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# A population-based nationwide study on total colectomy for ulcerative colitis and risk of ten prevalent inflammatory or autoimmune diseases

**Short title: Total colectomy and risk of inflammatory or autoimmune disease**

Anders Mark-Christensen[a,b], Line Riis Jølving[c], Pavithra Laxsen Anru[c], Joseph A. Murray[d], Rasmus Gaardskær Nielsen[e], Niels Qvist[a], Søren Laurberg[b], Henriette Engberg[f], Jens Kjeldsen[g], Bente Mertz Nørgård[c]

Affiliations: [a]Research Unit for Surgery and Centre for IBD Care, Odense University Hospital, Odense, Denmark and University of Southern Denmark, Odense, Denmark, [b]Department of Surgery, Aarhus University Hospital, [c]Center for Clinical Epidemiology and Research unit of Clinical Epidemiology, Odense University Hospital, Department of Clinical Research, University of Southern Denmark, Odense, Denmark, [d] Division of Gastroenterology and Hepatology, The Mayo Clinic, Rochester, Minnesota, USA, [e]Department of Pediatrics, Odense University Hospital, [f] The Danish Clinical Quality Program and Clinical Registries (RKKP), [g]Department of Medical Gastroenterology, Odense University Hospital and Research Unit of Medical Gastroenterology, Department of Clinical Research, University of Southern Denmark.

Corresponding author:
Anders Mark-Christensen, MD, PhD
E-mail: andersmark@dadlnet.dk
Phone: +45 5123 6691

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## ABSTRACT

**BACKGROUND** There is growing evidence to support a role of the gut microbiome in the development of chronic inflammatory and autoimmune disease (IAD). We used total colectomy (TC) for ulcerative colitis (UC) as a model for a significant disruption in gut microbiome to explore an association with subsequent risk of inflammatory or autoimmune disease.

**METHODS** We identified all patients with UC and no diagnosis of IAD prior to their UC diagnosis in Denmark from 1988 to 2015. Patients were followed from the date of UC to a diagnosis of IAD, death or end of follow-up, whichever occurred first. We used Cox regression to estimate hazard ratios (HRs) of IAD associated with TC, adjusting for age, sex, Charlson Comorbidity Index, and calendar year of UC diagnosis.

**RESULTS** 30,507 patients with UC (3,155 with TC and 27,352 without) were identified from the Danish National Patient Registry. During 43,266 person-years of follow-up, 2733 patients were diagnosed with an IAD. The risk of any IAD was higher for patients with TC compared to patients without (adjusted HR [aHR] 1.39 (95% CI: 1.24-1.57)). When the analyses were adjusted for exposure to antibiotics, immunomodulatory medicine and biologics (covering 2005-2018), the risk of IAD was still higher for patients with total colectomy (aHR = 1.41 (95% CI: 1.09;1.83)). Disease-specific analyses were weakened by a low number of outcomes.

**CONCLUSIONS** The risk of IAD was higher for patients who underwent TC for UC compared to patients who did not.

## KEY MESSAGES

- **What is already known?**
    - The gut microbiome plays an important role in host immune homeostasis, and changes in gut bacterial diversity and composition may change the individual's risk of inflammatory or autoimmune disease.
- **What is new here?**
    - Patients with ulcerative colitis who undergo total colectomy have a higher risk of being diagnosed with inflammatory or autoimmune disease, compared to patients with ulcerative colitis who do not undergo total colectomy.
- **How can this study help patient care?**
    - Future research can help uncover the mechanisms responsible for the higher risk of certain inflammatory or autoimmune diseases after total colectomy. If the microbiome plays a role, modifying the gut microbiome could prove a viable therapeutic strategy to reduce the risk of developing inflammatory or autoimmune diseases.

## SUMMARY

In this nationwide Danish cohort study of all Danish UC patients diagnosed in the period from 1988 to 2015, the risk of being diagnosed with inflammatory or autoimmune disease is higher for patients who underwent total colectomy compared to UC patients without total colectomy.

