anteriorly of the esophagus towards the azygos vein which is dissected at the level of the azygos arch.

Two thoracoscopic ports are used in the fifth and seventh intercostal spaces just posterior to the posterior axillary line. These ports are used for conventional thoracoscopic assistance such as suction, traction, and clipping. (figure 1). CO_2 insufflation of the thoracic cavity permits excellent vision, without the need for retracting the lung from the operative field. In case of a none-compliant lung, a retractor can be used.

Operative Steps

After division of any pulmonary adhesions and a proper overview of the operating field is achieved, the pulmonary ligament is divided. The parietal pleura is dissected at the anterior side of the esophagus from the diaphragm up to the azygos arch (figure 2). The azygos arch is carefully dissected and ligated using Hem-o-lok® clips (size Large, Teleflex Medical, Limerick PA, USA) applied with the robot. Then dissection of the parietal pleura is continued above the arch for a right paratracheal lymph node dissection. The right vagal nerve is dissected below the level of the carina. Subsequently, the parietal pleura is dissected at the posterior side of the esophagus cranially to caudally along the azygos vein, including the thoracic duct. At the level of the diaphragm, the thoracic duct is clipped

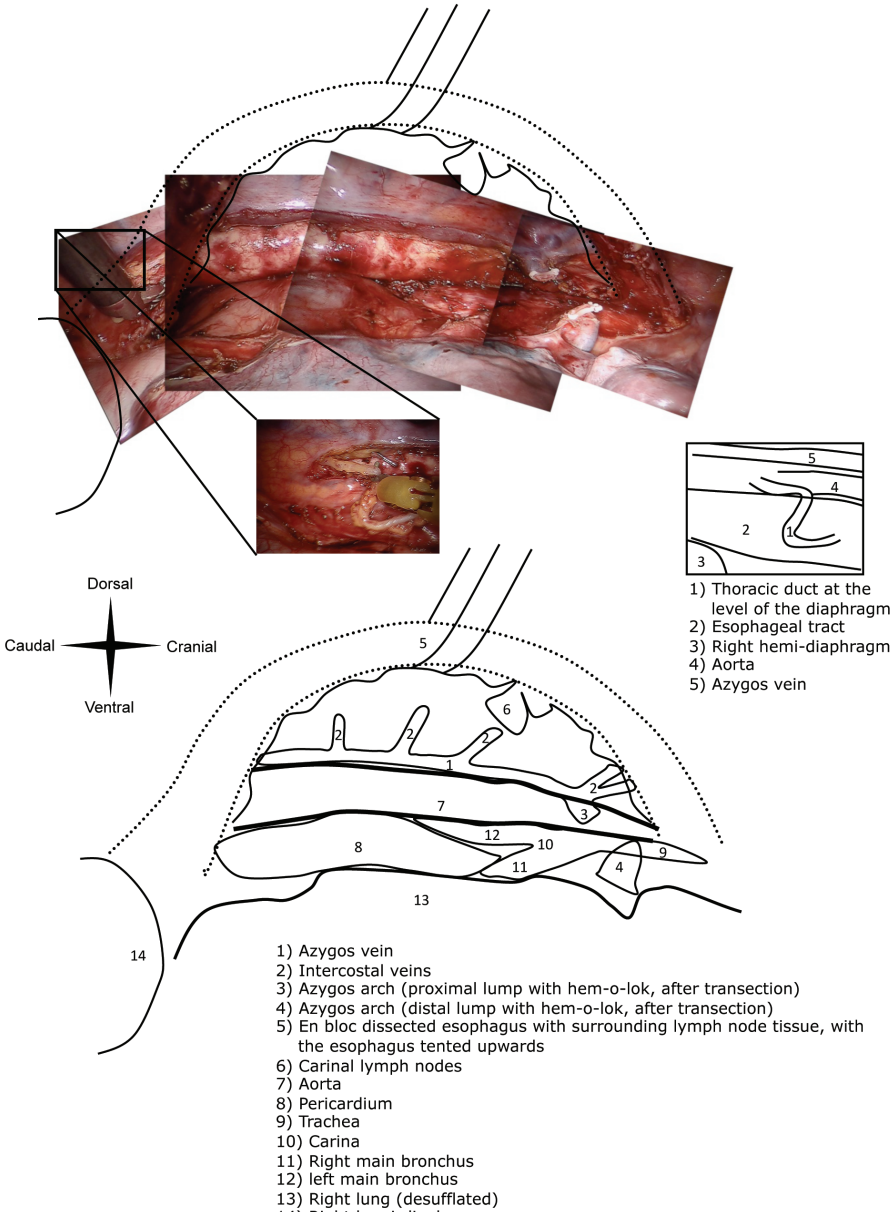


Figure 3 Thoracoscopic view of the mediastinum showing the esophagus (tented upwards) with surrounding lymph nodes, the azygos vein, the aorta, pericardium, the trachea and main bronchi and the thoracic duct which is clipped and dissected at the level of the right diaphragm.

