UPPSALA
UNIVERSITET

Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine 1756

Colon Cancer, Prognosis After Surgery

What Are the Risks of Recurrent Disease, and How Do We Find Those at Risk?

ERIK OSTERMAN

ACTA
UNIVERSITATIS
UPSALIENSIS
UPPSALA
2021

ISSN 1651-6206
ISBN 978-91-513-1253-8
URN urn:nbn:se:uu:diva-437831

Dissertation presented at Uppsala University to be publicly examined in Rudbecksalen, Rudbecklaboratoriet, Dag Hammarskjölds väg 20, Uppsala, Saturday, 25 September 2021 at 13:00 for the degree of Doctor of Philosophy (Faculty of Medicine). The examination will be conducted in English. Faculty examiner: Professor Ismail Gögenur (Center for Surgical Science, Copenhagen university, Copenhagen).

Abstract

Osterman, E. 2021. Colon Cancer, Prognosis After Surgery. What Are the Risks of Recurrent Disease, and How Do We Find Those at Risk? (Prognos efter kirurgi för koloncancer. Hur stor är recidivrisken och hur hittar vi dem med hög risk). *Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine* 1756. 56 pp. Uppsala: Acta Universitatis Upsaliensis. ISBN 978-91-513-1253-8.

Colon cancer is the fourth most common cancer worldwide with approximately 1.2 million yearly cases. Developments in the standard of care have improved prognosis.

In **Paper I** the recurrence risk was investigated in a national material consisting of 14,325 colon cancer patients. Three fourths of stage II patients have a risk of approximately 11%, indicating that adjuvant chemotherapy can marginally improve prognosis. In stage III, one fifth of the patients have such a low risk of recurrence that the addition of a second chemotherapeutic drug, oxaliplatin with its risk for late toxicity, may be questioned.

In **Paper II** emerging risk factors were investigated in a thoroughly staged and described material of 416 colon cancer patients from one county. All emerging risk factors correlated with an increased recurrence risk. Adjusting for established risk factors, pN-substage and postoperative carcinoembryonic antigen (CEA) correlated independently with recurrence.

Paper III investigated the completeness and correctness of recurrences in the Swedish Colorectal Cancer registry in 2,893 patients from two counties. In patients operated more than 5 years ago 2% of recurrences were not registered.

In **Paper IV** a nomogram for clinicians was developed using registry data to aid the interpretation of the recurrence risk and discussion with patients about treatment choices. It was validated in Norwegian cancer registry data and performed better than other available nomograms.

Future investigations of the cohort from paper II are planned with immunohistochemistry, tumour-normal tissue sequencing and biomarkers.

In summary, recurrence rates have decreased since the early 2000s and a large proportion of patients can probably be spared some or all adjuvant treatment. Established risk factors describe a large part of the risk, but there is room for improvement. The biomarker CEA taken after surgery could aid in the selection of patients to receive adjuvant treatment and guide follow-up. Adding other biomarkers might further improve the prediction of recurrence risk, though larger, prospective patient materials are needed.

Keywords: colon cancer, recurrence risk, prediction model, biomarkers

Erik Osterman, Department of Immunology, Genetics and Pathology, Experimental and Clinical Oncology, Rudbecklaboratoriet, Uppsala University, SE-751 85 Uppsala, Sweden.

ISSN 1651-6206
ISBN 978-91-513-1253-8
URN urn:nbn:se:uu:diva-437831 (http://urn.kb.se/resolve?urn=urn:nbn:se:uu:diva-437831)

Do. Or do not. There is no try.
-Yoda

List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals.

I. **Osterman E**, Glimelius B. (2018) Recurrence Risk After Up-to-Date Colon Cancer Staging, Surgery, and Pathology: Analysis of the Entire Swedish Population. *Diseases of the Colon & Rectum.* 61(9):1016–1025

II. **Osterman E**, Mezheyeuski A, Sjöblom T, Glimelius B. (2020) Beyond the NCCN risk factors in colon cancer – an evaluation in a Swedish population-based cohort. *Annals of Surgical Oncology*. 27(4):1036–45.

III. **Osterman E**, Hammarström K, Imam I, Osterlund E, Sjöblom T and Glimelius B. (2021) Completeness and accuracy of the registration of synchronous metastasis and recurrences in the Swedish Colorectal Cancer Registry (SCRCR) and an update of recurrence risk in colon cancer. *Acta Oncologica. 60(7):842-9.*

IV. **Osterman E**, Ekström J, Sjöblom T, Kørner H, Mycklebust T Å, Grønlie Guren M, Glimelius B. (2021) Accurate population-based model for individual prediction of colon cancer recurrence. *Acta Oncologica.* Epub ahead of print. 10.1080/0284186X.2021.1953138

Other papers by the PhD candidate not included in this thesis:

Mezheyeuski A, Hrynchyk I, Herrera M, Karlberg M, **Osterman E,** Ragnhammar P, Edler D, Portyanko A, Ponten F, Sjöblom T, Glimelius B, Östman A. (2019) Stroma-normalised vessel density predicts benefit from adjuvant fluorouracil-based chemotherapy in patients with stage II/III colon cancer. *British Journal of Cancer.* 121(4):303-311. doi: 10.1038/s41416-019-0519-1

Osterman E, Hammarström K, Imam I, Osterlund E, Sjöblom T, Glimelius B. (2020) Recurrence Risk after Radical Colorectal Cancer Surgery—Less Than before, But How High Is It? *Cancers*. *12*, 3308; doi:10.3390/cancers12113308

Glimelius B and **Osterman E**. (2020) Adjuvant Chemotherapy in Elderly Colorectal Cancer Patients. *Cancers*. *12*, 2289; doi:10.3390/cancers12082289

Contents

Abbreviations

AUC	Area under (Receiver Operating Characteristics) curve
BRAF	Proto-oncogene B-Raf
CAPOX	Chemotherapy regimen with oral fluoropyrimidine and oxaliplatin
CEA	Carcinoembryonic antigen
CI	Confidence interval, 95%
CIN	Chromosome instability
CMS	Consensus molecular subtypes
CRC	Colorectal cancer
CRP	C-reactive protein
ctDNA	Circulating tumour DNA
DFS	Disease-free survival
ESMO	European Society for Medical Oncology
FLOX, FOLFOX	Chemotherapy regimen with 5-fluorouracil, leucovorin, oxaliplatin
FLv	Chemotherapy regimen with 5-fluorouracil, leucovorin
HR	Hazard ratio
KRAS	Proto-oncogene Kirsten-RAS
LN	Lymph node
LNR	Lymph node ratio
MMR	Mismatch repair
MSI	Microsatellite instability
MSKCC	Memorial Sloan Kettering Cancer Centre
NCCN	National Comprehensive Cancer Network
OS	Overall survival
RFS	Recurrence-free survival
RR	Recurrence Risk
SCOC	Special Commission on Cancer
SCRCR	Swedish ColoRectal Cancer Registry
TNM	Tumour-Node-Metastasis
TTR	Time to recurrence
U-CAN	Uppsala-Umeå Comprehensive Cancer Consortium
UICC	Union for International Cancer Control

Introduction

Epidemiology

Colorectal cancer (CRC) is the third most common cancer worldwide with an estimated incidence of 1.9 million cases in 2020.[1] Two thirds of these cancers are in the colon, making colon cancer the fourth most common cancer worldwide. CRC is more common in developed regions of the world where incidence is between 30 and 50 cases per 100,000 persons. In less developed regions the incidence is lower, 10 – 20 per 100,000 persons, with the lowest incidence seen in South Central Asia, 5 per 100,000 persons.[2] The opposite relationship is seen for mortality where the prognosis is better in parts of the world with better access to care. This can in part be explained by the competing risk of dying from other diseases before CRC manifests and by lifestyle (see aetiology).

In Sweden 4,900 colon cancers were diagnosed in 2019, an increase compared with previous years due to both increasing incidence and population growth.[3] The sex distribution is almost equal with a higher age-standardized incidence among men but more cases among women due to their, on average, longer lifespan. The five-year OS after colon cancer diagnosis in Sweden was 40% in the sixties and had increased to above 60% in 2009 and the positive trend is expected to continue.[4,5]

Genomic classification of colon cancer

Cancer is a disease of genomic instability where the accumulation of mutations cause dysregulation and uncontrolled cell growth.[6] There are three main types of genomic instability in colon cancer: chromosomal, microsatellite and methylation of CpG-sites.[7] The most common type is chromosomal instability (CIN) representing 70-85% of colon cancers.[8] CIN tumours are characterised by multiple large chromosomal abnormalities. The causes of CIN are unknown, although the inactivation of the tumour suppressor gene, APC has been proposed as a potential initiator.[9] The majority of colon tumours exhibiting CIN have loss of function mutations in APC, resulting in the development of a precursor adenomatous polyp due to dysregulated Wnt signalling. Subsequent mutations in other pathways (e.g. Ras-MAPK, PI3K-Akt, TGFβ, p53) increase survivability and growth of the

mutated cells until they can invade the underlying tissue, at first manifesting as a late, growing adenoma and then a frank cancer; adenocarcinoma.[10]

Microsatellite instability (MSI) is caused by mutations or hypermethylation of the DNA-mismatch repair genes (MMR), typically *MLH1* or *MSH2*.[7] The MMR system is responsible for the repair of small genomic lesions (mismatches, insertions, and deletions) due to DNA-polymerase mistakes. Cells with deficient MMR accumulate large numbers of mutations in repeating sequences of DNA, known as microsatellites, some being in the same pathways as mentioned for CIN. Tumours with MSI are more common in the right colon and frequently feature *BRAF* mutations (60%).[11] In addition to CIN and MSI, cancers may have abnormal CpG methylation (CIMP-positive) leading to genomic instability from epigenetic modification and down-regulation of tumour suppressors genes.[7,12]

Key obstacles that the cancer needs to overcome were outlined by Hanahan and Weinberg in 2000 and 2011.[6,13] Cancers accomplish this in a multitude of ways, but the challenges are somewhat similar for all cancers. Continuous growth requires sustained proliferative signalling, avoidance of growth suppressor signals and replicative immortality. Blood and nutrient flow decrease as the tumour grows and requires induction of angiogenesis to improve it. The complex process of metastasis in turn requires its separate adaptations to allow for migration in tissue and growth away from the microenvironment of the primary tumour. Angiogenesis and growth factors can be acquired by promoting inflammation, but this carries the risk of destruction unless the cancer can avoid the immune response, another hallmark.[13] As the cells divide they create subpopulations diverging from the original cell with additional mutations, evolving the tumour. Because of this divergence some cancer cell populations may develop resistance to treatment.[14] Differences between the primary tumour and metastases are common, but alterations in targetable pathways are usually concordant.[15–17]

An alternative to the traditional CIN and MSI classification is the four "consensus molecular subtypes" (CMS) for colorectal cancer. However, these are also heterogenous and do not capture the full complexity of the disease. CMS1 contain the MSI and hypermethylated cancers and express both immune system stimulating and immune evasion genes. CMS2-4 are all CIN but differ in gene expression, CMS2 express genes usually associated with the adenoma-carcinoma sequence, CMS3 have a metabolic adaption with frequent *KRAS* mutations and CMS4 cancers are more invasive, overexpressing genes associated with infiltration, angiogenesis, and inflammation. In the clinical setting CMS1 correlates with right-sided, high-grade lesions with a low risk of metastasis but have the worst prognosis if metastasized.[18] CMS2 is usually left-sided as implicated by the CIN and adenoma-carcinoma sequence association, and both CMS2 and CMS3 are neutral in terms of metastatic potential and prognosis. CMS4 is more aggressive, often diagnosed at advanced stages and confer a poor prognosis.[12]

Aetiology

It takes several years, if not decades, from the first predisposing mutation to a clinically relevant colon cancer.[19] A major risk factor of colon cancer is thus age, with an increasing likelihood of colon cancer diagnosis after age 50.[20] The median age at colon cancer diagnosis in Sweden was 74 years between 2007 and 2011.[21] Approximately 5% of colon cancers are part of hereditary syndromes and 15-30% of cancers have a hereditary component. Inherited mutations in MMR genes cause the syndrome hereditary non-polyposis colorectal cancer (HNPCC/Lynch syndrome) representing 2-6% of colon cancer cases.[20] Inherited *APC* mutations are rare (1%), and patients with *APC*-mutations develop hundreds to thousands of colon (and rectal) polyps in a syndrome called "familial adenomatous polyposis" (FAP). Some of these polyps will acquire further mutations and develop into cancer prompting preventive measures such as early screening and early total colectomy.[22] Some other genes linked to hereditary CRC include *POLE*, *MUTYH*, *SMAD4*, *STK11* and *TP53*.[23] Inflammatory bowel disease also increases the risk of colon cancer, the exact reasons are not known but inflammation, genetic, and acquired factors are implicated.[24]

Environmental factors explain much of the difference in colon cancer incidence between parts of the world. Diets rich in fat and meat and lacking in greens and dietary fibre increase the risk of colon cancer. Other risk factors are low physical activity, obesity, cigarette smoking and high alcohol consumption.[20]

Staging

Colon cancer is staged according to the Tumour-Node-Metastasis (TNM) system, either the American Joint Committee on Cancer 8th edition or the Union for International Cancer Control (UICC) 8th edition. There are no major differences in the staging of colon cancer between the systems. The tumour (T) is classified depending on the depth of invasion where T1 tumours grow in the submucosa of the colon, T2 grows into the muscle, T3 grows through the muscle and into the subserosa and T4 tumours grows through the subserosal tissue to the peritoneal surface or on/into other organs. T1 is further divided by how many thirds of the mucosa it invades (sm1-3). Cancerous pedunculated polyps are instead subdivided by how far down the stalk the cancer invades (Haggitt 0-4).[25] T3 is subdivided by the subserosal invasion, <1 mm, 1-5 mm, 6-15 and >15 mm (a, b, c, d, respectively). Growth on the peritoneal surface is classified as T4a and growth on/into other organs is classified as T4b.

Table 1: Stage by TNM

Stage	T	N	M
I	T1 - 2	N0	M0
II	T3 – 4	N0	M0
III	Any T	N1 – 2	M0
IV	Any T	Any N	M1

Node classification (N) depends on tumour spread to lymph nodes (LN) in the mesentery and omentum. To reliably classify the N-stage, it is stated that at least 12 LN need to be investigated from a surgical specimen. However, to reliably stage less advanced cancers a higher number of investigated nodes is needed.[26] N1a means spread to 1 LN, N1b 2-3 LN, N2a 4-6 LN and seven or more LN metastases are classified as N2b. Additionally localized and non-continuous spread of tumour tissue is classified as tumour deposits and categorized as pN1c if LN are not completely taken over by tumour. Lymph node ratio (LNR) is calculated by dividing LN with tumour spread (positive LN) by examined LN, as with the subclassification of N-stage this gives additional information regarding the extent of LN involvement and is prognostic.[27,28] Any distant metastasis results in a positive metastasis classification (M1), this can also be subclassified into M1a-c depending on the extent of metastatic spread. TNM-staging can be performed before surgery and is then denoted cTNM, where c stands for clinical. When staging is performed by a pathologist after surgery it is denoted pTNM and staging of cancers treated with radiation or chemotherapy before surgery is denoted ypTNM.