## INTRODUCTION

It is increasingly recognized that the human gut microbiome plays an important role in health and disease,[1] and susceptibility to many chronic diseases such as inflammatory or autoimmune disease may to some extent be modified by the composition and function of the gut bacterial flora.[2] Dysbiosis, altered polysaccharide fermentation, chronic low-grade inflammation, and host immunological reactions[3,4] may be responsible for this increased risk of inflammatory or autoimmune diseases associated with modifications of the gut microbiome. Several studies have implied a role of the gut flora in the development of several inflammatory or autoimmune diseases that involve a dysregulated immune response, e.g., diabetes mellitus, rheumatoid arthritis and thyroid disease.[5-7]. The underlying mechanisms are complex and possibly unique to each disease. Molecular mimicry, where antibodies induced by Lactobacillus and Bifidobacterium species cross-react with thyroperoxidase and thyroglobulin, may in part explain the association between gut microbiome composition and risk of autoimmune thyroid disease.[5] Animal studies have found that mice housed under germ-free conditions have a reduced risk of autoimmune arthritis, and that gut microbiota mediates the induction of Th 17 cells in the lamina propria of the small intestine. These cells then migrate to the peripheral lymphoid tissues and secrete IL-17, which ultimately helps in the differentiation of B cells and production of autoantibodies that define autoimmune arthritis and leads to its phenotypic traits.[6] It therefore seems plausible that changes in the gut microbiome, could affect the risk of developing inflammatory and autoimmune disease.

Ulcerative colitis (UC) is a chronic inflammatory bowel disease that involves a dysregulated immune response to commensal gut bacteria in genetically susceptible individuals,[8] and 4-7% of patients with UC will require total colectomy for either medically refractory disease or colorectal neoplasia after two years, increasing to more than 25% at 20 years.[9,10] With this procedure, the greatest mass of gut bacteria is removed together with fermented byproducts such as free fatty acids important for immune homeostasis. On this background, total colectomy may modify the individual's risk of inflammatory and autoimmune diseases, but this hypothesis remains untested. Total colectomy is most frequently undertaken in patients with UC, thus providing an important opportunity to examine the potential role for the colonic flora on the risk for autoimmunity.

It is important to explore whether an association between UC and inflammatory or autoimmune disease exists, as this may contribute to our understanding of these diseases and ultimately have implications for both prevention and therapies for these diseases, including UC. The aim of this study was thus to examine if the risk of inflammatory or autoimmune disease is associated with a significant and severe reduction in gut microbiome mass, composition and diversity. We used total colectomy for UC as a model to test this hypothesis and defined an inflammatory or autoimmune disease as a disease with a relatively high prevalence in the general population and a suspected full or partial autoimmune etiology.

## METHODS

### Setting

This nationwide cohort study comprised all patients diagnosed with UC from 1988 to 2015 in Denmark from a source population of approximately 5.7 million citizens (>90% Caucasian) at any given time. At birth or emigration, all citizens in Denmark are provided with a unique personal identification number (the CPR number) issued by the Civil Registration System and allows valid record linkage of individual-level data between numerous health registries.[11] All in-hospital treatment in Denmark is tax-funded, while out-patient medical treatment may include a small part self-payment and government reimbursements.

### Identification of study population

From the Danish National Patient Registry (DNPR),[12] we identified individuals aged ≥ 12 years recorded with a diagnosis of UC (ICD-8: 563.19, 569.04 and ICD-10: K51) in the period from January 1, 1988 to December 31, 2015. To increase the specificity of the UC diagnosis, we only included individuals with at least two UC discharge diagnosis in the DNPR, or at least three outpatient visits with a UC diagnosis after a first recorded UC diagnosis.[13]