with a 10-mm endoscopic clipping device (EndoclipTM II; Covidien, Mansfield, Massachusetts, USA) to prevent postoperative chylous leakage (figure 3). At the level of the diaphragm, a Penrose drain is placed around the esophagus to provide traction, which facilitates esophageal mobilization. The esophagus is then resected en bloc with the surrounding mediastinal lymph nodes and the thoracic duct from the diaphragm up to the thoracic inlet. Aortoesophageal vessels are identified and clipped by the assisting surgeon. The extensive lymphadenectomy includes the right-sided paratracheal (lymph node station 2R), tracheobronchial (lymph node station 4), aortopulmonary window (station 5), carinal (station 7) and peri-esophageal (station 8) lymph nodes. A 24-Fr chest tube is placed, and the lung is insufflated under direct vision.

LAPAROSCOPIC DISSECTION

Instruments

- Harmonic scalpel
- 2x fenestrated bowel clamps
- Endopaddle
- Clipper

Positioning

After completion of the robot-assisted thoracoscopic esophageal mobilization, the patient is put in supine position. An 11-mm camera port is introduced left paraumbilically, and an 11-mm working port is placed at the left midclavicular line at the umbilical level. A 5-mm working port is placed more cranially at the right midclavicular line. A 5-mm assisting port is placed in the left subcostal area, and a 12-mm port is placed pararectally right for the liver retractor (figure 4). The abdomen is insufflated to a carbon dioxide pressure level of 15 mmHg.

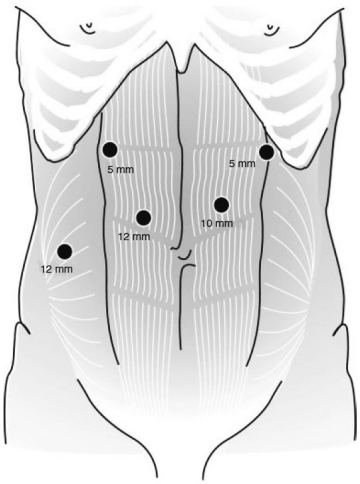
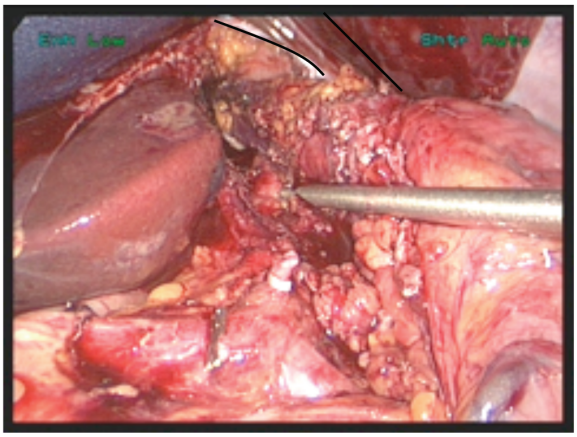


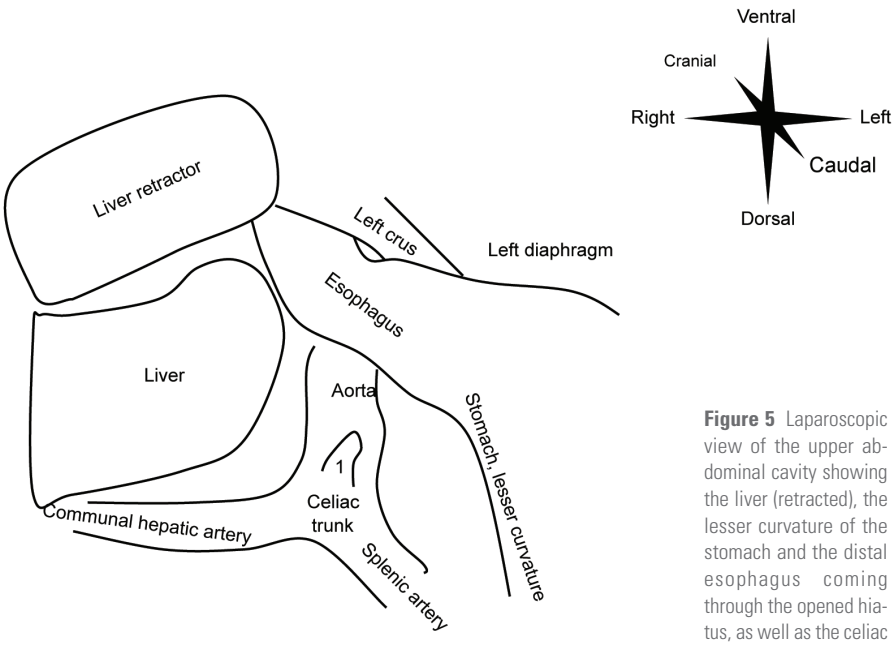
Figure 4 Laparoscopic phase of robot-assisted thoracolaparoscopic esophagectomy; an overview showing the setup of the operating room, the patient in supine position and port placement.