Once the TNM stage has been determined tumours are divided into stages, see Table 1. In 2020, 19% of patients in Sweden were in stage I at diagnosis, 30% each in stage II and III, and 23% in stage IV.[29] Tumour invasion of vessels, lymphatics (lymphovascular invasion/vascular invasion) and perineural tissue, margins of resection, circumferential resection margin and tumour differentiation, or grade, should also be noted during pathology examination. These factors do not influence the tumour stage but are risk factors of recurrence.

Diagnosis

Symptoms of colon cancer may include iron-deficiency anaemia, change in bowel habits, macroscopic blood in stool, weight loss, and local symptoms from the tumour.[30] However, these symptoms are not specific for colon cancer and some patients have no symptoms at all. A quarter of all patients, and a third of those with metastasis present in the emergency department because of bowel obstruction, perforation, bleeding or other symptoms.[31] Survival is worse after emergency surgery due to postoperative complications, comorbidities and more advanced tumours[32]. In addition, emergency surgery increases the risk of recurrence by delaying the time to adjuvant treatment.[33]

Efforts to facilitate early diagnosis of symptomatic CRC have been implemented in many countries in the form of standardized cancer patient pathways.[34] Patients presenting with certain symptoms are referred to specialist care with the aim to perform all the necessary work-up within a defined number of weeks from the first healthcare contact. The cancer patient pathways decrease wait times, improve early detection, and survival for patients with symptoms.[34,35] Screening of asymptomatic patients aims to decrease the incidence and mortality by removing precursors to cancer (polyps) and facilitate early detection of cancer.[36] Several methods exist, each with benefits and drawbacks. Multi-step programmes involving either repeat faecal testing and follow-up colonoscopy, or immediate colonoscopy every few years is the norm. These programmes decrease the incidence and mortality, are cost effective and are recommended by central health-care agencies in large parts of the world including Sweden.[37]

Colonoscopy with biopsy is the gold standard for diagnosing colon cancer although this is not always possible. In those cases a virtual colonoscopy computed tomography can be used, however this is without the possibility for biopsies.[38] The advantage of the computed tomography scan is that it can cover the most common metastatic sites, the lungs and liver, provides a radiology assessed cTNM stage, and informs about other differential diagnoses. Carcinoembryonic antigen (CEA) is a biomarker of CRC but has a low sensitivity and specificity as a diagnostic tool. Elevated CEA (>5 ng/ml) before surgery indicates worse prognosis according to most publications.[39,40] CEA after surgery can aid in the detection of local and metastatic spread, especially if it was elevated before surgery.[41]

Colorectal Surgical Anatomy

The colon is an important part of the gastrointestinal tract participating in digestion and re-uptake of fluids and electrolytes.[22] It also stores and participates in the elimination of faecal matter. The proximal colon is a continuation of the small bowel with the anatomic division at the ileocolic

valve. The colon is easily identified by its longitudinal muscle fibres, taenia coli, the variable width, haustra, and adipose tissue raindrops, epiploic appendices. From proximal to distal, the colon is divided into cecum, ascending, right/hepatic flexure, transverse, left/splenic flexure, descending and sigmoid, Figure 1.[42] The first three parts and the first two-thirds of the transverse colon are considered parts of the right colon, and derives from the embryological plane of the midgut. The final third of the transverse colon and the last three parts derives from the hindgut and are considered part of the left colon. The ascending and descending colon are retroperitoneal and fixed against the posterior abdominal wall. The cecum, transverse and sigmoid are intraperitoneal and more mobile.

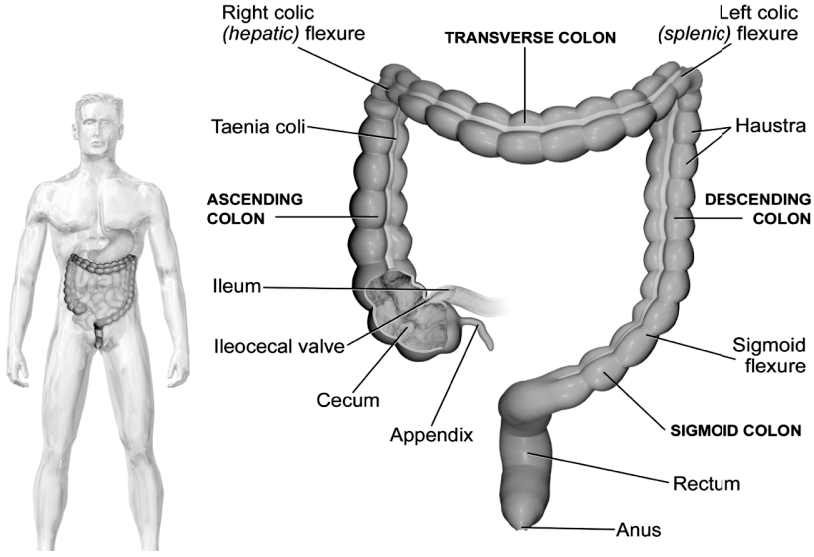

Figure 1: Anatomy of the colon, from wikiversity.com by Blausen.com Staff (2014). CC BY 3.0.

The superior mesenteric artery and its branches supply the right colon and the proximal two-thirds of the transverse colon. The inferior mesenteric artery and branches supplies the distal one-third of the transverse and the left colon. The two arterial systems are often connected, increasing redundancy (by the marginal artery of Drummond and Arc of Riolan). The venous drainage follows the main arteries and ends up in the hepatic portal vein. The lymphatic flow is similarly divided and passes through lymphatic nodes in the mesentery towards nodes closer to the aorta. Distal of the sigmoid colon is the rectum, sometimes defined as the last 12-15 cm of large intestine[43,44] but more recently defined as beginning from the sigmoid take-off.[45] The rectum is often included when discussing cancer of the large intestine, but there are important differences. The biological function is closely related to that of the colon with

emphasis on storage and elimination of faecal matter. It is supplied from the inferior mesenteric artery (superior rectal vessels) and from branches of the interior iliac arteries, the middle rectal and inferior rectal arteries. Because of this two-way vascular supply, the rectum is more resistant to ischemia. The lymphatic drainage follows the vessels but also drains towards iliac and inguinal nodes.[46]

Surgery for colorectal cancer

Early and complete resection of the tumour is the single most important intervention to improve the chances of curing the patient. For small lesions it is sometimes achievable with a local excision, polypectomy. Otherwise, based on the knowledge of vascular and lymphatic drainage, a few standard procedures are used. The right-sided hemicolectomy is used for right-sided cancers and includes resection the proximal transverse colon following the embryological planes and vascular supply. If the distal transverse colon is involved the resection should be extended towards the left colon. There is limited evidence regarding the surgical approach for tumours in the transverse colon.[47] Resection of only the transverse colon is usually not performed due to the embryological and vascular anatomy, and because the ascending and descending colon are retroperitoneal organs and fixed to the posterior wall of the abdominal cavity which makes re-establishing bowel continuity and maintaining circulation more difficult. The left-sided hemicolectomy is performed for left-sided cancers, although a sigmoidectomy can be performed for sigmoid cancers. If there is a hereditary component, multiple tumours, or the patient has had previous surgery for CRC, a complete or partial colectomy can be performed as a preventive measure. The bowel continuity is either restored with an anastomosis or left discontinued and a diverting stoma created. If there is a high risk of anastomotic leakage (e.g. emergency surgery, fragile patient), a temporary loop-diversion can be used to avoid re-operation in case of a leak.[48] Continuity can be restored later, often after adjuvant treatment has been given and the patient has recovered sufficiently to tolerate surgery again.

The concept of complete mesocolic excision was introduced in the early 2000s and came from the concept of total mesorectal excision for rectal cancer.[49] Utilizing the embryological planes of development to perform the surgery allows for a protective sheet of fascia to cover the vessels and lymph nodes in the mesentery, in theory decreasing the risk of spreading the cancer at surgery. Dissection of the mesentery allows for access to the vessels at their branching and allows a high-tie or central vascular ligature. Complete mesocolic excision and central vascular ligature result in higher lymph node yields and potentially better oncological outcomes.[49–51] Laparoscopic colon and rectal surgery have the same long-term oncological results and better

perioperative outcomes compared to open surgery.[52–54] Drawbacks of laparoscopic surgery are longer surgery duration and higher risk of wound infection. Benefits include shorter hospital stay, less blood loss and less postoperative pain.[53] Mortality and comorbidity after surgery have been further reduced by the introduction of evidence based "enhanced recovery after surgery" programmes.[55] Examples of interventions included in these programmes are limited perioperative fluids, early mobilization, early oral nutrition and standardized care guidelines.

Surgery in the emergency setting has a higher rate of complications, mortality, and risk of recurrence.[32,56] It may be performed by non-specialized surgeons, at night and in a critically ill patient where resolving the situation at hand is (often) more important than doing an oncologically safe procedure that takes longer time and might risk the life of the patient. The most common reason for emergency surgery is bowel obstruction, with or without perforation of the bowel and sepsis. The recommended approach is to solve the situation using diverting stomas if an oncologically safe procedure is impossible and leave the tumour to be removed later.[57]

In the metastatic setting surgery and interventional radiology are currently the curative options. Liver and lung metastases can be resected if there is enough healthy organ left and no other signs of metastasis. Methods are available to increase the healthy liver, making more patients eligible for surgery. Radiofrequency ablation or other ablative approaches are interventional options for small metastases and may provide a cure.[58] Patients with metastases to the peritoneum can, if the spread is somewhat limited, be operated and cured. Addition of warm intraperitoneal chemotherapeutic agents further improves the prognosis after surgery for peritoneal metastases.[58,59]

Oncological treatment

Systemic chemotherapeutic drugs seldom target specific cells but exploit that rapidly dividing cells are disproportionally sensitive to disruption. The aim of the treatment is halted growth, apoptosis, and/or necrosis. Fluoropyrimidines are the basis for chemotherapy in colon cancer. The most used drug, a fluoropyrimidine, 5-fluorouracil, was discovered in 1957 and the first published use in colon cancer was in 1962. Fluoropyrimidine drugs inhibit the synthesis of DNA and RNA causing apoptosis and necrosis.[60–62] Initial results were mixed as the optimal dosage and regime were worked out.[63] It was not until it was biochemically modulated, initially with methotrexate and subsequently, with a reduced folate, leucovorin, that it gave clinically meaningful effects. Leucovorin is commonly used as an adjunct to fluoropyrimidine treatment and enhances the cytotoxic effect by stabilizing the enzyme-drug complex.[64] 5-fluorouracil is today administered as an

intravenous rapid injection or, more commonly, as an infusion over about 48 hours. An oral alternative is capecitabine, a pro-drug that is converted to fluoropyrimidine in three steps.[60] Cancer cells have an upregulation of the enzymes responsible for the last step of conversion giving capecitabine some selectivity. It is as effective as intravenous fluoropyrimidine in pooled analyses of several trials.[65]

Oxaliplatin is another main chemotherapeutic agent in colon cancer. It was discovered in the 70s and initially used in metastatic colon cancer.[66,67] Oxaliplatin exerts its effect by reaction with macromolecules in the cell, especially with DNA, forming cross links and inhibiting both replication and transcription.[62] In the MOSAIC trial, published 2004, it was given with 5-fluorouracil and leucovorin as adjuvant treatment for advanced stages of colon cancer, reducing the recurrence rate by approximately 20% compared with 5-fluorouracil and leucovorin.[65,68] Two other studies also found 5-fluorouracil, leucovorin and oxaliplatin more effective than a fluoropyrimidine alone and this is standard therapy for patients with stage III since more than a decade.[38,65,69]

Preoperative treatment

Preoperative (neoadjuvant) therapy is the practice of treating patients before surgery with the aim to treat subclinical disease more effectively than if delivered postoperatively. Preoperative chemotherapy affects the primary tumour and metastases if present, reducing size and potentially increasing resectability. There are theoretical benefits to this practice, which has been adopted for cancer of e.g. the stomach.[70] After promising initial results in colon cancer, with downstaging of the tumour and improved resection margins,[71] a randomized multicentre trial FOxTROT, including slightly over 1,000 patients reported that the risk of recurrence after 2 years was decreased, albeit not statistically significant.[72] Yet another study from Denmark has recently completed patient recruitment. In high-risk rectal cancers neoadjuvant radiation therapy has been the standard of care since the 90s[73] and two trials could in 2020 report better tumour outcomes delivering neo-adjuvant chemotherapy rather than postoperative.[74,75]

Postoperative treatment

In the adjuvant setting the aim of chemotherapy is to treat any micro-metastasis that are left after surgery. The effect of adjuvant treatment in stage III colon cancer is well documented and recommended in patients who are healthy enough to tolerate the side-effects.[69,76] In Sweden, about 20% of stage II patients and 65% of stage III patients are treated with adjuvant chemotherapy.[77] The recommended regimen in most guidelines is a fluoropyrimidine (either intravenous with leucovorin, FLv, or oral) and oxaliplatin (FOLFOX, CAPOX or FLOX) which reduces the risk of recurrence compared with fluoropyrimidines and leucovorin (FULV) by

approximately 20% (3-year disease free survival, DFS, 77% compared with 72%).[38] In the 90s the three-year DFS was 44% in stage III without chemotherapy and treatment with FLv decreased recurrences by 30-35%.[78,79] In stage II the risk of recurrence is lower and the recommended adjuvant treatment is either single agent (FLv) chemotherapy or a combination with oxaliplatin for high-risk patients (see below, Prognosis).[38,80] Here treatment with fluoropyrimidines reduces recurrences by slightly above 20%[81] and there appears to be no additional benefit from oxaliplatin,[82] although this is highly controversial and many guidelines recommend an oxaliplatin combination if the disease is at high risk of recurrence and the patient is <70 years old.[83]

Treatment with chemotherapy is not without risk and it is important that the patient is monitored for side-effects and deteriorating health. Life-threatening side effects occurred in 10% for FLv and 12% for FLOX in the NSABP C-07 trial.[69] Acute side effects include nausea, diarrhoea, skin reactions, allergic and local reactions to infusion, and intense treatment naturally causes more side effects.[68] Sensory neuropathy can develop as a long-term side effect, more common when oxaliplatin is added.[84,85]

Studies are ongoing to explore the optimal schedule, intensity, and duration of chemotherapy. The first results from studies comparing 3 months of treatment with an oxaliplatin combination (FOLFOX or CAPOX) with 6 months of treatment came in 2018. Together the studies included over 10,000 patients. Overall DFS was not non-inferior for 3 months of treatment with fluoropyrimidines and oxaliplatin. In sub-group analysis there was no difference in the low-risk groups between 3 and 6 months but 3 months was inferior in the high-risk stage III group. There were also differences between fluoropyrimidine formulations. CAPOX for 3 months was non-inferior to 6 months, whereas non-inferiority could not be shown using FOLFOX. Side-effects were less common and less severe in the group with 3 months of treatment.[86,87]

Table 2: Endpoints in cancer trials. Adapted from Punt et al 2007

Event	OS	TTR	RFS	DFS
Recurrence	I	E	E	E
Second cancer	I	I	I	E*
Death from same cancer	E	E	E	E
Death from other cancer	E	C	E	E
Death, other	E	C	E	E

OS: Overall Survival, TTR: Time to Recurrence, RFS: Recurrence Free Survival, DFS: Disease Free Survival. C: Censor, E: Event, I: Ignore
*Events from second other cancers are ignored in 2020 guidelines

Prognosis

The measurements of outcomes in cancer have not been standardized but guidelines exist. Punt et al (2007) suggested several endpoints, which will be used in this thesis.[88] Overall survival (OS) includes all mortality and disregards other cancer related events, it is useful for assessing patient benefit from treatment but does not necessarily reflect a treatment specific effect on the cancer. Time to recurrence (TTR) is instead sensitive to the effect of surgery and chemotherapy but patients who die of other causes count as not "needing" treatment. Disease-free survival (DFS) includes both death and recurrence as endpoints and accumulate events quicker than both OS and TTR. DFS is useful for early evaluation of treatment in trials where it is important to consider both gains and potential lethal negative effects. DFS is thus useful in assessing the benefit of chemotherapy as all deaths including treatment-related deaths are included and was recommended for adjuvant treatment trials in a consensus guideline released 2020.[89] In Table 2 the four common endpoints used in adjuvant trials in primary solid tumours are summarized.