The DNPR is a nationwide health registry that was established in 1977 and holds data on all in-patient hospital contacts made in Denmark since this inception year, and on out-patient treatments since 1995.[12] Each contact contains information on discharge diagnoses based on the International Classification of Diseases (ICD) version 8 from 1977

to 1993, and the ICD-10 version from 1994 onwards. Within the study population, individuals with UC and total colectomy were identified by the surgical procedure codes: 45020, 45060, 45061, 45065, 45080, 45081, 45840, 45880, KJFH00, and KJFH01. Procedures were coded using The Danish Classification of Surgical Procedures until 1997, where the Nordic Medico-Statistical Committee Classification of Surgical Procedures replaced it. All contact to the Danish healthcare system is linked to the individual by their CPR number.

**Definition of outcomes**

As there is no international consensus on how to define an inflammatory or autoimmune disease based on ICD coding, we chose an operational definition, where disease with an established full or partial autoimmune etiology were defined as an inflammatory or autoimmune disease. We considered a patient to have a relevant inflammatory or autoimmune disease if any of the following diagnoses were recorded in the DNPR during follow-up: Type 1 diabetes (ICD-8: 249; ICD-10: E10) and type 2 diabetes (ICD-8: 250; ICD-10: E11), celiac disease (ICD-8: 269.00, 269.98; ICD-10: K900), multiple sclerosis (ICD-8: 340; ICD-10: G35), thyroid disease (ICD-8: 242.00, 245.03; ICD-10: E050, E063), seropositive rheumatoid arthritis (ICD-8: 712.19, 712.39, 712.59; ICD-10: M05, M06), psoriasis vulgaris (ICD-8: 696.09, 696.10, 696.19; ICD-10: L40 (excluding L404)), ankylosing spondylitis (ICD-8: 712.49; ICD-10: M459), systemic lupus erythematosus (ICD-8: 734.19; ICD-10: M321, M329), and polymyalgia rheumatica (ICD-8: 446.30, 446.31, 446.39; ICD-10: M315, M316, M353).

**Covariates and confounders**

Potential confounders were defined *a priori* and included: age at the time of UC diagnosis, sex, Charlson Comorbidity Index[14] (calculated using diagnoses from the DNPR registered within 10 years prior to UC diagnosis), calendar year of UC diagnosis, exposure to azathioprine/6-mercaptopurine (Anatomical Therapeutic Chemical [ATC] Classification System codes L04AX01, and L01BB02, with data available from 1995 onwards), antibiotics (ATC codes D06BX01, J01MA02, J01DC02, and J01CR05, available from 1995 onwards), and biological therapy (procedure codes BOHJ18A1, BOHJ18A3, BOHJ18A4, BOHJ19H4, with data available from 2005 onwards). Information on biological therapy was

extracted from the DNPR, whereas the remaining medication data on prescriptions of azathioprine and 6-mercaptopurine was extracted from The Danish National Prescription Register (NPR).[15,16] Data on out-patient drug prescriptions were collected from the nationwide prescription database maintained by the Danish Medicine Agency. All pharmacies in Denmark are equipped with a computerized accounting system, which sends data on prescriptions to the nationwide prescription database. The data transferred to the prescription database include the CPR number, the type of drug prescribed, and the date of filed prescription.

**Statistical analysis**

Characteristics were given for the main study variables according to colectomy status as exposed and unexposed cohorts. A Chi-squared test was used to test differences in categorical variables between the exposed and unexposed cohorts, and a p-value $< 0.05$ was considered statistically significant.

Patients with UC diagnosed between January 1 1988 and December 31 2015 were followed from their first UC diagnosis, where they entered the 'no colectomy' cohort. Patients from the 'no colectomy' cohort transitioned to the 'colectomy' cohort in the event of total colectomy. All patients were followed for a diagnosis of inflammatory or autoimmune disease, and were censored in the event of emigration, death or end of follow-up on December 31, 2018, whichever occurred first. Individuals with a diagnosis of inflammatory or autoimmune disease prior to the index date were excluded from the analyses.