Operative Steps

The hepatogastric ligament is opened. The greater and lesser curvatures are dissected with ultrasonic harmonic scalpel. The hiatus is opened, and the distal esophagus is dissected from the right and left crus. The carbon dioxide pressure level is reduced to 6 mmHg to avoid excessive intrathoracic pressure and a chest tube is placed in the left pleural sinus. Dissection

and lymphadenectomy then continues around the celiac trunk (figure 5). The left gastric artery and vein then are transected at their origin. Abdominal lymphadenectomy includes lymph nodes surrounding the left gastric artery and the lesser omental lymph nodes.
The cervical esophagus is mobilized through a left-side longitudinal neck incision along the sternocleidoid muscle. No formal cervical lymph node dissection is carried out, but cervical lymph nodes are dissected if lymph node metastases are suspected macroscopically during the cervical phase of esophagectomy. The esophagus is dissected and a cord is attached to the proximal part of the specimen to enable pull-up of the gastric conduit along the anatomical tract of the esophagus.





1) Left gastric artery, proximal lump with hem-o-lok (after transection)

Figure 5 Laparoscopic view of the upper abdominal cavity showing the liver (retracted), the lesser curvature of the stomach and the distal esophagus coming through the opened hiatus, as well as the celiac trunk with the transacted left gastric artery.

The esophagus and surrounding lymph nodes are pulled into the abdomen under laparoscopic vision. A 7-cm transverse incision is made at the level of the left paraumbilical port for extraction of the specimen and stomach using a wound protector. Outside the abdomen, a 5-cm-wide gastric tube is constructed with staplers (GIATM 80, 3·8 mm; Covidien, Dublin, Ireland), and the stapled line is oversewn with 3–0 polydioxanone. Routine extracorporal oversewing was reintroduced as two serious complications occurred when the staple line was not oversewn.[20,33] The specimen consisting of the esophagus and cardia of the stomach is sent for pathological examination. After the gastric tube has been pulled to the neck, a hand-sewn end-to-side esophagogastrostomy is performed in the neck using using 3–0 polydioxanone single-layer running sutures. Excess gastric tubing is removed using a GIA stapler. A feeding jejunostomy is placed at the level of the transverse incision (Freka® FCJ-Set, Fresenius Kabi AG, Bad Homburg vd H., Germany).

POSTOPERATIVE CARE

CLINICAL CARE

Postoperatively, patients are transferred to the intensive care unit (ICU). After leaving the operating room, mechanical ventilation is continued briefly usually extubating later that evening. After 1 day in the ICU patients are transferred to a medium care (MC) ward.
Important for postoperative care are a nasogastric tube, feeding jejunostomy and an epidural catheter. The nasogastric tube is used for gastric decompression and to provide a splinting in case of anastomotic dehiscence. Fixation of the tube is imperative, as re-introduction can cause damage to the anastomosis. No oral intake is allowed for 7 days minimum. During that first week, feeding is provided by the feeding jejunostomy. After 7 days without any indication of anastomotic dehiscence sips of water are initiated. If there is no evidence of anastomotic leak, oral intake is gradually supplemented to solid foods under close supervision of a clinical nutritionist. The feeding jejunostomy is left *in situ* up to 6 weeks after discharge from the hospital. Only after sufficient intake is maintained, the jejunostomy is removed at the outpatient clinic. Pain medication through the epidural catheter is required to improve postoperative ventilation and coughing. Other strategies to prevent postoperative pulmonary complications include elevation of the bed by 15–30 degrees, physical respiratory therapy and early mobilization.

POINTS OF INTEREST

ANASTOMOTIC LEAKAGE

Leakage of the esophago-gastrostomy can present itself in various ways. Possible clinical signs are fever, swelling, erythema or fluctuations in the neck, subcutaneous emphysema

and pneumothorax. In case of anastomotic leakage, the neck wound is re-opened to enable drainage. Frequent cleaning and flushing of the wound is required. Leakage can occasionally drain to the mediastinum. This causes fever and mostly pleural effusion and atelectasis. The pleural cavity should be drained. The mediastinum is drained through the neck.