It is uncommon for colon cancer to recur locally, instead recurrence commonly manifests as metastasis to the closest capillary and lymphatic networks, the liver, and lungs. Screening for recurrent disease is considered worthwhile in patients where a recurrence could be treated, either through surgery or oncologic treatment. Most recurrences occur in the first three years after surgery.[82,90] Clinical check-up, computed tomography of liver and lungs and CEA are the modes employed to search for distant metastasis one and three years after surgery in Sweden.[91] Other guidelines frequently recommend more intense and longer follow-up,[92] but there is no proven survival benefit to

this practice.[93] Colonoscopy after surgery is performed to look for second cancers and polyps and continued every 5 years after the initial check-ups have been completed until the patient is 75 years old.

More advanced but still non-disseminated stages of colon cancer are associated with increased risk of recurrence. For stage III the sharpest increase in risk is with more advanced T- and N-classification. Other risk factors are identical between stages and are used to make adjuvant treatment decisions (see section on oncological treatment). The risk factors that the European Society for Medical Oncology (ESMO) and National Comprehensive Cancer Network (NCCN) agree upon are: pT4, pN2, less than 12 investigated lymph nodes, poor diffrentiation, lymphovascular and perineural invasion, and obstruction.[38,80,92,94–96] In addition, preoperative CEA above 5 ng/ml was previously considered a risk factor by NCCN but has been removed from the 2019 guidelines although it was added by ESMO in 2020. The side of the primary tumour also correlates with recurrence risk but there is controversy over which side is worse.[11,97–100] Rough estimates of the recurrence risk for each stage are presented in Table 3, compiled from older sources, and published in a review by Böckelman et al (2015).[101] OS is highly dependent on patient comorbidities, but estimates from a 2016 study is also presented in Table 3.[38,76,101,102] Data for stage III is more challenging since most patients should receive chemotherapy if treated according to guidelines and the data from randomized studies which included placebo are old and not indicative of the care provided today.[78]

Table 3: Risk of recurrence without chemotherapy by stage. Adapted from Böckelman et al 2015, Casadaban et al 2016, André et al 2009, Labianca et al 2013.

Stage and treatment	5-year DFS	5-year OS
No adjuvant treatment		
Stage II	81 %	65 %
Stage III	49 %	30-60 % *
Adjuvant treatment		
Stage II	79 %	81 %
Stage III	64 %	75 %

DFS: Disease-Free Survival, OS: Overall Survival
*Mostly old studies, not indicative of current prognosis.

Nomograms and prognostic tools

Nomograms and other tools may be used to help clinicians put together risk factors of recurrence and poor survival in an easy way. A nomogram is the visual representation of an equation, for logistic regressions and proportional hazard regressions several axes are used to calculate a final score corresponding to the predicted risk. For colon cancer several nomograms have been developed, the ACCENT-based web calculator[103], the Memorial Sloan Kettering Cancer Centre (MSKCC) nomogram[104], a Japanese nomogram (Hoshino)[105] and a Special Commission on Cancer nomogram (SCOC)[106]. There was a predictive tool previously available, ”Adjuvant! Online”, but the webpage has been down for updates since 2015. A problem with nomograms is that the formula used to calculate both the contribution of individual risk factors and the underlying hazard function are static. When the prognosis and knowledge about a disease improves, the old nomograms will overestimate the risk and may include risk factors of less importance and/or be missing factors previously unknown. This was partially the case with the MSKCC nomogram which was re-validated in 2019 and the underlying hazard function was changed, but not the risk factors, to reflect modern patients. [107]

Biomarkers in colon cancer

In primary disease, the molecular subgroups are used clinically when selecting treatment with regards to MSI cancers in stage II, which roughly corresponds to CMS1, which have a lower risk of recurrence, thus adjuvant treatment is generally not recommended even in the presence of risk factors. It is still controversial, but patients with MSI tumours also appear to have little benefit from chemotherapy.[108] Other expression profiles have been constructed but are not in use. [109] Mutations in the KRAS-gene impart immunity to certain biological drugs, e.g. EGFR-inhibitors, in CRC exclusively used when patients have metastasized cancer. Mutations in BRAF are uncommon in metastasized disease but confers worse prognosis.[110]

Cell-free DNA or circulating tumour DNA (ctDNA) can be detected in blood and sequenced. It may be the biomarker with most promise as it is both prognostic and can measure treatment effect. [111–113]

There are many non-genetic biomarkers in CRC. One of the more publicised is “Immunoscore®”, a measure of the number and proportion of different inflammatory cells in and around the tumour. It correlates well with prognosis and explains almost 50% of the risk in adjusted models. Unfortunately, it does not seem to be predictive of treatment effect (e.g. benefit from chemotherapy), only prognosis.[114]

Another immunohistochemical method are specific protein stains. There are several proteins that have been identified as prognostic, but the effect is

often limited and not always adjusted for other risk factors making it difficult to assess how it fits into prognostic models. [115–118]

Individual proteins and panels have mainly been explored in the setting of detection of disease and for screening, where the aim has been to replace or supplement faecal testing.[119–121] CEA is a well-established protein biomarker as described previously, it can be used both for monitoring and prognosis/prediction. Other prognostic protein biomarkers are not used clinically. [122]

SCRCR

The Swedish Colorectal Cancer Registry (SCRCR) covers approximately 99% of the patients diagnosed with colon cancer in Sweden, and survival is updated every week with the help of the National Death Registry.[21] The SCRCR was created in 2007 with the inclusion of colon cancers to a previously existing rectal cancer registry. The registry was created to evaluate the national quality of care and outcomes and enables follow-up, evaluation of the adherence to guidelines, and research. It contains information about individual patients and their cancer, detailed information regarding diagnosis, treatment, tumour characteristics and follow-up. Initiatives to investigate patient-related outcomes (quality of life) are ongoing.[123] Use of personal identification numbers enable linkage to other registries (e.g. National Death Registry) and ensures follow-up.

There are guidelines for validation of the Swedish cancer registries published by the Swedish Association of Local Authorities and Regions.[124] The guidelines recommend a random sample from all participating regions and re-abstraction using the registry infrastructure.[125,126] Correction of already registered data is to be avoided since this may bias comparisons between validated and non-validated clinics, and in the case of a random sample, between patients. Validation of the SCRCR shows generally good coverage, but some variables are less covered.[21,127] The primary registration in SCRCR was validated in 2018 with good completeness (98.5%), timeliness (98% are registered within 1 year), and validity (median agreement at re-abstraction approximately 90%). [127]

All recurrences are to be reported to the SCRCR once they have occurred or at least after three and five years when a request is sent to the responsible physician at the hospital where the patient had surgery or is followed.

U-CAN

The Uppsala-Umeå Comprehensive Cancer Consortium is a cooperative bio-banking effort led by Uppsala University and includes Umeå University,

Stockholm University, and KTH Royal Institute of Technology.[128] The aim is to develop a high-quality cancer biobank comprised of different cancer types from defined geographical locations. It includes longitudinal blood samples, tissue samples, self-reported health data and clinical data. In June 2021 it included 2,245 patients from the Uppsala-Örebro region with CRC. For non-metastatic colon cancer, blood is drawn at diagnosis, after surgery but before any adjuvant treatment and at one and three years. If patients recur blood is drawn at diagnosis of recurrence and before initiation of treatment. Tissue samples are taken as biopsies and surgical specimens and kept in the biobank for analysis.

Present investigations

Aims of the thesis

To investigate the prognosis after surgery for colon cancer (Papers I-II), validate these findings (Paper III) and create a tool for individual patient prediction (Paper IV). The specific aims were

I. To investigate the current risk of recurrence after surgery for colon cancer and validate clinical risk factors
II. To investigate the value of assessing the sidedness, pT3- and pT4-subclassification, LNR, tumour deposits, CEA and CRP against a baseline of routinely recorded clinicopathological variables in predicting risk of recurrence and OS
III. To validate the reporting of recurrences to the SCRCR
IV. To create and validate a nomogram for colon cancer recurrence risk

Patients and methods

Study design

Papers I, III and IV were retrospective studies of SCRCR patients. Paper II combined prospective testing for biomarkers and retrospective collection of data from electronic patient records. Study design, period of inclusion, number of patients and data sources are shown in Table 4.

Table 4. Study designs, period of patient diagnosis and surgery, number of patients and data sources

Study	Design	Period	Patients	Data sources
I	Retrospective national cohort study	2007-2012	14,325	SCRCR
II	Retrospective and prospective regional cohort study	2010-2015	416	SCRCR, EPR
III	Retrospective multicentre study	2010-2018	2,500	SCRCR, EPR
IV	Retrospective international cohort study	2007-2014	Model: 11,943 Validation: 4,191 and 12,769	SCRCR, Norwegian Cancer registry

SCRCR: Swedish Colorectal Cancer Registry. EPR: Electronic Patient Records

Patients

SCRCR data was requested through the Regional Cancer Centres for papers I-IV and electronic patient records were scrutinized, and selected data collected manually for papers II-III. In paper I, data for all patients who had undergone surgery for colon cancer in stage I through III in Sweden between 2007 and 2012 was requested from the SCRCR. Of the 16,659 patients, 14,352 patients who were alive 30 days after surgery, received no neoadjuvant chemotherapy and underwent radical surgery were selected.

The cohort in paper II consisted of all 685 patients diagnosed with colon cancer (ICD-code C18 and C19) and registered as an adenocarcinoma in the SCRCR from Uppsala County that had surgery between January 2010 and December 2015. Of these, 504 patients had TNM7/UICC stage I-III disease. Exclusion of patients that were not resected, with non-radical resections, polypectomies, patients treated with neoadjuvant chemotherapy, or who died

within 30 days of surgery yielded a final cohort size of 416 patients. Blood samples for analysis were collected at diagnosis and approximately 6-10 weeks after surgery but before any adjuvant chemotherapy was initiated.

For paper III all patients diagnosed with (Uppsala) or operated for (Gävleborg) stage I-III CRC in Uppsala and Gävleborg counties between 2010 - 2018 were included (n=2,500). Patients with stage IV disease were included in the review in Uppsala County (n=393).

In paper IV patients with stage I-III colon cancer diagnosed between 2007 and 2014 were identified in the SCRCR (n=21,008). After exclusion of less advanced synchronous cancers, patients where no radical resection was performed or who received neo-adjuvant chemotherapy or early death 11,943 patients were used to construct a prediction model for recurrences within 5-years. The model was validated with an internal material consisting of 4,191 patients. The model was then externally validated with 12,769 stage II-III patients from the Norwegian colon cancer registry selected using the same criteria as the Swedish patients.

Statistical Methods

Descriptive statistics were calculated using either the Pearson χ^2 or Fisher exact test for categorical data, and non-parametric tests, Mann–Whitney *U* and Kruskal Wallis, for continuous data. To calculate survival at different time-points the Kaplan-Meier method and log-rank test were used with censoring.

Unadjusted (one variable) and adjusted (multiple variables) Cox proportional hazards regressions were used to calculate hazard ratios for time to event outcomes. IBM SPSS Statistics version 24 and 25 (IBM Corp, Armonk, NY) was used to calculate statistics for paper I and II. *R* version 3.6.1 was used to calculate the statistics for papers III and IV. Results were considered statistically significant if p value <0.05.

In paper I patients were stratified according to the risk factors of recurrence as defined by NCCN and available in the SCRCR (emergency surgery, pT4, pN2, <12 investigated lymph nodes, high grade malignancy, vascular invasion, perineural invasion). NCCN then also defined CEA above 5 ng/ml before surgery as a risk factor, however, this was not available in the SCRCR and thus not included.[95]

The 5-year risk of recurrence was calculated for each stage and further stratified by the number of risk factors. Unadjusted and adjusted hazard ratios were calculated for OS and TTR. Changes in recurrence risk over time were investigated using the Pearson χ^2-test.

In paper II unadjusted and adjusted hazard ratios (HR) for TTR and OS were calculated using the Cox proportional hazards model. The variables investigated were sidedness, pT- and pN-subclassification, LNR, tumour deposits, CEA and CRP. Each predictor variable was evaluated with

adjustment for age, sex, comorbidities, emergency presentation, ≥12 surveyed LN, grade of malignancy, vascular and perineural invasion, and adjuvant chemotherapy.

Patient records were scrutinized in paper III for the existence of metastatic disease, both synchronous (primary metastatic) and metachronous (recurrence). Discrepancies between the registry and records were noted and tabulated. After review of the records ensuring that all recurrences were recorded, recurrence risks were calculated for colon cancer patients (n=1,416) stratified by stage and ESMO risk factors as published in 2020.[96]

In paper IV a Cox proportional hazards regression model was constructed using available registry data. Variables were included if deemed clinically relevant and of statistical significance. The model was evaluated for proportional hazards and collinearity. Area under the receiver operation curve/c-index (AUC) and bootstrapped calibration curves were used to assess the model fit. Model validation was then performed using one-out cross validation in the model data set before being validated in an internal and external data set with AUC and calibration plots. Calibration curves were created by plotting the predicted outcome versus the actual outcome. A nomogram was then drawn using the package rms in R, the nomogram was adjusted to no adjuvant treatment to allow for separate axes for the benefit from monotherapy (25%) and combination therapy (40%) as reported in clinical trials and guidelines.[38,65,69,78] Comparisons with other nomograms were then performed by calculating the predicated risks for all patients and comparing the AUC and calibration with the new nomogram.

Ethical considerations

The regional ethical review committee (2010/198, 2013/093/1, 2014/419, 2015/419, 2018/490), the Swedish Ethical Review Authority (2019/06156) and Norwegian regional Ethics Committee South-East (2020/91617) approved the studies.

Patients included in papers I-IV were included based upon assumed consent, they had consented to the SCRCR and the invasion of privacy from scrutinization of the records was deemed small enough that separate consent was not needed. Patients with data for CEA and CRP in paper II had signed consent forms for the U-CAN biobank.

Results

In paper I recurrences were seen in 16% of patients after radical surgery, with 4% being local recurrences. The five-year recurrence rate was 5% in stage I, 12% in stage II, and 33% in stage III. The rate was heterogenous within stages II-III when stratified by risk factors and pT-stage and ranged between 9% - 31% in stage II and between 17% - 44% in stage III. Recurrence risks from papers I, III and IV are summarised in Table 5 stratified by stage and risk factors for patients not receiving chemotherapy.