CHYLOUS LEAKAGE

The diagnosis of chylous leakage is based on excessive drainage of milk-like fluid from a thoracic drain containing an elevated level of triglycerides and increased concentration of chylomicrons. Long chain triglycerides (LCT) are drained through the thoracic duct. By eliminating LCTs from the diet less triglycerides will be drained, reducing chylous production. To sustain energy intake, medium chain triglycerides which are absorbed directly through the portal system, can be added to the patient's diet.

When a clinical suspicion of chylous leakage arises, concentrations of triglycerides and cholesterol in both serum and drain fluids need to be examined. A triglyceride drainfluid-serum ratio of more than 10 and a cholesterol drainfluid-serum ratio of less than 1 is typical for chylous leakage.

Mild chylous leakage (<500cc/24h) is conservatively treated with adapted enteral feeding (MCT) and ceasing oral intake. Mild to serious leakage (500–1000cc/24h) requires MCT feeding or total parenteral feeding (TPF). Serious leakage of more than 1000cc/24h is treated with TPF without any enteral or oral type of feeding.

OUTPATIENT CARE

After discharge from hospital, patients are seen frequently every 2–4 weeks to make sure their weight is stable and eating is tolerated. In case of weight loss of more than 2 kilograms, tube feeding is started through the jejunostomy. After the initial visits, patients are seen every 3–4 months in the first year, at 6-month intervals in the second year, and annually thereafter. At each visit, a medical interview and physical examination are carried out. Diagnostic modalities such as gastroscopy with biopsy, CT, FDG-PET or magnetic resonance imaging are only performed if tumor recurrence is suspected, in accordance with the 2006 National Comprehensive Cancer Network Esophageal Cancer Clinical Practice Guidelines.[34]

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(14)2137-2150.
2. Stoner GD, Gupta A. Etiology and chemoprevention of esophageal squamous cell carcinoma. Carcinogenesis. 2001;22(11)1737-1746.
3. Marsman WA, Tytgat GN, ten Kate FJ, van Lanschot JJ. Differences and similarities of adenocarcinomas of the esophagus and esophagogastric junction. J Surg Oncol. 2005;92(3)160-168.
4. Holmes RS, Vaughan TL. Epidemiology and pathogenesis of esophageal cancer. Semin Radiat Oncol. 2007;17(1)2-9.
5. Mariette C, Piessen G, Triboulet JP. Therapeutic strategies in oesophageal carcinoma: role of surgery and other modalities. Lancet Oncol. 2007;8(6)545-553.
6. Hulscher JB, Tijssen JG, Obertop H, van Lanschot JJ. Transthoracic versus transhiatal resection for carcinoma of the esophagus: a meta-analysis. Ann Thorac Surg. 2001;72(1)306-313.
7. Orringer MB, Marshall B, Chang AC, Lee J, Pickens A, Lau CL. Two thousand transhiatal esophagectomies: changing trends, lessons learned. Ann Surg. 2007;246(3)363-372.
8. Espat NJ, Jacobsen G, Horgan S, Donahue P. Minimally invasive treatment of esophageal cancer: laparoscopic staging to robotic esophagectomy. Cancer J. 2005;11(1)10-17.
9. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96(8)878-886.
10. Omloo JM, Lagarde SM, Hulscher JB, Reitsma JB, Fockens P, van Dekken H, ten Kate FJ, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the mid/distal esophagus: five-year survival of a randomized clinical trial. Ann Surg. 2007;246(6)992-1000.
11. Rizzetto C, DeMeester SR, Hagen JA, Peyre CG, Lipham JC, DeMeester TR. En bloc esophagectomy reduces local recurrence and improves survival compared with transhiatal resection after neoadjuvant therapy for esophageal adenocarcinoma. J Thorac Cardiovasc Surg. 2008;135(6)1228-1236.
12. Stark SP, Romberg MS, Pierce GE, Hermreck AS, Jewell WR, Moran JF, Cherian G, Delcore R, Thomas JH. Transhiatal versus transthoracic esophagectomy for adenocarcinoma of the distal esophagus and cardia. Am J Surg. 1996;172(5)478-481.
13. Rentz J, Bull D, Harpole D, Bailey S, Neumayer L, Pappas T, Krasnicka B, Henderson W, Daley J, Khuri S. Transthoracic versus transhiatal esophagectomy: a prospective study of 945 patients. J Thorac Cardiovasc Surg. 2003;125(5)1114-1120.
14. Colvin H, Dunning J, Khan OA. Transthoracic versus transhiatal esophagectomy for distal esophageal cancer: which is superior? Interact Cardiovasc Thorac Surg. 2011;12(2)265-269.
15. Luketich JD, velo-Rivera M, Buenaventura PO, Christie NA, McCaughan JS, Litle VR, Schauer PR, Close JM, Fernando HC. Minimally invasive esophagectomy: outcomes in 222 patients. Ann Surg. 2003;238(4)486-494.
16. Gemmill EH, McCulloch P. Systematic review of minimally invasive resection for gastro-oesophageal cancer. Br J Surg. 2007;94(12)1461-1467.
17. Verhage RJ, Hazebroek EJ, Boone J, van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64(2)135-146.
18. Ruurda JP, van Vroonhoven TJ, Broeders IA. Robot-assisted surgical systems: a new era in laparoscopic surgery. Ann R Coll Surg Engl. 2002;84(4)223-226.
19. Camarillo DB, Krummel TM, Salisbury JK, Jr. Robotic technology in surgery: past, present, and future. Am J Surg. 2004;188(4A Suppl)2S-15S.

20. van Hillegersberg R, Boone J, Draaisma WA, Broeders IA, Giezeman MJ, Borel Rinkes IH. First experience with robot-assisted thoracoscopic esophagolymphadenectomy for esophageal cancer. Surg Endosc. 2006;20(9)1435-1439.

21. Kernstine KH, DeArmond DT, Karimi M, Van Natta TL, Campos JH, Yoder MR, Everett JE. The robotic, 2-stage, 3-field esophagolymphadenectomy. J Thorac Cardiovasc Surg. 2004;127(6)1847-1849.

22. Wittekind C, Greene FL, Hutter RVP, Klimpfinger M, Sobin LH. TMM Atlas. Illustrated Guide to the TNM/pTNM Classification of Malignant Tumors. 2004.

23. Cense HA, Lagarde SM, de JK, Omloo JM, Busch OR, Henny C, van Lanschot JJ. Association of no epidural analgesia with postoperative morbidity and mortality after transthoracic esophageal cancer resection. J Am Coll Surg. 2006;202(3)395-400.

24. Block BM, Liu SS, Rowlingson AJ, Cowan AR, Cowan JA, Wu CL. Efficacy of postoperative epidural analgesia: a meta-analysis. JAMA. 2003;290(18)2455-2463.

25. Michelet P, Roch A, D'Journo XB, Blayac D, Barrau K, Papazian L, Thomas P, Auffray JP. Effect of thoracic epidural analgesia on gastric blood flow after oesophagectomy. Acta Anaesthesiol Scand. 2007;51(5)587-594.

26. American Society of Anesthesiologists. Standards for basic anesthetic monitoring. http://www asahq org/For-Healthcare-Professionals/Standards-Guidelines-and-Statements aspx. 2011.

27. Sato N, Koeda K, Ikeda K, Kimura Y, Aoki K, Iwaya T, Akiyama Y, Ishida K, Saito K, Endo S. Randomized study of the benefits of preoperative corticosteroid administration on the postoperative morbidity and cytokine response in patients undergoing surgery for esophageal cancer. Ann Surg. 2002;236(2)184-190.

28. Pennefather SH, Russell GN. Placement of double lumen tubes: time to shed light on an old problem. Br J Anaesth. 2000;84(3)308-310.

29. Patane PS, Sell BA, Mahla ME. Awake fiberoptic endobronchial intubation. J Cardiothorac Anesth. 1990;4(2)229-231.

30. Gothard J. Lung injury after thoracic surgery and one-lung ventilation. Curr Opin Anaesthesiol. 2006;19(1)5-10.

31. Renner J, Gruenewald M, Quaden R, Hanss R, Meybohm P, Steinfath M, Scholz J, Bein B. Influence of increased intra-abdominal pressure on fluid responsiveness predicted by pulse pressure variation and stroke volume variation in a porcine model. Crit Care Med. 2009;37(2)650-658.

32. Chandrashekar MV, Irving M, Wayman J, Raimes SA, Linsley A. Immediate extubation and epidural analgesia allow safe management in a high-dependency unit after two-stage oesophagectomy. Results of eight years of experience in a specialized upper gastrointestinal unit in a district general hospital. Br J Anaesth. 2003;90(4)474-479.

33. Boone J, Borel Rinkes IH, van Hillegersberg R. Gastric conduit staple line after esophagectomy: to oversew or not? J Thorac Cardiovasc Surg. 2006;132(6)1491-1492.