Table 5: Five-year recurrence risks without adjuvant treatment in papers I, III and IV

Stage	**Risk Factors / Risk group***	**Paper I (2007-12)**		**Paper III (2010-18)**		**Paper IV (2010-14)**	
		N	***RR%***	***N***	***RR%***	***N***	***RR%***
I	**0**	1,330	5%				
	1	874	5%				
	≥2	109	6%				
II	**0/Low**	2,743	9%	337	6%	3,389	8%
	1/Intermediate	1,836	11%	105	9%	2,086	10%
	≥2/High	835	22%	79	23%	912	17-27%
III	**0/Low**	410	22%	111	26%	590	23%
	1	627	29%			719	28%
	≥2/High	987	45%	94	43%	834	36-53%

N: N at start (paper I) and at risk (paper III-IV). RR% 5-year recurrence risk
*Risk factors/groups. Paper I: pT4, pN2, emergency surgery, high-grade malignancy, vascular or perineural invasion, and <12 sampled lymph nodes.
Paper III: Low risk= pT1-3N0-1, no risk factors. Intermediate risk = pT3N0 with one of perioperative obstruction, high grade malignancy, lympho-vascular invasion and perineural invasion. High risk = pT4 and/or <12 investigated nodes
Paper IV: Risk factors: emergency surgery, perforation, pT4, high grade malignancy, lymphovascular invasion and <12 investigated lymph nodes. Patients split by 0,1,2, ≥3 risk factors in original paper.

In the multivariable analysis, an increased risk for recurrence was seen for male sex, emergency surgery, distal vascular ligature or no reported ligature, pT and pN classification, vascular and perineural invasion, and postoperative complications. Adjuvant treatment correlated with a lower risk of recurrence.

When analysing OS additional factors were of importance; high grade malignancy, low lymph node yield, high age, low body mass index and comorbidities all correlated with worse prognosis. Better prognosis was seen when the tumour was staged before surgery, surgery was performed with intermediate resection margins and in patients who received adjuvant treatment (adjusted HRs in Table 6).

Improvements in the quality of care were demonstrated by increasing circumferential excision margins and higher lymph node yields. Reporting to the registry also increased during the investigated time-period. No correlation between year of surgery and recurrence rate or OS was seen.

Patients in paper II (Uppsala County) did not differ from the national cohort in paper I regarding mean age, sex distribution and tumour sidedness. However, there were proportionally more T1-2 and N0 cancers in the national material. Missingness for these variables was low in both materials. Data on malignancy grade, vascular invasion and perineural invasion were missing in a higher proportion in the national material (6-27%) than the Uppsala County material (3-5%). The percentage of complete cases regarding the above variables was 68% in the SCRCR and 95% in the Uppsala County material. In a more modern material from the SCRCR covering 2010-2014, the missingness was at most 10% (paper IV). Baseline parameters were similar in both distribution and univariable Cox regressions for TTR and OS.

In the adjusted model, sidedness, pN-substage and postoperative CEA correlated with recurrence. Increased hazard of mortality was correlated with right-sided tumours, pT4, node positivity (except pN1c), LNR, postoperative CEA and CRP when adjusting for baseline variables (Table 6). Right-sided tumours were correlated with worse prognosis in paper II. Sidedness was not analysed in the adjusted model in paper I since it was not significantly correlated with recurrences in the unadjusted model. Stage-stratified analysis was done as an exploratory analysis in paper I where right-sided tumours correlated with better prognosis in stage II with no difference in stage III. In paper IV right-sided tumours had a more favourable prognosis.

Table 6: Multivariable/Adjusted hazard ratios for time to recurrence from papers I, II and III

Variable (reference)	Level	Paper I HR (CI)	Paper II HR (CI)	Paper III (Stage II) HR (CI)	Paper III (Stage III) HR (CI)
Sex (Male)	***Female***	0.9 (0.8-0.9)			
Surgery (Elective)	***Emergency/ Obstruction***	**1.6 (1.5-1.8)**	**2.5 (1.5-4.1)**	1.8 (0.9-3.8)	**1.9 (1.3-2.7)**
pT (pT1)	***pT3ab/a/pT3***	**2.3 (1.6-3.2)**	1.6 (0.4-5.9)		1.2 (0.5-3)
	pT3b		1.6 (0.5-5)		
	pT3cd/c	**3.3 (2.4-4.7)**	1.5 (0.4-4.8)		
	pT3d		2.3 (0.7-8.1)		
	pT4a/pT4a/ pT4	**5.0 (3.5-7.2)**	2.1 (0.6-7.2)	1.8 (0.9-3.7)	1.8 (0.7-4.6)
	pT4b	**4.3 (2.8-6.4)**	1.9 (0.5-7)		
pN (pN0)	***pN1/pN1a***	**2.1 (1.9-2.4)**	**2.6 (1.2-5.7)**		
	pN1b		**2.3 (1-5.1)**		
	pN1c		1.5 (0.3-6.9)		
	pN2a/pN2	**3.4 (2.9-3.9)**	**5.4 (2.4-12)**		**2.3 (1.7-3.2)**
	pN2b	**4.3 (3.5-5.2)**	**6.4 (2.8-15)**		
Grade (Low)	***High-grade***	1 (0.9-1.1)	1.6 (0.9-2.8)	0.9 (0.4-1.9)	1.2 (0.8-1.8)
LVI (No)	***Yes***	**1.5 (1.3-1.6)**	1.4 (0.8-2.5)	**2.4 (1.2-4.7)**	**1.6 (1.1-2.3)**
PNI (No)	***Yes***	**1.3 (1.2-1.5)**	1.7 (1-3)	2.0 (1-4.3)	1.1 (0.8-1.7)
Adjuvant (No)	***Yes***	**0.8 (0.7-0.9)**	0.6 (0.4-1.1)		
Investigated nodes (≥12)	***<12***		2 (0.6-6.8)	0.7 (0.2-3)	**0.7 (0.3-0.9)**
LNR	***0-1***		3.6 (0.7-18)		
TD (No)	***Yes***		0.8 (0.4-1.5)		
CEA ng/ml (<5)	***Preoperative >5***		1.2 (0.7-2.2)		
	Postoperative >5		**2.6 (1.2-5.6)**		
CRP mg/l (>10)	***Preoperative >10***		1 (0.6-1.7)		
	Postoperative >10		1.8 (0.9-3.9)		
Side (Right)	***Left***		**0.5 (0.3-0.9)**		

CI=95% Confidence intervals in parentheses.
LVI: lymphovascular invasion, PNI: Perineural invasion, TD: Tumour deposits
Values in bold were statistically significant ($p < 0.05$).

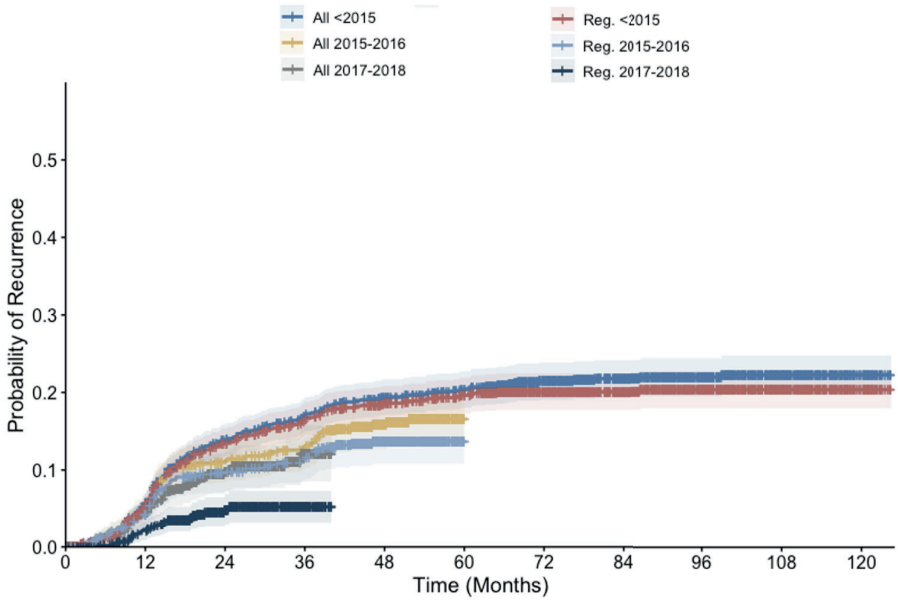

Figure 2: ***Recurrences rates were less accurate in colorectal cancer patients operated <5 years ago.*** *Registered recurrences and all recurrences (after validation) were plotted by year of diagnosis for all M0-patients included in the validation (paper III), i.e. also non-radically operated patients. Censoring for loss to follow-up and death. Shaded area denotes 95% confidence intervals.*

In paper III, 3% (13/393) of M1 patients in Uppsala County were M0, while 0.6% (8/1,314) of M0 patients were M1. Among non-recurrent M0 patients, 0.5% (5/1,048) had non-registered recurrences. Three percent (7/213) of patients registered as recurrent M0 were non-recurrent. No patients with more than 5 years of follow-up had missed recurrences.

In Gävleborg County, primary metastatic patients were not scrutinized. One percent (12/1,186) of M0 patients were M1. Among patients registered as non-recurrent M0 patients, 5% (54/1,024) had non-registered recurrences. One percent (2/138) of patients registered as recurrent M0 were non-recurrent. In patients with more than 5-years of follow-up, 1% (9/630) of the patients had recurrences within 5-years of surgery that were missing from the registry and 1% (7/630) had recurrences after 5-years that were missing from the registry. There were no significant differences between patients with a registered and non-registered recurrence regarding demographic and disease variables (data not published). Overall, there was only a small difference for patients followed >5 years while 3-year recurrence rates are somewhat reliable for patients followed over 3 years (Figure 2). The curves for patients with a

more recent diagnosis (2017-2018) differ substantially and more follow-up time is needed before evaluation of recurrence rate based on the SCRCR.

In the complete data set with regards to recurrences 1,416 patients had been diagnosed with colon cancer and fulfilled the inclusion criteria used in papers I-II. The 5-year recurrence risk was 17%. In stage II patients without adjuvant treatment the 5-year recurrence risks in low, intermediate, and high-risk groups were 6%, 9% and 23 %, respectively. In stage III the 5-year recurrence risk was 26% and 43% in low and high-risk groups, respectively. Stage stratified risk of recurrence is presented in Figure 3 and Table 5. In regression analysis of the ESMO risk factors for stage II colon cancer, only vascular invasion was statistically significant (Table 6).

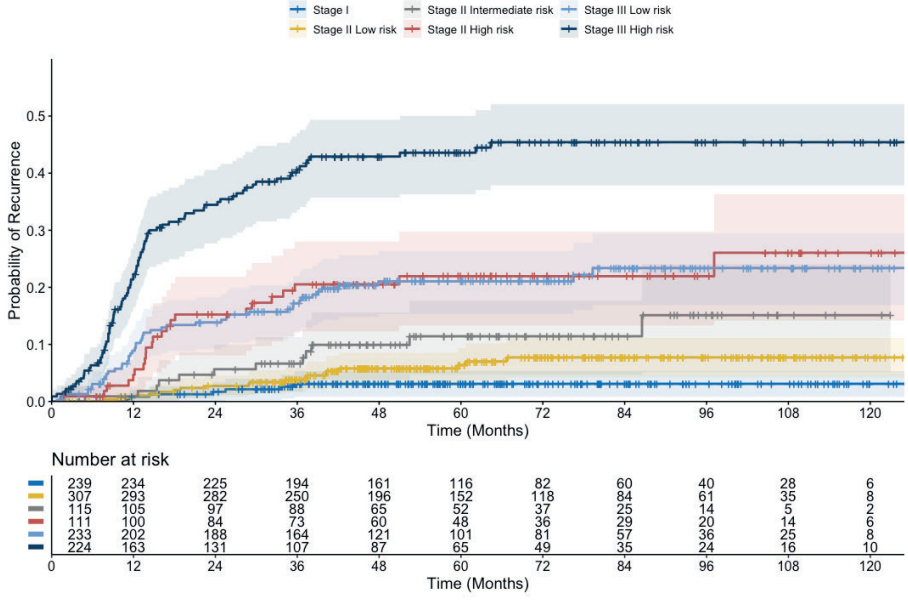

Figure 3: ***Stage and risk group stratified probability of recurrence*** *in material of 1,416 colon cancer patients where all recurrences were registered (Paper III). Patients were stratified according to stage and risk group. Shaded areas denote 95% confidence intervals.*

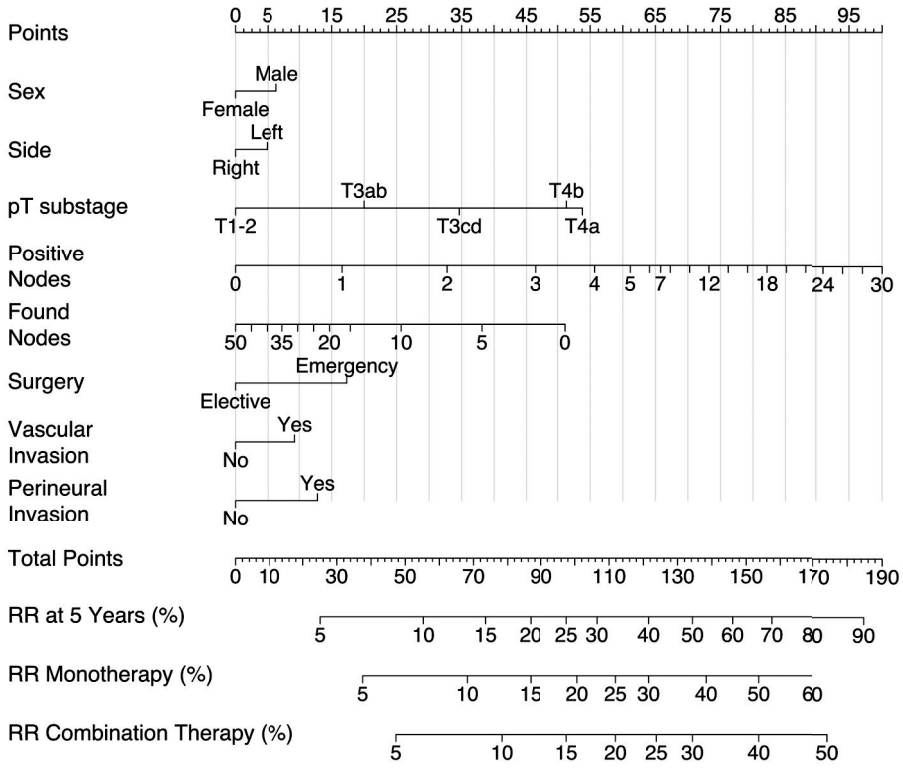

Figure 4: Nomogram. Start from second axis (sex), sum corresponding points read from top axis (points), continue with each axis until you reach the axis for total points. The total points correspond to a predicted recurrence risk at 5 years. Subsequent axes represent predicted recurrence risk if mono- or combination chemotherapy is given.
RR: Recurrence risk
E.g. a male with a right-sided, pT3cd with 2/20 positive nodes and vascular invasion gets 98 points (7+0+34+33+15+0+9+0) corresponding to a recurrence risk of about 25%. The next two axes describe the risk of recurrence if adjuvant therapy is given, with monotherapy it is 19% and with combination therapy the recurrence risk is 15%.