34. Ajani J, Bekaii-Saab T, D'Amico TA, Fuchs C, Gibson MK, Goldberg M, Hayman JA, Ilson DH, Javle M, Kelley S, Kurtz RC, Locker GY, Meropol NJ, Minsky BD, Orringer MB, Osarogiagbon RU, Posey JA, Roth J, Sasson AR, Swisher SG, Wood DE, Yen Y. Esophageal Cancer Clinical Practice Guidelines. J Natl Compr Canc Netw. 2006;4(4)328-347.

D

Ventilation strategies during robot-assisted thoracoscopic esophagectomy

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Roy JJ Verhage
Judith Boone
Richard van Hillegersberg

DEAR EDITOR

With great interest we read the article by Choi *et al.*, reporting the effects of volume-controlled (VCV) and pressure-controlled ventilation (PCV) on oxygenation and intrapulmonary shunting during one lung ventilation (OLV) in robot-assisted thoracoscopic esophagectomy (RTE) in prone position.[1] Since the results of their randomized clinical study indicate that PVC provides no advantage over VCV, the authors conclude that both ventilation techniques can safely be applied during RTE in prone position. Although this is an interesting study providing new insights into this specific field of anaesthesiology, some remarks have to be made.

The study population was randomly divided into two groups, yet, the authors omitted an overview of the baseline characteristics of both study groups. This raises the question whether there were statistical significant differences between both groups. Secondly, the authors failed to describe intra-operative data that may have influenced the endpoints of this study, such as total surgery time and blood loss.

In addition, the primary outcomes were limited to haemodynamic and respiratory variables. It would have been of greater clinical interest if the authors analysed the effect of the different ventilation strategies on post-operative outcome, such as intensive care unit stay and postoperative pulmonary morbidity (e.g. pneumonia, atelectasis and Adult Respiratory Distress Syndrome).

In 2005, we have extensively described our surgical technique of RTE in the left-lateral decubitus position[2]. Our short- and mid-term results of 47 esophageal cancer patients having undergone RTE have shown this technique to be very promising.[3] Regrettably, Choi *et al*. have not described the surgical technique of RTE in prone position in their article. We look forward to a new publication of this research group on this topic.

REFERENCES

1. Choi YS, Shim JK, Na S, Hong SB, Hong YW, Oh YJ. Pressure-controlled versus volume-controlled ventilation during one-lung ventilation in the prone position for robot-assisted esophagectomy. Surg Endosc. 2009;23(10):2286-91.
2. van Hillegersberg R, Boone J, Draaisma WA, Broeders IA, Giezeman MJ, Borel Rinkes IH. First experience with robot-assisted thoracoscopic esophagolymphadenectomy for esophageal cancer. Surg Endosc. 2006;20(9):1435-9.
3. Boone J, Schipper MEI, Moojen WA, Borel Rinkes IHM, Cromheecke GJE, van Hillegersberg R. Robot-assisted thoracoscopic esophagectomy for esophageal cancer: short- and mid-term results. Br J Surg. 2009;96(8):878-86.

E

Incarcerated hiatal hernia after robot-assisted esophagectomy; transhiatal versus thoracoscopic approach

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Judith Boone
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DEAR EDITOR

With interest we read the article by Sutherland *et al.*, in which they reported their initial experience with transhiatal robot-assisted total esophagectomy in 36 patients with esophageal cancer.(1) Seven (19.4%) patients had postoperative incarcerated hiatal hernias, of which 1 died due to complications of the hernia repair. According to the authors, due to performing the mediastinal dissection through laparoscopy, the hiatus is extensively dilated by the angles of the robotic arms, thereby increasing the risk of postoperative hiatal hernia. They therefore recommend to perform a primary closure and reinforcement with mesh sutured to the gastric wall.

In 2009 we have published the world's largest series of robot-assisted thoracolaparoscopic esophagectomy (RTE).(2) At this moment, we have performed this procedure in more than 140 patients. Minimally invasive esophagectomy substantially reduces blood loss and postoperative hospital and intensive care stay compared to open esophagectomy.(3)

We adhere to the combined thoracic and abdominal approach to provide optimal oncological clearance. The largest study to date comparing transhiatal with transthoracic surgery showed an important trend towards improved survival after transthoracic esophagectomy.(4) The survival benefit is attributed to a more extensive lymphadenectomy in the mediastinum. Furthermore, studies have shown that distal esophageal carcinomas frequently metastasize to lymph nodes in the upper mediastinum.(2, 5)

In our series we did not experience any postoperative hiatal hernias so far. In our opinion, this is the result of performing the mediastinal dissection of the esophagus and surrounding lymph nodes through thoracoscopy instead of through the diaphragm. During the laparoscopic phase of RTE, the stomach is mobilized, regional abdominal lymph nodes are dissected and the esophagus is pulled out of the mediastinum. By performing the mediastinal dissection thoracoscopically, the integrity of the hiatus is preserved and no incarcerated hiatal hernias will be encountered postoperatively. In this way, the abovementioned recommendation of the authors can be abandoned. We would encourage Sutherland *et al.* to continue performing robot-assisted esophagectomies and to experience the many advantages of the robot-assisted thoracoscopic approach.

REFERENCES

1. Sutherland J, Banerji N, Morphew J, Johnson E, Dunn D. Postoperative incidence of incarcerated hiatal hernia and its prevention after robotic transhiatal esophagectomy. Surg Endosc. 2011;25:1526-1530.
2. Boone J, Schipper ME, Moojen WA, Borel Rinkes IH, Cromheecke GJ, van Hillegersberg R. Robot-assisted thoracoscopic oesophagectomy for cancer. Br J Surg. 2009;96:878-886
3. Verhage RJ, Hazebroek EJ, Boone J, van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. Minerva Chir. 2009;64:135-146.
4. Omloo JM, Lagarde SM, Hulscher JB, Reitsma JB, Fockens P, van Dekken H, ten Kate FJ, Obertop H, Tilanus HW, van Lanschot JJ. Extended transthoracic resection compared with limited transhiatal resection for adenocarcinoma of the mid/distal esophagus: five-year survival of a randomized clinical trial. Ann Surg. 2007;246:992-1000.
5. Schroder W, Monig SP, Baldus SE, Gutschow C, Schneider PM, Holscher AH. Frequency of nodal metastases to the upper mediastinum in Barrett's cancer. Ann Surg Oncol. 2002;9:807-811.

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LIST OF PUBLICATIONS

Prins MJ, **Verhage RJ**, ten Kate FJ, van Hillegersberg R. Cyclooxygenase isoenzyme-2 and vascular endothelial growth factor expression are associated with poor prognosis in esophageal adenocarcinoma. *Journal of Gastrointestinal Surgery. 2012 Jan 19. [Epub ahead of print]*

Verhage RJ, van der Horst S, van der Sluis P, Lolkema MP, van Hillegersberg R . Risk of thromboembolic events after perioperative chemotherapy versus surgery alone for esophageal adenocarcinoma. *Annals of Surgical Oncology. 2012;19(2):684-92*.

Boone J, **Verhage RJ**, van der Sluis PC, van Hillegersberg R. RE: Incarcerated hiatal hernia after robot-assisted esophagectomy: transhiatal versus thoracoscopic approach. *Surgical Endoscopy. 2012;26(3):886-7.*

Komanduri S, Deprez PH, Atasoy A, Hofmann G, Pokieser P, Ba-Ssalamah A, Collard JM, Wijnhoven BP, **Verhage RJ**, Brücher B, Schuhmacher C, Feith M, Stein H. Barrett's esophagus: treatments of adenocarcinomas I. *Annals of the New York Academy of Sciences*. 2011;1232:248-64.

Twaddell WS, Wu PC, **Verhage RJ**, Feith M, Ilson DH, Schuhmacher CP, Luketich JD, Brücher B, Vallböhmer D, Hofstetter WL, Krasna MJ, Kandioler D, Schneider PM, Wijnhoven BP, Sontag SJ. Barrett's esophagus: treatments of adenocarcinomas II. *Annals of the New York Academy of Sciences*. 2011;1232:265-91.

Verhage RJ, Zandvoort HJ, ten Kate FJ, van Hillegersberg R. How to define a positive circumferential resection margin for T3 adenocarcinoma of esophagus. *American Journal of Surgical Pathology*. 2011;35(6):919-26.

van der Sluis PC, **Verhage RJ**, van Hillegersberg R. Robotgeassisteerde minimaal invasieve thoracoscopische oesofagolymfadenectomie: chirurgische behandeling van het oesofaguscarcinoom. *Nederlands Tijdschrift voor Oncologie*. 2011;8:59-67.

Verhage RJ, Boone J, van Hillegersberg R. RE: Ventilation strategies during robot-assisted thoracoscopic esophagectomy. *Surgical Endoscopy*. 2010;24(3):721-721.

Verhage RJ, Hazebroek EJ, Boone J, van Hillegersberg R. Minimally invasive surgery compared to open procedures in esophagectomy for cancer: a systematic review of the literature. *Minerva Chirurgica*. 2009;64(2):135-46.