In paper IV the 5-year recurrence rate was 8% in stage II and 23% in stage III in patients without risk factors (pT4, emergency surgery, perforation, pT4, <12 investigated lymph nodes, high grade malignancy and lymphovascular or perineural invasion) who did not receive adjuvant treatment (Table 5). Variables for the nomogram (Figure 4) were selected from unadjusted regressions, guidelines and known collinearity (e.g. nodes). The final predictive model included: sex, sidedness, pT-substages, number of positive and found lymph nodes, emergency surgery, lymphovascular and perineural invasion, and adjuvant treatment. Perforation and malignancy grade were non-significant in the adjusted model and contributed little with regards to HR and

were removed to simplify calculation of the nomogram. Model assumptions were confirmed and bootstrapped calibration and leave-one-out cross-validation confirmed a good fit in the model data. The nomogram was then drawn and adjusted to no adjuvant treatment to allow for separate axes for the benefit from monotherapy and combination therapy. AUC was 0.78 in the model data while it was 0.76 in the internal validation data. The model was fitted to the external data from Norway, performing somewhat worse with an AUC of 0.70. The calibration curve indicated that the model was more pessimistic than reality but that it fitted well in the range of 5-25% risk of recurrence. When four other nomograms were applied to the data, they received AUC ranging from 0.69-0.78, while calibration curves revealed both pessimistic and optimistic models compared to reality, and all were calibrated worse than the SCRCR nomogram in the data (Figure 5).

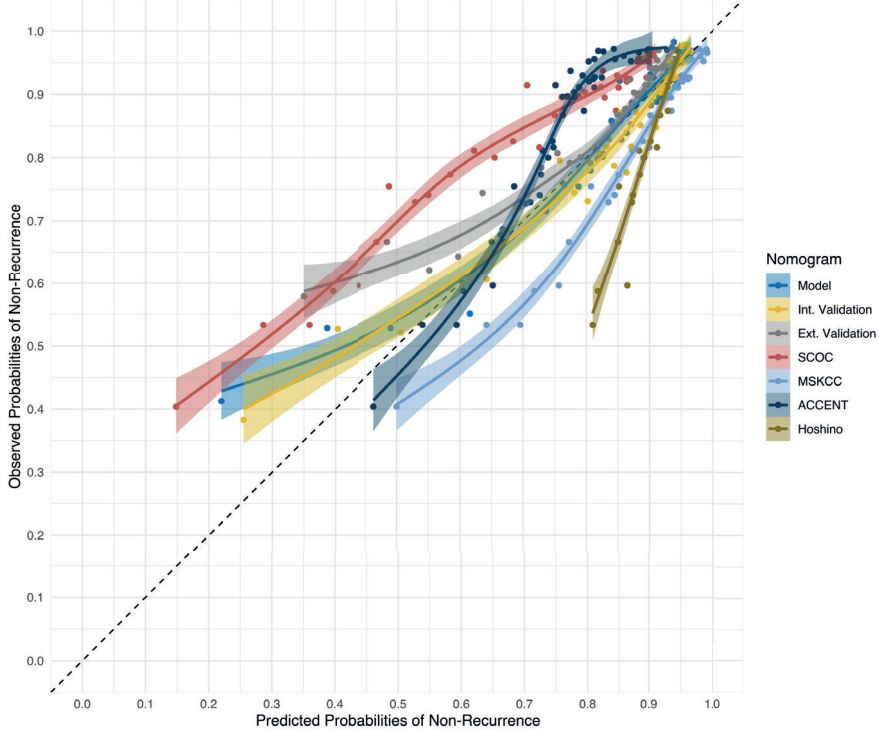

Figure 5: Predicted probabilities of recurrences vs observed probability of recurrence in model data, validation data (Int. Validation), external validation in Norwegian data (Ext. Validation), and calibration of other nomograms in SCRCR data (SCOC; Special commission on cancer, MSKCC; Memorial Sloan Kettering Cancer Centre, ACCENT, Hoshino; Japanese nomogram). Shaded area denotes confidence intervals. Dashed line is the optimal fit where predicted risk equals actual risk.

Discussion

Current prognosis

In papers I-III, the routinely used risk factors, as defined by international guidelines and staging recommendations were confirmed. Compared with the recurrence risks presented in guidelines, a large proportion has such a low risk that adjuvant treatment is of questionable value considering the negative impact of the treatment. Patients with no risk factors in both stage II and III could potentially be spared all or some adjuvant treatment. This was first pointed out by Lars Påhlman et al. in 2016[129] and hinted at by others[107,130] but the findings in this thesis are the first to put it in numbers.[131]

Many oncologists consider a 5% absolute reduction in recurrence enough to initiate treatment, but no consensus exists,[132–134] and patients may be of different opinions why shared decision making is of utmost importance.[135,136] The relative benefit of chemotherapy is probably the same over time, i.e. the 20% reduction in recurrences as seen in several early trials including patients during the 1980s and 1990s is similar to patients treated in 2020. This was not investigated in this thesis since registry data cannot reliably answer this.

As prognosis improves, the absolute benefit decreases. If 100 patients are at risk of recurrence and 20 will recur, chemotherapy would decrease this to 16 recurrences. This was the situation in stage II in trials reported some 20 years ago. It is estimated that the risk has decreased since then, but by how much is not clear.[131] If the risk is 15 in 100, chemotherapy would prevent three recurrences for 100 treated patients. As the risk decrease further, fewer patients are helped, even though chemotherapy prevents 20% of recurrences and at some point, the risk-benefit balance shifts towards risk rather than benefit.

Prognostic factors

pT3-subclassification was prognostic in paper I, but not reported with high accuracy in the SCRCR. It was investigated further in paper II and the importance of deeper tumour invasion was confirmed. pT3d tends to have a worse prognosis than pT3a-c and was numerically as bad as pT4. pT4a similarly had a worse prognosis than pT4b in both the large, national material and the Uppsala County material. In previous versions of the TNM staging manual the classification was the reverse, but visceral involvement (previously T4a, now T4b) was deemed worse than tumour growth involving

the peritoneum and the categories changed in TNM6. Further studies are needed, but perhaps this indicates a need to change it back to preserve the logic of worse prognosis with higher number or letter.

Stratification by N-subclassification contributes to improve the risk assessment in both papers I and II and the largest difference is seen in the pN2 stage. LNR was analysed in paper II and had a strong correlation with recurrence, HR 3.7 for every 0.1 increase in unadjusted analysis but not when adjusting for the base classification by pN-stage since they are strongly correlated. When using the optimal LNR cut-off of 0.13, also described elsewhere,[137] 46% of patients above cut-off recurred compared with 16% below.

Elevated CEA, taken after surgery and within the window of potential initiation of adjuvant therapy, was independently correlated with increased risk of recurrence. CEA is commonly used to follow patients at risk, indicating recurrent or residual disease, but currently it does not guide treatment decisions. If elevated, as many as 48% of patients recurred in paper II.

In both paper I and II there were correlations between risk factors of recurrence and OS, but the strongest risk factors for increased risk of death were patient comorbidities and age. Elevated CRP after surgery correlated with increased mortality, which may be explained by complications suffered after surgery, a risk factor for recurrence and mortality in paper I and mortality in paper II (unadjusted analysis).

Sidedness was correlated with recurrence risk in both the national cohorts and the county cohort, but the results were not consistent with right sided tumours being favourable in the national material while it was worse in the county material. Stage seems to be an important differentiator with more impact from sidedness in stage II, with the results from the larger cohorts being more reliable since sidedness is well reported in the SCRCR.

The results of paper I and II both underline the importance of clinicopathological factors for estimating the risk of recurrence and the need for adjuvant treatment.

Validity

After the publication of article I, some international researchers inquired about the accuracy and correctness in the SCRCR regarding recurrences. It was estimated in paper III that only 1-2% of recurrences (occurring within 5-years after surgery) are missing from the registry in patients operated more than 5 years ago, if Uppsala County is assumed to be representative of other university hospital counties and Gävleborg County is representative of non-university hospital counties. The risk factors presented in the new ESMO guidelines[96] were then investigated and additional risk stratification seemed possible in stage III. Since ESMO recommends 3-months of combination therapy for the low-risk group, there is thus little room to step this down to

monotherapy for low-risk patients in stage III if treatment is given according to international guidelines.

Nomogram

Work to put the information from papers I-III together in a user-friendly way was then commenced with paper IV. By creating a tool that is easy to use and understand (compared to calculating the risk from hazard ratios) the aim was to aid in the risk-benefit discussion taking place daily in GI-oncologist offices. To improve this further a web-based calculator is in the works. The newly constructed nomogram performed well in external validation, especially if accounting for the percentage of unreported recurrences in the Norwegian registry (94% concordance between patient records and the registry).[138] The SCRCR nomogram performed better compared with the other nomograms in the validation data set but received the same AUC as the MSKCC nomogram. However, AUC measures the ranking of patients but does not consider the actual predicted value. Calibration on the other hand takes this into account and is good measure of the accuracy of the predictions. The SCOC nomogram overestimated the risk of recurrence by about 20% and the MSKCC and ACCENT nomograms underestimated the risk in high-risk patients. The Japanese nomogram (Hoshino) was only applicable to stage II patients and overestimated the risk of recurrence.

It seems difficult to get the AUC above about 0.8 in a colon cancer predictive model (paper IV), in part because of chance, but also because of the limits of clinicopathological factors. The combination of a new biomarker and CEA was investigated to improve the prediction and detection of recurrences. It was left out of this thesis due to a wish for anonymity from the industry partner developing the new biomarker. The combination was sensitive enough but when the specificity was evaluated in controls it was so low that it would not be viable as a marker in the clinic, too many patients would be identified as high-risk patients. The combination could potentially be used in low-risk stage II patients where the negative predictive value would be higher. There are several lessons to be learned from the design of the study that will ease the evaluation of future markers. The selection of patients and sample times should be stringent to facilitate easy analysis, more samples are not necessarily better, and the first evaluation should be done in a limited number of longitudinal samples. The study was performed in a stepwise fashion, recurrent cases were first evaluated with controls included when sensitivity was confirmed to be high. This saves biobanked material if the sensitivity is low but can give false hope if specificity is low instead.

Strengths

The strength of a large population-based registry was used in papers I and IV to estimate the risks in the population and calculate the baseline risk of already known risk factors and construct a model. The results should be representative

of the health care provided in Sweden, with colorectal surgical units varying in size and case volume but with common guidelines. Compared with previous studies, performed in the early 2000s, it represents a modern material where diagnostics, surgical technique and oncological treatment are optimized. No formal, statistical, comparison was carried out in papers I-IV with previous materials since it was beyond the scope of these papers. To meet this need a review of most, if not all, large trials of adjuvant treatment and population-based cohorts was performed and we found that the yearly improvement in proportion of recurrence free patients was 0.54% after adjusting for case numbers, tumour location and stage distribution.[131] It is unlikely that the improvement will continue at this rate and it should be viewed in the light of major health care improvements over the last 40-60 years. The finding of reduced recurrence rates[139] and improved survival[140] over time has also been seen in other retrospective materials. Since we stress the risk of recurrence as the proper endpoint for evaluating the need for adjuvant therapy rather than DFS or OS, crucial for the validity of the results are the reporting of recurrences which was evaluated in paper III as only differing by 1-2% from reality.

Death is a firm endpoint but the reporting of recurrences behind TTR is up to the carefulness of the clinician, usually a surgeon or nurse. The registry has tried to secure that as many as possible of diagnosed recurrences are reported. The completeness of reporting of recurrences has not been formally validated in the entirety of Sweden. In Uppsala and Gävleborg counties, clinical records for every CRC patient have been scrutinized why no single clinically detected recurrence is missed. In the age groups where CRC is most common, virtually no patients are missed due to moving to another area in Sweden or abroad. The similarity in recurrence risks for each stage between Uppsala and Gävleborg after correction, and the rest of Sweden indirectly suggest that missingness nationwide is low. The results are also likely representative of at least Norway,[138] but potentially all the Nordic countries considering our similar standard of health care. The model constructed in paper IV could thus be used in other similar countries.

Limitations

Because of the limitations of retrospective materials, no attempt was made to investigate the benefit from chemotherapy, only the risk of recurrence in untreated patients. This is less sensitive to bias and selection compared to treated patients (who must both be at risk and fit enough to tolerate treatment). The information about which chemotherapy patients received was not complete for all patients, these patients were counted as treated in the analyses. To truly know the recurrence risk a large untreated, unselected cohort would be needed, something that would be unethical with the current knowledge about the benefits from chemotherapy.

The SCRCR covers at least 99% of patients diagnosed with CRC in Sweden. There are however some patients who may be missing from the SCRCR and national cancer registry. Autopsy rates are low in Sweden and patients dying from other causes with unknown cancers are unlikely to be reported. Frail or otherwise sick patients may decline (or be declined) work-up, and sometimes other patients decide that not knowing is best. Patients with unknown primaries with extensive spread where no treatment is available may also be left out for the simple reason that there is no definitive diagnosis. However, these are not the patients who will have surgery and then potentially receive chemotherapy to reduce the risk of recurrence. They are equally important when talking about colorectal cancer, but beyond the scope of this thesis.

There is also limit to what can be collected in a registry and some risk factors could only be assessed in the smaller material collected for paper II impacting the power of these conclusions. It would have been of value to have MSI-status and CEA levels in papers I and IV, but it was not available in the registry. MSI/MRR status was added in 2017 and CEA is reported for metastatic patients since 2016. MSI data was available for half the patients in paper II but more frequently analysed in patients with metastatic disease introducing bias why it was not published.

Missing data was more common in the purely registry-based papers I and IV, while it was less of a problem in paper II where data could be checked in the electronic patient records and pathology slides re-evaluated. Complications are also known to be underreported in the registry which impacts paper I.[141] In paper III the aim was to check the coverage level of the registry regarding recurrences, but analysis of recurrence rates were also performed according to risk groups. The cohort was newer (2010-2018 vs 2007-2012) than in paper I and missingness decreased with time making it a smaller problem.

Conclusions

Risk factors for recurrence could be confirmed and investigated utilizing the strength of both a population-based registry, and a smaller, thoroughly staged and controlled cohort. The finding of lower risk than previously was then validated by investigating the accuracy of the registry in both a previously investigated and a new material. The recurrence rate in SCRCR data was accurate to between 1-2% percent. A risk prediction model, nomogram, was then constructed to aid clinicians and patients, the model was validated in external data and performed better than other nomograms. Some emerging factors added little value and most of the risk was predicted by tumour and node stage in papers I-IV. To better predict the recurrence risk after colon cancer surgery combinations of biomarkers and clinicopathological features are needed. Already biobanked material offers the opportunity to produce data, however, care must be taken when designing these retrospective studies.