Verhage RJ, Kroese AC, van Hillegersberg R. Robot-Assisted Thoracolaparoscopic Esophagectomy. *Thoracic Robotic Surgery: Atlas and Guide*. W. Randolph Chitwood and Kemp Kernstine (Eds). Springer (London, Unitited Kingdom), 2013

van der Sluis PC, **Verhage RJ**, van der Horst S, Lolkema MP, Siersema PD, Braunius WW, van Hillegersberg R. Local gastric conduit resection with free jejunal interposition for postoperative locoregional recurrent esophageal adenocarcinoma at the site of the anastomosis: a case report. *Annals of Thoracic Surgery* (in print)

Verhage RJ, Ruiz AP, Verheem A, Goldschmeding R, Borel Rinkes IH, van Hillegersberg R. Fibrin-thrombin coated sealant increases strength of esophagogastric anastomoses in a rat model. *Journal of Surgical Research* (accepted for publication)

Verhage RJ, Boone J, Rijkers GT, Cromheecke GJ, Kroese AC, Borel Rinkes IH, van Hillegersberg R. Reduced local immune response with CPAP during single lung ventilation for esophagectomy. (submitted)

Verhage RJ, van der Sluis PC, van der Horst S, van der Wal WM, Ruurda JP, van Hillegersberg R. A new clinical scoring system to define pneumonia following esophagectomy for cancer. (submitted)

CURRICULUM VITAE AUCTORIS

Roy Jaap Jan Verhage was born on August 9, 1981 in Leiderdorp, The Netherlands. In 1999, he graduated cum laude from secondary school, the Stedelijk Gymnasium in Leiden. Hereafter he studied Pre-Medicine at University College Utrecht graduating cum laude as a Bachelor of Science in 2002. From 2002 to 2008 he studied Medicine at Utrecht University. During his bachelors and medical studies he was an active member of several student bodies and societies. In 2005 he worked for the Dutch attaché of Healthcare and the attaché of Economics at the Royal Netherlands Embassy in Washington DC, USA, for six months. As a student during the last year of medical school, he conducted research at the department of Traumatology in the University Medical Center Utrecht (UMCU), The Netherlands. In April 2008 he obtained his medical degree after which he started working at the emergency department of the St Jansdal Hospital in Harderwijk. In October 2008 he commenced a PhD program at the department of Surgery in the UMCU focusing on the surgical management of esophageal cancer under the supervision of Prof. Dr. R van Hillegersberg and Prof. Dr. IHM Borel Rinkes. The results of his research are presented in this thesis. During his PhD program he was actively involved in organizing academic events and initiated a postgraduate course on minimally invasive surgery for esophageal cancer. In January 2012 he started his residency in general surgery at the Jeroen Bosch Hospital in 's Hertogenbosch (Dr. K Bosscha). The last two years of his surgical training will be completed at the UMCU (Dr. MR Vriens).

Roy Jaap Jan Verhage werd geboren op 9 augustus 1981 te Leiderdorp. In 1999 behaalde hij cum laude het VWO diploma aan het Stedelijk Gymnasium Leiden. Hierna studeerde hij Pre-Medicine aan het University College Utrecht waar hij in 2002 cum laude zijn Bachelor of Science behaalde. Aansluitend ging hij Geneeskunde studeren aan de Universiteit Utrecht. Tijdens zijn studie werkte hij gedurende 6 maanden op de Koninklijke Nederlandse Ambassade in Washington DC, Verenigde Staten, voor de attachés van Volksgezondheid, Welzijn en Sport en Economische Zaken. Tijdens zijn laatste jaar van de studie Geneeskunde verrichtte hij onderzoek bij de afdeling Traumatologie (Prof. Dr. LPH Leenen) in het Universitair Medisch Centrum Utrecht. In april 2008 behaalde hij zijn artsexamen waarna hij ging werken als arts-assistent niet-in-opleiding op de afdeling Spoedeisende Hulp van het Sint Jansdal Ziekenhuis in Harderwijk (Drs. S Kruisinga). Van oktober 2008 tot oktober 2011 was hij werkzaam als promovendus in het Universitair Medisch Centrum Utrecht bij de afdeling Heelkunde. Onder leiding van Prof. Dr. R van Hillegersberg en Prof. Dr. IHM Borel Rinkes deed hij onderzoek naar de chirurgische behandeling van slokdarmkanker, wat geresulteerd heeft in dit proefschrift. Tijdens zijn promotie onderzoek heeft hij zich tevens ingezet voor o.a. de organisatie het Heelkunde Voorjaarssymposium en de cursus Minimally Invasive Esophageal Cancer Surgery. Vanaf 1 januari 2012 is hij in opleiding tot chirurg. De opleiding start met vier jaar in het Jeroen Bosch Ziekenhuis te 's Hertogenbosch (opleider Dr. K Bosscha) en wordt gevolgd door twee jaar in het Universitair Medisch Centrum Utrecht (opleider Dr. MR Vriens).