Future perspectives

As noted in the discussion further stratification should be possible, and the key to this lies in analyses on a molecular level such as proteomics and genomics. Multiple molecular and genetic risk factors have been identified previously, but most of these investigations were performed for a single factor, adjusting for basic clinicopathological variables.[114–118,120,122,142–146] The aim should be to improve identification of low-risk patients who can be spared treatment and intensive follow-up and those at high risk needing more intense treatment.[147]

A project mainly using U-CAN samples was started in 2018 to sequence both tumour DNA and RNA, and analyse serum markers for approximately 1,000 patients with CRC. Among these are some of Uppsala County cases from papers II and III, but the project also includes rectal cancer and metastatic cases. Further investigations are ongoing, and the huge amount of data that is generated will be a treasure trove for researchers for years to come.
Immunohistochemical staining is ongoing for all colon cancer cases included in the Uppsala County cohort, including cases from 2016-2018. This will allow analysis of known genetic factors (*KRAS*, *BRAF*, *PIK3CA*), protein expression (e.g. MMR proteins, CDX2, SATB-2, SMADs) but also previously unknown factors in combination with basic clinical factors and DNA, RNA, and protein data. A subset of patients with large plasma aliquots in the biobank are also suitable for circulating tumour DNA (ctDNA) analysis.[111–113,148,149] The aim is to identify both new high- and low-risk features, potential biomarkers and improve our understanding of the disease.

A protein panel for CRC diagnosis will be the basis for a new attempt at identifying biomarkers correlated to recurrence risk. It is a retrospective case-control study similar to the previous attempt but with more rigorous selection of patients and material. A novel method of developing cut-off points has already been tested for diagnostics and in other public data sets and will be used to set it apart from previous studies. The diagnostic capabilities will be evaluated in a prospective material of patients referred for colonoscopies due to suspected cancer.

The nomogram will hopefully be developed into a web-tool and published for free with graphs detailing the risk to help clinicians and patients even further in their discussion on risks versus benefits of chemotherapy. One can envision a similar nomogram for rectal cancer patients, perhaps even to try to predict a complete clinical response to neoadjuvant therapy.

Future studies could incorporate the risk stratification gains that the updated nomogram provide, and any biomarkers and risk factors identified during the above investigations. Introduction of these into routine healthcare should be performed in a controlled way and assessed in prospectively collected materials, potentially randomized, especially if treatment is guided by biomarkers. The landmark IDEA collaboration finding that shorter CAPOX is non-inferior in at least low-risk patients may be taken one step further with treatment-effect markers. In patients at risk and with positive markers one could sample the markers again after 3 months of treatment, and if it they are negative stop treatment, or if positive treatment may continue for another 3 months. Faster inclusion would be possible if the investigation is performed as a (inter)national multi-arm study, with additional samples taken to allow exploration of other markers. Patients would potentially benefit with regards to recurrence rates, treatment intensity and quality of life.

Populärvetenskaplig sammanfattning

Tjock- och ändtarmscancer är den tredje vanligaste cancern i Sverige och drabbar årligen cirka 6 000 personer, varav 4 000 har tjocktarmscancer. Hälften av patienterna med tjocktarmscancer har måttlig till hög risk för återfall (cancerstadie II och III). Behandlingen är kirurgi och eventuellt efterföljande cellgiftsbehandling, vilken minskar risken för återfall och ökar överlevnaden, men behandlingen är inte biverkningsfri. Den kirurgiska samt onkologiska behandlingen har senaste åren förbättrats avsevärt och det finns indikationer på att studier som gjordes i början av 2000-talet beskriver en högre risk för återfall och död i tjocktarmscancer än vad som är aktuellt idag.

Cirka 14 000 patienter från det svenska tjock- och ändtarmscancerregistret (SCRCR) utgjorde grunden för den första studien (artikel I) där risken för återfall idag beskrivs. Vid större tumör, men utan spridning till lymfan (stadie II), har 3/4 av patienterna en låg risk för återfall. Vid spridning till lymfan (stadie III) är risken högre, men 1/5 har så låg risk att de sannolikt inte tjänar tillräckligt på intensiva cellgifter. Risken för återfall beskrevs bra av de faktorer som används kliniskt och som varit kända sedan länge.

Ytterligare riskfaktorer undersöktes hos en mindre grupp patienter som alla bodde i Uppsala län (artikel II). Om CEA, ett protein som används som tumörmarkör och mäts i blod, var förhöjt efter kirurgin var det en riskfaktor för återfall men huvuddelen av risken beskrevs av de redan kända faktorerna.

För att vara säkra på att risken för återfall var så låg som i artikel I granskades alla journaler från patienter som opererats för tjock- och ändtarmscancer i två regioner (artikel III). Nästan 2% av återfallen saknades i SCRCR hos patienter som opererats för mer än 5 år sedan. Risken för återfall, när alla återfall var medräknade i de två regionerna, var dock lägre än i artikel I vilket talar för att siffrorna som presenterats stämmer väl med verkligheten.

För att underlätta för läkare att förutsäga risken och kommunicera den till patienter skapades ett riskberäkningsverktyg med hjälp av data från SCRCR (artikel IV). Verktyget testades i registerdata från Norge. I jämförelse med andra liknande verktyg var det nyutvecklade verktyget minst lika bra på att förutsäga återfall. När förmågan att förutsäga risken för återfall testades fungerade det nya verktyget bättre än liknande, tidigare publicerade verktyg.

Sammanfattningsvis är risken för återfall lägre idag än tidigare, hur låg denna risk är har inte tidigare satts i siffror. Genom att använda registerdata från historiska patienter kan mycket av risken förutsägas för de som insjuknar idag.

Acknowledgments

This work was conducted at the Department of Immunology, Genetics and Pathology at Uppsala University, and supported by the Swedish Cancer Foundation.

I want to thank my main supervisor **Tobias Sjöblom** for supporting my work and supplying me with ample challenges, collaborative research opportunities, and timely feedback.

These first tentative steps into research would not have been possible if not for my co-supervisor **Bengt Glimelius**, and I want to express the greatest gratitude to you. Thank you for your support, your mentoring, for sending me feedback on the latest revision at midnight (on your vacation), and for always being curious!

Lasse Karlsson, for co-supervising my thesis, but also for your mentorship regarding balancing the life of being a surgical resident and scientist. **Mats Hellström** for checking up on how my thesis is going and always signing the yearly review.

The late, great **Lars Påhlman** also needs an acknowledgement, if not for him and Bengt this would not be happening.

Klara for the discussions, comparisons and exchange of ideas improving our work. **Joakim** for the statistical and out of the box discussions. **Luis** for your work with the BGI project and jovial pun-contests, **PH** for your help with the U-CAN data and your handle on the proceedings. **Artur** for helping us out with annotations and review of thousands of slides. The entire **MolCan** team, past and present, an excellent group to work with. **RCC** helping with the raw data for Papers I-IV. Our research partners in **Norway** and the **Kreftregister**.

The **Department of Surgery in Gävle** for allowing the time off to do research and supporting me throughout, I promise to one day finish my Residency.

Carolina, my love, thank you for putting up with me and my multiple jobs and supporting me through it all. **Idun** for not deleting my entire thesis while slamming on the keyboard. My **father** for inspiring me to be a physician and scientist and pushing me too always be better. My **mother** for always supporting me and boosting my confidence since I was little. And last, but not least, all my **friends** and **family** for your support.

References

1 Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A *et al.* Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA Cancer J Clin* 2021; 71: 209–249.

2 Ferlay J, Colombet M, Soerjomataram I, Mathers C, Parkin DM, Piñeros M *et al.* Estimating the global cancer incidence and mortality in 2018: GLOBOCAN sources and methods. *Int J Cancer* 2019; 144: 1941–1953.

3 OFFICIAL STATISTICS OF SWEDEN Statistics – Health and Medical Care Cancer Incidence in Sweden 2019. www.socialstyrelsen.se/statistik-och-data/statistik/statistikamnen/cancer/ (accessed 13 Jan 2021).

4 Allemani C, Weir HK, Carreira H, Harewood R, Spika D, Wang X-S *et al.* Global surveillance of cancer survival 1995–2009: analysis of individual data for 25 676 887 patients from 279 population-based registries in 67 countries (CONCORD-2). *The Lancet* 2014; 385: 977–1010.

5 Birgisson H, Talbäck M, Gunnarsson U, Påhlman L, Glimelius B. Improved survival in cancer of the colon and rectum in Sweden. *Eur J Surg Oncol EJSO* 2005; 31: 845–853.

6 Hanahan D, Weinberg RA. The Hallmarks of Cancer. *Cell* 2000; 100: 57–70.

7 Jass JR. Classification of colorectal cancer based on correlation of clinical, morphological and molecular features. *Histopathology* 2007; 50: 113–130.

8 Lengauer C, Kinzler KW, Vogelstein B. Genetic instability in colorectal cancers. *Nature* 1997; 386: 623–627.

9 Fodde R, Kuipers J, Rosenberg C, Smits R, Kielman M, Gaspar C *et al.* Mutations in the APC tumour suppressor gene cause chromosomal instability. *Nat Cell Biol* 2001; 3: 433–438.

10 Vogelstein B, Papadopoulos N, Velculescu VE, Zhou S, Diaz LA, Kinzler KW. Cancer Genome Landscapes. *Science* 2013; 339: 1546–1558.

11 Missiaglia E, Jacobs B, D'Ario G, Di Narzo AF, Soneson C, Budinska E *et al.* Distal and proximal colon cancers differ in terms of molecular, pathological, and clinical features. *Ann Oncol* 2014; 25: 1995–2001.

12 Guinney J, Dienstmann R, Wang X, de Reyniès A, Schlicker A, Soneson C *et al.* The consensus molecular subtypes of colorectal cancer. *Nat Med* 2015; 21: 1350–1356.

13 Hanahan D, Weinberg RA. Hallmarks of Cancer: The Next Generation. *Cell* 2011; 144: 646–674.

14 Roerink SF, Sasaki N, Lee-Six H, Young MD, Alexandrov LB, Behjati S *et al.* Intra-tumour diversification in colorectal cancer at the single-cell level. *Nature* 2018; 556: 457–462.

15 Naxerova K, Reiter JG, Brachtel E, Lennerz JK, van de Wetering M, Rowan A *et al.* Origins of lymphatic and distant metastases in human colorectal cancer. *Science* 2017; 357: 55–60.

16 del Carmen S, Sayagués JM, Bengoechea O, Anduaga MF, Alcazar JA, Gervas R *et al.* Spatio-temporal tumor heterogeneity in metastatic CRC tumors: a mutational-based approach. *Oncotarget* 2018; 9. doi:10.18632/oncotarget.26081.
17 Bhullar DS, Barriuso J, Mullamitha S, Saunders MP, O'Dwyer ST, Aziz O. Biomarker concordance between primary colorectal cancer and its metastases. *EBioMedicine* 2019; 40: 363–374.
18 Holch JW, Ricard I, Stintzing S, Modest DP, Heinemann V. The relevance of primary tumour location in patients with metastatic colorectal cancer: A meta-analysis of first-line clinical trials. *Eur J Cancer 1990* 2017; 70: 87–98.
19 Jones S, Chen W -d., Parmigiani G, Diehl F, Beerenwinkel N, Antal T *et al.* Comparative lesion sequencing provides insights into tumor evolution. *Proc Natl Acad Sci* 2008; 105: 4283–4288.
20 Haggar FA, Boushey RP. Colorectal Cancer Epidemiology: Incidence, Mortality, Survival, and Risk Factors. *Clin Colon Rectal Surg* 2009; 22: 191–197.
21 Kodeda K, Nathanaelsson L, Jung B, Olsson H, Jestin P, Sjövall A *et al.* Population-based data from the Swedish Colon Cancer Registry. *Br J Surg* 2013; 100: 1100–1107.
22 Brunicardi FC, Andersen DK, Billiar TR, Dunn DL, Hunter JG (eds.). *Schwartz's principles of surgery*. Eleventh edition. McGraw-Hill Education: New York, 2019.
23 Valle L, Vilar E, Tavtigian SV, Stoffel EM. Genetic predisposition to colorectal cancer: syndromes, genes, classification of genetic variants and implications for precision medicine. *J Pathol* 2019; 247: 574–588.
24 Dyson JK, Rutter MD. Colorectal cancer in inflammatory bowel disease: What is the real magnitude of the risk? *World J Gastroenterol WJG* 2012; 18: 3839–3848.
25 Haggitt RC, Glotzbach RE, Soffer EE, Wruble LD. Prognostic factors in colorectal carcinomas arising in adenomas: Implications for lesions removed by endoscopic polypectomy. *Gastroenterology* 1985; 89: 328–336.
26 Joseph NE, Sigurdson ER, Hanlon AL, Wang H, Mayer RJ, MacDonald JS *et al.* Accuracy of Determining Nodal Negativity in Colorectal Cancer on the Basis of the Number of Nodes Retrieved on Resection. *Ann Surg Oncol* 2003; 10: 213–218.
27 Jestin P, Påhlman L, Glimelius B, Gunnarsson U. Cancer staging and survival in colon cancer is dependent on the quality of the pathologists' specimen examination. *Eur J Cancer* 2005; 41: 2071–2078.
28 Zhang M-R, Xie T-H, Chi J-L, Li Y, Yang L, Yu Y-Y *et al.* Prognostic role of the lymph node ratio in node positive colorectal cancer: a meta-analysis. *Oncotarget* 2016; 7: 72898–72907.
29 Svenska Kolorektalcancerregistret (Tjocktarm) Interaktiv Statistik. 2021. www.statistik.incanet.se/kolorektal/kolon/ (accessed 22 Jul 2021).
30 Astin M, Griffin T, Neal RD, Rose P, Hamilton W. The diagnostic value of symptoms for colorectal cancer in primary care: a systematic review. *Br J Gen Pract* 2011; 61: e231–e243.
31 Gunnarsson H, Ekholm A, Olsson LI. Emergency presentation and socioeconomic status in colon cancer. *Eur J Surg Oncol* 2013; 39: 831–836.
32 Jestin P, Nilsson J, Heurgren M, Påhlman L, Glimelius B, Gunnarsson U. Emergency surgery for colonic cancer in a defined population. *Br J Surg* 2005; 92: 94–100.

33 Fransgaard T, Caspar Thygesen L, Gögenur I. The impact of postoperative complications and delay of adjuvant chemotherapy on oncological outcomes in patients with colorectal cancer. *Colorectal Dis* 2021; 23: 1132–1140.
34 Valentín-López B, Ferrándiz-Santos J, Blasco-Amaro J-A, Morillas-Sáinz J-D, Ruiz-López P. Assessment of a rapid referral pathway for suspected colorectal cancer in Madrid. *Fam Pract* 2012; 29: 182–188.
35 Jensen KH, Maina PJ-C. Cancer pathways are associated with improved long-term survival. *Dan Med J* 2015; 62.
36 Lieberman D, Ladabaum U, Cruz-Correa M, Ginsburg C, Inadomi JM, Kim LS *et al.* Screening for Colorectal Cancer and Evolving Issues for Physicians and Patients: A Review. *JAMA* 2016; 316: 2135–2145.
37 Schreuders EH, Ruco A, Rabeneck L, Schoen RE, Sung JJY, Young GP *et al.* Colorectal cancer screening: a global overview of existing programmes. *Gut* 2015; 64: 1637–1649.
38 Labianca R, Nordlinger B, Beretta GD, Mosconi S, Mandalà M, Cervantes A *et al.* Early colon cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2013; 24: vi64–vi72.
39 Thirunavukarasu P, Talati C, Munjal S, Attwood K, Edge SB, Francescutti V. Effect of Incorporation of Pretreatment Serum Carcinoembryonic Antigen Levels Into AJCC Staging for Colon Cancer on 5-Year Survival. *JAMA Surg* 2015; 150: 747–755.
40 Lin B-R, Lin Y-L, Lai H-S, Lee P-H, Chang K-J, Liang J-T. Overall Survival of Stage III Colon Cancer with Only One Lymph Node Metastasis Is Independently Predicted by Preoperative Carcinoembryonic Antigen Level and Lymph Node Sampling Status. *PLoS ONE* 2015; 10. doi:10.1371/journal.pone.0137053.
41 Hansen HJ, Snyder JJ, Miller E, Vandevoorde JP, Miller ON, Hines LR *et al.* Carcinoembryonic antigen (CEA) assay. A laboratory adjunct in the diagnosis and management of cancer. *Hum Pathol* 1974; 5: 139–147.
42 Blausen.com Staff. Colon Anatomy. 2014 www.en.wikiversity.org/wiki/WikiJournal_of_Medicine/Medical_gallery_of_Blausen_Medical_2014 (accessed 30 Jul 2021).
43 Kenig J, Richter P. Definition of the rectum and level of the peritoneal reflection – still a matter of debate? *Videosurgery Miniinvasive Tech* 2013; 8: 183–186.
44 Wasserman MA, McGee MF, Helenowski IB, Halverson AL, Boller A-M, Stryker SJ. The anthropometric definition of the rectum is highly variable. *Int J Colorectal Dis* 2016; 31: 189–195.
45 D'Souza N, de Neree tot Babberich MPM, d'Hoore A, Tiret E, Xynos E, Beets-Tan RGH *et al.* Definition of the Rectum: An International, Expert-based Delphi Consensus. *Ann Surg* 2019; 270: 955–959.
46 Standring S, Gray H (eds.). *Gray's anatomy: the anatomical basis of clinical practice*. 40. ed., reprinted. Churchill Livingstone Elsevier: Edinburgh, 2009.
47 Roy MK, Pipara A, Kumar A. Surgical management of adenocarcinoma of the transverse colon: What should be the extent of resection? *Ann Gastroenterol Surg* 2021; 5: 24–31.
48 Lightner A, Pemberton J. The Role of Temporary Fecal Diversion. *Clin Colon Rectal Surg* 2017; 30: 178–183.
49 Hohenberger W, Weber K, Matzel K, Papadopoulos T, Merkel S. Standardized surgery for colonic cancer: complete mesocolic excision and central ligation – technical notes and outcome. *Colorectal Dis* 2009; 11: 354–364.

50 Bernhoff R, Martling A, Sjövall A, Granath F, Hohenberger W, Holm T. Improved survival after an educational project on colon cancer management in the county of Stockholm – A population based cohort study. *Eur J Surg Oncol* 2015; 41: 1479–1484.
51 Bernhoff R, Holm T, Sjövall A, Granath F, Ekbom A, Martling A. Increased lymph node harvest in patients operated on for right-sided colon cancer: a population-based study. *Colorectal Dis* 2012; 14: 691–696.
52 On behalf of the COLOR (COlon cancer Laparoscopic or Open Resection) study group, Deijen CL, Vasmel JE, de Lange-de Klerk ESM, Cuesta MA, Coene P-PLO *et al.* Ten-year outcomes of a randomised trial of laparoscopic versus open surgery for colon cancer. *Surg Endosc* 2017; 31: 2607–2615.
53 Ohtani H, Tamamori Y, Arimoto Y, Nishiguchi Y, Maeda K, Hirakawa K. A Meta-Analysis of the Short- and Long-Term Results of Randomized Controlled Trials That Compared Laparoscopy-Assisted and Conventional Open Surgery for Colorectal Cancer. *J Cancer* 2011; 2: 425–434.
54 Green BL, Marshall HC, Collinson F, Quirke P, Guillou P, Jayne DG *et al.* Long-term follow-up of the Medical Research Council CLASICC trial of conventional *versus* laparoscopically assisted resection in colorectal cancer: Conventional *versus* laparoscopically assisted surgery for colonic and rectal cancer. *Br J Surg* 2013; 100: 75–82.
55 Liu VX, Rosas E, Hwang J, Cain E, Foss-Durant A, Clopp M *et al.* Enhanced Recovery After Surgery Program Implementation in 2 Surgical Populations in an Integrated Health Care Delivery System. *JAMA Surg* 2017; : e171032.
56 Brännström F, Gunnarsson U. Risk Factors for Local Recurrence after Emergency Resection for Colon Cancer: Scenario in Sweden. *Dig Surg* 2016; 33: 503–508.
57 Pisano M, Zorcolo L, Merli C, Cimbanassi S, Poiasina E, Ceresoli M *et al.* 2017 WSES guidelines on colon and rectal cancer emergencies: obstruction and perforation. *World J Emerg Surg WJES* 2018; 13: 36.
58 Van Cutsem E, Cervantes A, Adam R, Sobrero A, Van Krieken JH, Aderka D *et al.* ESMO consensus guidelines for the management of patients with metastatic colorectal cancer. *Ann Oncol* 2016; 27: 1386–1422.
59 Huang C-Q, Min Y, Wang S-Y, Yang X-J, Liu Y, Xiong B *et al.* Cytoreductive surgery plus hyperthermic intraperitoneal chemotherapy improves survival for peritoneal carcinomatosis from colorectal cancer: a systematic review and meta-analysis of current evidence. *Oncotarget* 2017; 8: 55657–55683.
60 Xeloda® - FASS Vårdpersonal. www.fass.se/LIF/product?userType=0&nplId=20010202000047 (accessed 29 Jul 2021).
61 Eidinoff ML, Knoll JE, Klein D. Effect of 5-fluorouracil on the incorporation of precursors into nucleic acid pyrimidines. *Arch Biochem Biophys* 1957; 71: 274–275.
62 Rang HP, Ritter J, Flower RJ, Henderson G. *Rang & Dale's pharmacology*. Eight edition. Elsevier/Churchill Livingstone: Edinburgh 2016
63 Lawrence W, Terz JJ, Horsley S, Donaldson M, Lovett WL, Brown PW *et al.* Chemotherapy as an adjuvant to surgery for colorectal cancer. *Ann Surg* 1975; 181: 616–623.
64 Morgan RG. Leucovorin enhancement of the effects of the fluoropyrimidines on thymidylate synthase. *Cancer* 1989; 63: 1008–1012.

65 Schmoll H-J, Twelves C, Sun W, O'Connell MJ, Cartwright T, McKenna E *et al.* Effect of adjuvant capecitabine or fluorouracil, with or without oxaliplatin, on survival outcomes in stage III colon cancer and the effect of oxaliplatin on post-relapse survival: a pooled analysis of individual patient data from four randomised controlled trials. *Lancet Oncol* 2014; 15: 1481–1492.
66 Kidani Y, Noji M, Tashiro T. Antitumor activity of platinum(II) complexes of 1,2-diamino-cyclohexane isomers. *Gan* 1980; 71: 637–643.
67 Díaz-Rubio E, Sastre J, Zaniboni A, Labianca R, Cortés-Funes H, de Braud F *et al.* Oxaliplatin as single agent in previously untreated colorectal carcinoma patients: a phase II multicentric study. *Ann Oncol* 1998; 9: 105–108.
68 André T, Boni C, Mounedji-Boudiaf L, Navarro M, Tabernero J, Hickish T *et al.* Oxaliplatin, fluorouracil, and leucovorin as adjuvant treatment for colon cancer. *N Engl J Med* 2004; 350: 2343–2351.
69 Yothers G, O'Connell MJ, Allegra CJ, Kuebler JP, Colangelo LH, Petrelli NJ *et al.* Oxaliplatin As Adjuvant Therapy for Colon Cancer: Updated Results of NSABP C-07 Trial, Including Survival and Subset Analyses. *J Clin Oncol* 2011; 29: 3768–3774.
70 Selcukbiricik F, Sag AA, Kanıtez M, Bilici A, Mandel NM. Neoadjuvant systemic therapy for patients with gastric cancer: Current concepts and outcomes. *J Oncol Sci* 2016; 1: 25–30.
71 FOxTROT Collaborative Group. Feasibility of preoperative chemotherapy for locally advanced, operable colon cancer: the pilot phase of a randomised controlled trial. *Lancet Oncol* 2012; 13: 1152–1160.
72 Seymour M, Morton D. FOxTROT: an international randomised controlled trial in 1052 patients (pts) evaluating neoadjuvant chemotherapy (NAC) for colon cancer. *Journal of Clinical Oncology*. 2019, p Abstract 3504.
73 Glimelius B, Tiret E, Cervantes A, Arnold D, on behalf of the ESMO Guidelines Working Group. Rectal cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2013; 24: vi81–vi88.
74 Conroy T, Lamfichekh N, Etienne P-L, Rio E, FRANCOIS E, Mesgouez-Nebout N *et al.* Total neoadjuvant therapy with mFOLFIRINOX versus preoperative chemoradiation in patients with locally advanced rectal cancer: Final results of PRODIGE 23 phase III trial, a UNICANCER GI trial. *J Clin Oncol* 2020; 38: 4007–4007.
75 Bahadoer RR, Dijkstra EA, van Etten B, Marijnen CAM, Putter H, Kranenbarg EM-K *et al.* Short-course radiotherapy followed by chemotherapy before total mesorectal excision (TME) versus preoperative chemoradiotherapy, TME, and optional adjuvant chemotherapy in locally advanced rectal cancer (RAPIDO): a randomised, open-label, phase 3 trial. *Lancet Oncol* 2021; 22: 29–42.
76 André T, Boni C, Navarro M, Tabernero J, Hickish T, Topham C *et al.* Improved Overall Survival With Oxaliplatin, Fluorouracil, and Leucovorin As Adjuvant Treatment in Stage II or III Colon Cancer in the MOSAIC Trial. *J Clin Oncol* 2009; 27: 3109–3116.
77 Regionalt cancercentrum Norr, Norra sjukvårdsregionen. Onkologirapport 2019 från Svenska kolorektalcancerregistret. Regionalt cancercentrum Norr, Norra sjukvårdsregionen, 2021.
78 Efficacy of adjuvant fluorouracil and folinic acid in colon cancer: International Multicentre Pooled Analysis of Colon Cancer Trials (IMPACT) investigators. *The Lancet* 1995; 345: 939–944.
79 Ragnhammar P, Hafström L, Nygren P, Glimelius B, SBU-group. Swedish Council of Technology Assessment in Health Care. A systematic overview of chemotherapy effects in colorectal cancer. *Acta Oncol* 2001; 40: 282–308.

80 National Comprehensive Cancer Network, Inc. NCCN Guidelines Version 2.2017 Colon Cancer. 2017. www.nccn.org/professionals/physician_gls/pdf/colon.pdf (accessed 10 Jun 2017).
81 Quasar Collaborative Group, Gray R, Barnwell J, McConkey C, Hills RK, Williams NS *et al.* Adjuvant chemotherapy versus observation in patients with colorectal cancer: a randomised study. *The Lancet* 2007; 370: 2020–2029.
82 Shah MA, Renfro LA, Allegra CJ, André T, de Gramont A, Schmoll H-J *et al.* Impact of Patient Factors on Recurrence Risk and Time Dependency of Oxaliplatin Benefit in Patients With Colon Cancer: Analysis From Modern-Era Adjuvant Studies in the Adjuvant Colon Cancer End Points (ACCENT) Database. *J Clin Oncol* 2016; 34: 843–853.
83 Tournigand C, André T, Bonnetain F, Chibaudel B, Lledo G, Hickish T *et al.* Adjuvant therapy with fluorouracil and oxaliplatin in stage II and elderly patients (between ages 70 and 75 years) with colon cancer: subgroup analyses of the Multicenter International Study of Oxaliplatin, Fluorouracil, and Leucovorin in the Adjuvant Treatment of Colon Cancer trial. *J Clin Oncol* 2012; 30: 3353–3360.
84 Stefansson M, Nygren P. Oxaliplatin added to fluoropyrimidine for adjuvant treatment of colorectal cancer is associated with long-term impairment of peripheral nerve sensory function and quality of life. *Acta Oncol* 2016; 55: 1227–1235.
85 Kidwell KM, Yothers G, Ganz PA, Land SR, Ko CY, Cecchini RS *et al.* Long-term neurotoxicity effects of oxaliplatin added to fluorouracil and leucovorin as adjuvant therapy for colon cancer: results from National Surgical Adjuvant Breast and Bowel Project trials C-07 and LTS-01. *Cancer* 2012; 118: 5614–5622.
86 André T, Vernerey D, Mineur L, Bennouna J, Desrame J, Faroux R *et al.* Three Versus 6 Months of Oxaliplatin-Based Adjuvant Chemotherapy for Patients With Stage III Colon Cancer: Disease-Free Survival Results From a Randomized, Open-Label, International Duration Evaluation of Adjuvant (IDEA) France, Phase III Trial. *J Clin Oncol* 2018; 36: 1469–1477.
87 Grothey A, Sobrero AF, Shields AF, Yoshino T, Paul J, Taieb J *et al.* Duration of Adjuvant Chemotherapy for Stage III Colon Cancer. *N Engl J Med* 2018; 378: 1177–1188.
88 Punt CJA, Buyse M, Köhne C-H, Hohenberger P, Labianca R, Schmoll HJ *et al.* Endpoints in Adjuvant Treatment Trials: A Systematic Review of the Literature in Colon Cancer and Proposed Definitions for Future Trials. *J Natl Cancer Inst* 2007; 99: 998–1003.
89 Cohen R, Vernerey D, Bellera C, Meurisse A, Henriques J, Paoletti X *et al.* Guidelines for time-to-event end-point definitions in adjuvant randomised trials for patients with localised colon cancer: Results of the DATECAN initiative. *Eur J Cancer* 2020; 130: 63–71.
90 Sargent DJ, Wieand HS, Haller DG, Gray R, Benedetti JK, Buyse M *et al.* Disease-Free Survival Versus Overall Survival As a Primary End Point for Adjuvant Colon Cancer Studies: Individual Patient Data From 20,898 Patients on 18 Randomized Trials. *J Clin Oncol* 2005; 23: 8664–8670.
91 Cancercentrum i Samverkan. National Guidelines: Colorectal Cancer. Natl. Guidel. Colorectal Cancer. www.cancercentrum.se/samverkan/cancerdiagnoser/ tjocktarm-andtarm-och-anal/tjock--och-andtarm/vardprogram/gallande-vardprogram/ (accessed 1 Jul 2021).

92 Benson AB, Venook AP, Al-Hawary MM, Arain MA, Chen Y-J, Ciombor KK *et al.* National Comprehensive Cancer Network. Colon Cancer (Version 2.2019). 2019. www.nccn.org/professionals/physician_gls/pdf/colon.pdf (accessed 10 Jun 2019).
93 Wille-Jørgensen P, Syk I, Smedh K, Laurberg S, Nielsen DT, Petersen SH *et al.* Effect of More vs Less Frequent Follow-up Testing on Overall and Colorectal Cancer–Specific Mortality in Patients With Stage II or III Colorectal Cancer: The COLOFOL Randomized Clinical Trial. *JAMA* 2018; 319: 2095.
94 Schmoll HJ, Van Cutsem E, Stein A, Valentini V, Glimelius B, Haustermans K *et al.* ESMO Consensus Guidelines for management of patients with colon and rectal cancer. a personalized approach to clinical decision making. *Ann Oncol* 2012; 23: 2479–2516.
95 Benson AB, Venook AP, Al-Hawary MM, Cederquist L, Chen Y-J, Ciombor KK *et al.* NCCN Guidelines Insights: Colon Cancer, Version 2.2018. *J Natl Compr Canc Netw* 2018; 16: 359–369.
96 Argilés G, Tabernero J, Labianca R, Hochhauser D, Salazar R, Iveson T *et al.* Localised colon cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2020; 31: 1291–1305.
97 Brungs D, Aghmesheh M, de Souza P, Ng W, Chua W, Carolan M *et al.* Sidedness is prognostic in locoregional colon cancer: an analysis of 9509 Australian patients. *BMC Cancer* 2017; 17: 251.
98 Petrelli F, Tomasello G, Borgonovo K, Ghidini M, Turati L, Dallera P *et al.* Prognostic Survival Associated With Left-Sided vs Right-Sided Colon Cancer: A Systematic Review and Meta-analysis. *JAMA Oncol* 2017; 3: 211–219.
99 Weiss JM, Pfau PR, O'Connor ES, King J, LoConte N, Kennedy G *et al.* Mortality by stage for right- versus left-sided colon cancer: analysis of surveillance, epidemiology, and end results--Medicare data. *J Clin Oncol* 2011; 29: 4401–4409.
100 Huang C-W, Tsai H-L, Huang M-Y, Huang C-M, Yeh Y-S, Ma C-J *et al.* Different clinicopathologic features and favorable outcomes of patients with stage III left-sided colon cancer. *World J Surg Oncol* 2015; 13: 257.
101 Böckelman C, Engelmann BE, Kaprio T, Hansen TF, Glimelius B. Risk of recurrence in patients with colon cancer stage II and III: A systematic review and meta-analysis of recent literature. *Acta Oncol* 2015; 54: 5–16.
102 Casadaban L, Rauscher G, Aklilu M, Villenes D, Freels S, Maker AV. Adjuvant Chemotherapy is Associated with Improved Survival in Patients with Stage II Colon Cancer. *Cancer* 2016; 122: 3277–3287.
103 Renfro LA, Grothey A, Xue Y, Saltz LB, André T, Twelves C *et al.* ACCENT-Based Web Calculators to Predict Recurrence and Overall Survival in Stage III Colon Cancer. *JNCI J Natl Cancer Inst* 2014; 106. doi:10.1093/jnci/dju333.
104 Weiser MR, Landmann RG, Kattan MW, Gonen M, Shia J, Chou J *et al.* Individualized prediction of colon cancer recurrence using a nomogram. *J Clin Oncol* 2008; 26: 380–385.
105 Hoshino N, Hasegawa S, Hida K, Kawada K, Ganeko R, Sugihara K *et al.* Nomogram for predicting recurrence in stage II colorectal cancer. *Acta Oncol* 2016; 55: 1414–1417.
106 Zafar SN, Hu C-Y, Snyder RA, Cuddy A, You YN, Lowenstein LM *et al.* Predicting Risk of Recurrence After Colorectal Cancer Surgery in the United States: An Analysis of a Special Commission on Cancer National Study. *Ann Surg Oncol* 2020. doi:10.1245/s10434-020-08238-7.

107 Konishi T, Shimada Y, Hsu M, Wei IH, Pappou E, Smith JJ *et al.* Contemporary Validation of a Nomogram Predicting Colon Cancer Recurrence, Revealing All-Stage Improved Outcomes. *JNCI Cancer Spectr* 2019; 3: pkz015.
108 Ribic CM, Sargent DJ, Moore MJ, Thibodeau SN, French AJ, Goldberg RM *et al.* Tumor Microsatellite-Instability Status as a Predictor of Benefit from Fluorouracil-Based Adjuvant Chemotherapy for Colon Cancer. *N Engl J Med* 2003; 349: 247–257.
109 Van Schaeybroeck S, Allen WL, Turkington RC, Johnston PG. Implementing prognostic and predictive biomarkers in CRC clinical trials. *Nat Rev Clin Oncol* 2011; 8: 222–232.
110 Van Cutsem E, Köhne C-H, Láng I, Folprecht G, Nowacki MP, Cascinu S *et al.* Cetuximab Plus Irinotecan, Fluorouracil, and Leucovorin As First-Line Treatment for Metastatic Colorectal Cancer: Updated Analysis of Overall Survival According to Tumor KRAS and BRAF Mutation Status. *J Clin Oncol* 2011; 29: 2011–2019.
111 Diehn M, Alizadeh AA, Adams H-P, Lee JJ, Klassen S, Palma JF *et al.* Early prediction of clinical outcomes in resected stage II and III colorectal cancer (CRC) through deep sequencing of circulating tumor DNA (ctDNA). *J Clin Oncol* 2017; 35: 3591–3591.
112 Parikh A, Seventer EV, Boland G, Hartwig A, Jaimovich A, Raymond V *et al.* O-016 Serial assessment of cell-free circulating tumor DNA (ctDNA) to assess treatment effect and minimal residual disease during neoadjuvant and adjuvant therapy in colorectal cancer. *Ann Oncol* 2019; 30. doi:10.1093/annonc/mdz154.015.
113 Reinert T, Henriksen TV, Christensen E, Sharma S, Salari R, Sethi H *et al.* Analysis of Plasma Cell-Free DNA by Ultradeep Sequencing in Patients With Stages I to III Colorectal Cancer. *JAMA Oncol* 2019. doi:10.1001/jamaoncol.2019.0528.
114 Pagès F, Mlecnik B, Marliot F, Bindea G, Ou F-S, Bifulco C *et al.* International validation of the consensus Immunoscore for the classification of colon cancer: a prognostic and accuracy study. *The Lancet* 2018; 391: 2128–2139.
115 Siesing C, Sorbye H, Dragomir A, Pfeiffer P, Qvortrup C, Pontén F *et al.* High RBM3 expression is associated with an improved survival and oxaliplatin response in patients with metastatic colorectal cancer. *PLOS ONE* 2017; 12: e0182512.
116 Eberhard J, Gaber A, Wangefjord S, Nodin B, Uhlén M, Ericson Lindquist K *et al.* A cohort study of the prognostic and treatment predictive value of SATB2 expression in colorectal cancer. *Br J Cancer* 2012; 106: 931–938.
117 Kaprio T, Fermér C, Hagström J, Mustonen H, Böckelman C, Nilsson O *et al.* Podocalyxin is a marker of poor prognosis in colorectal cancer. *BMC Cancer* 2014; 14. doi:10.1186/1471-2407-14-493.
118 Hansen TF, Kjær-Frifeldt S, Eriksen AC, Lindebjerg J, Jensen LH, Sørensen FB *et al.* Prognostic impact of CDX2 in stage II colon cancer: results from two nationwide cohorts. *Br J Cancer* 2018; 119: 1367–1373.
119 Jones JJ, Wilcox BE, Benz RW, Babbar N, Boragine G, Burrell T *et al.* A Plasma-Based Protein Marker Panel for Colorectal Cancer Detection Identified by Multiplex Targeted Mass Spectrometry. *Clin Colorectal Cancer* 2016; 15: 186-194.e13.

120 Mahboob S, Ahn SB, Cheruku HR, Cantor D, Rennel E, Fredriksson S *et al.* A novel multiplexed immunoassay identifies CEA, IL-8 and prolactin as prospective markers for Dukes' stages A-D colorectal cancers. *Clin Proteomics* 2015; 12. doi:10.1186/s12014-015-9081-x.
121 Chen H, Zucknick M, Werner S, Knebel P, Brenner H. Head-to-Head Comparison and Evaluation of 92 Plasma Protein Biomarkers for Early Detection of Colorectal Cancer in a True Screening Setting. *Clin Cancer Res* 2015; 21: 3318–3326.
122 Ghanipour L, Darmanis S, Landegren U, Glimelius B, Påhlman L, Birgisson H. Detection of Biomarkers with Solid-Phase Proximity Ligation Assay in Patients with Colorectal Cancer. *Transl Oncol* 2016; 9: 251–255.
123 QoLiCOL - Quality of Life in Colon Cancer - Full Text View - ClinicalTrials.gov. www.clinicaltrials.gov/ct2/show/NCT02530593 (accessed 15 Jul 2021).
124 Lambe, Mats, Rejmyr-Davis M. Validering av kvalitetsregister på INCA Version 2.0. 2021.
125 Bray F, Parkin DM. Evaluation of data quality in the cancer registry: Principles and methods. Part I: Comparability, validity and timeliness. *Eur J Cancer* 2009; 45: 747–755.
126 Parkin DM, Bray F. Evaluation of data quality in the cancer registry: Principles and methods Part II. Completeness. *Eur J Cancer* 2009; 45: 756–764.
127 Moberger P, Sköldberg F, Birgisson H. Evaluation of the Swedish Colorectal Cancer Registry: an overview of completeness, timeliness, comparability and validity. *Acta Oncol* 2018; 57: 1611–1621.
128 Glimelius B, Melin B, Enblad G, Alafuzoff I, Beskow A, Ahlström H *et al.* U-CAN: a prospective longitudinal collection of biomaterials and clinical information from adult cancer patients in Sweden. *Acta Oncol* 2017; 57: 187–194.
129 Påhlman LA, Hohenberger WM, Matzel K, Sugihara K, Quirke P, Glimelius B. Should the Benefit of Adjuvant Chemotherapy in Colon Cancer Be Re-Evaluated? *J Clin Oncol* 2016; 34: 1297–1299.
130 Salem ME, Yin J, Goldberg RM, Pederson LD, Wolmark N, Alberts SR *et al.* Evaluation of the change of outcomes over a 10-year period in patients with stage III colon cancer: pooled analysis of 6501 patients treated with fluorouracil, leucovorin, and oxaliplatin in the ACCENT database. *Ann Oncol* 2020; 31: 480–486.
131 Osterman E, Hammarström K, Imam I, Osterlund E, Sjöblom T, Glimelius B. Recurrence Risk after Radical Colorectal Cancer Surgery—Less Than before, But How High Is It? *Cancers* 2020; 12: 3308.
132 Stiggelbout AM, de Haes JC, van de Velde CJ. Adjuvant chemotherapy in node negative breast cancer: patterns of use and oncologists' preferences. *Ann Oncol* 2000; 11: 631–633.
133 Engelhardt EG, de Haes HCJM, van de Velde CJH, Smets EMA, Pieterse AH, Stiggelbout AM. Oncologists' weighing of the benefits and side effects of adjuvant systemic therapy: Has it changed over time? *Acta Oncol* 2015; 54: 956–959.
134 Ragnhammar P, Brorsson B, Nygren P, Glimelius B, SBU-group. Swedish Council of Technology Assessment in Health Care. A prospective study of the use of chemotherapy in Sweden and assessment of the use in relation to scientific evidence. *Acta Oncol* 2001; 40: 391–411.

135 Jansen SJT, Kievit J, Nooij MA, Haes JCJM de, Overpelt IME, Slooten H van *et al.* Patients' preferences for adjuvant chemotherapy in early-stage breast cancer: is treatment worthwhile? *Br J Cancer* 2001; 84: 1577–1585.
136 Simes RJ, Coates AS. Patient preferences for adjuvant chemotherapy of early breast cancer: how much benefit is needed? *J Natl Cancer Inst Monogr* 2001; : 146–152.
137 Greenberg R, Itah R, Ghinea R, Sacham-Shmueli E, Inbar R, Avital S. Metastatic lymph node ratio (LNR) as a prognostic variable in colorectal cancer patients undergoing laparoscopic resection. *Tech Coloproctology* 2011; 15: 273–279.
138 Møller. LMD Inger Kristin Larsen, Kristin Oterholt Knudsen, Stein Aaserud, Barthold Vonen og Bjørn. *Evaluering av datakvaliteten i Nasjonalt kvalitetsregister for tykk- og endetarmskreft*. Kreftregistret: Oslo, Norway, 2017.
139 Gately L, Jalali A, Semira C, Faragher I, Croxford M, Ananda S *et al.* Stage dependent recurrence patterns and post-recurrence outcomes in non-metastatic colon cancer. *Acta Oncol* 2021; : 1–8.
140 Iversen LH, Green A, Ingeholm P, Østerlind K, Gögenur I. Improved survival of colorectal cancer in Denmark during 2001–2012 – The efforts of several national initiatives. *Acta Oncol* 2016; 55: 10–23.
141 Rutegård M, Kverneng Hultberg D, Angenete E, Lydrup M-L. Substantial underreporting of anastomotic leakage after anterior resection for rectal cancer in the Swedish Colorectal Cancer Registry. *Acta Oncol* 2017; 56: 1741–1745.
142 Kopetz S, Tabernero J, Rosenberg R, Jiang Z-Q, Moreno V, Bachleitner-Hofmann T *et al.* Genomic classifier ColoPrint predicts recurrence in stage II colorectal cancer patients more accurately than clinical factors. *The Oncologist* 2015; 20: 127–133.
143 Nozawa H, Ishihara S, Kawai K, Hata K, Kiyomatsu T, Tanaka T *et al.* A high preoperative carbohydrate antigen 19-9 level is a risk factor for recurrence in stage II colorectal cancer. *Acta Oncol* 2017; 56: 634–638.
144 Niedzwiecki D, Frankel WL, Venook AP, Ye X, Friedman PN, Goldberg RM *et al.* Association Between Results of a Gene Expression Signature Assay and Recurrence-Free Interval in Patients With Stage II Colon Cancer in Cancer and Leukemia Group B 9581 (Alliance). *J Clin Oncol* 2016; 34: 3047–3053.
145 Marcker Espersen ML, Linnemann D, Christensen IJ, Alamili M, Troelsen JT, Høgdall E. SOX9 expression predicts relapse of stage II colon cancer patients. *Hum Pathol* 2016; 52: 38–46.
146 Dalerba P, Sahoo D, Paik S, Guo X, Yothers G, Song N *et al.* CDX2 as a Prognostic Biomarker in Stage II and Stage III Colon Cancer. *N Engl J Med* 2016; 374: 211–222.
147 Sundström J, Byberg L, Gedeborg R, Michaëlsson K, Berglund L. Useful tests of usefulness of new risk factors: Tools for assessing reclassification and discrimination. *Scand J Public Health* 2011; 39: 439–441.
148 Tie J, Wang Y, Tomasetti C, Li L, Springer S, Kinde I *et al.* Circulating tumor DNA analysis detects minimal residual disease and predicts recurrence in patients with stage II colon cancer. *Sci Transl Med* 2016; 8: 346ra92.
149 Wang Y, Li L, Cohen JD, Kinde I, Ptak J, Popoli M *et al.* Prognostic Potential of Circulating Tumor DNA Measurement in Postoperative Surveillance of Nonmetastatic Colorectal Cancer. *JAMA Oncol* 2019; 5: 1118.

Acta Universitatis Upsaliensis

Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine 1756

Editor: The Dean of the Faculty of Medicine

A doctoral dissertation from the Faculty of Medicine, Uppsala University, is usually a summary of a number of papers. A few copies of the complete dissertation are kept at major Swedish research libraries, while the summary alone is distributed internationally through the series Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine. (Prior to January, 2005, the series was published under the title "Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine".)

Distribution: publications.uu.se
urn:nbn:se:uu:diva-437831

ACTA
UNIVERSITATIS
UPSALIENSIS
UPPSALA
2021