The risk of developing inflammatory or autoimmune disease during follow-up was illustrated graphically as cumulative incidences using death as a competing risk. To reduce the risk of introducing a detection bias where patients hospitalized after total colectomy were coincidentally diagnosed with inflammatory or autoimmune diseases, we excluded outcome events that occurred within 1 month after total colectomy. Also, due to statistical power considerations, the HRs were not calculated if the number of outcomes in those having total colectomy were less than 10.

The association between total colectomy and incident inflammatory or autoimmune disease was estimated using a stratified Cox proportional hazards regression model of the hazard ratios (HRs) of inflammatory or autoimmune disease, comparing

patients with UC and total colectomy to patients with UC without total colectomy. We used i) a composite outcome of any inflammatory or autoimmune diseases and ii) each type of disease as separate outcomes. Crude and adjusted HRs were calculated, adjusting for age, sex, Charlson Comorbidity Index and calendar year of diagnosis in model 1. Data on medications were only available for a restricted time period from 1995 to 2018.

The use of antibiotics was added as confounder in model 2, and exposure to immunomodulatory medicine and biological medicine was added as confounder in model 3. Patients with no use of antibiotics, immunomodulatory medicine or biologics were followed from their first UC diagnosis, where they entered the group of patients with *no prescriptions* in the analysis. Patients from this group transitioned during follow-up to the group of patients with *prescriptions* if the patient had received either antibiotics, immunomodulatory medicine, or biologics in the time period from the index date to either an outcome of interest, death or end of follow-up. In order to explore if exposure to medication could be considered a pertinent confounder, we looked at the proportion of patients with an outcome of interest, who were exposed to antibiotics, immunomodulatory medicine and biological medicine in the time period from 6 months before the first occurrence of any of the 10 listed inflammatory or autoimmune diseases after the time of UC diagnosis, hence follow-up.

In a sensitivity analysis, we excluded any outcomes diagnosed within 6 months after total colectomy and in a second sensitivity analysis, we restricted the definition of autoimmune disease to only include cases with at least two hospital diagnoses with the outcome disease of interest. In these sensitivity analyses, we adjusted for age, sex, Charlson Comorbidity Index and calendar year of diagnosis.

The assumption of proportional hazards was evaluated graphically on log-log plots and found to be satisfied in all analyses.

Stata V. 15.0 software (StataCorp LP, College Station, TX, USA) was used in all analyses.

**Ethical considerations**

This study was approved by the Danish Data Protection Agency under the current joint notification of the Region of Southern Denmark (journal number 17/34575). According to Danish law, ethical review board approval or individual patient consent are not required for register-based studies.

## Data availability

According to Danish legislation, our approvals to use these register data for the current study do not allow us to make patient data available to other parties. Any interested researchers may apply for access to data through an application to the Research Service at the Danish Health Data Authority (Forskerservice). Also, access to data from the Danish Health Data Authority requires approval from the Danish Data Protection Agency.

## RESULTS

We identified 30,507 patients with UC, of whom 3,155 underwent total colectomy, and 27,352 who did not undergo total colectomy during follow-up. Baseline characteristics are listed in Table 1. Patients who underwent total colectomy were typically younger and had fewer comorbidities than those who did not undergo total colectomy.

In both the 'no colectomy' and 'colectomy' cohorts, few individuals had received treatment with immunomodulatory medicine, antibiotics, or biological therapy six months prior to an outcome event. The median time of follow-up was 15.03 years in patients with total colectomy (25-75% percentiles: 8.62-21.77), and the total years of follow-up was 43,266 years for outcomes in the category of any autoimmune disease. Among patients without total colectomy, the median time of follow-up was 10.16 years (25-75% percentiles: 5.80-16.94), and the total follow-up was 290,828 years.

During follow-up, 325 (11.4%) patients with total colectomy were diagnosed with any of the prespecified diseases, compared to 2408 (9.4%) patients who did not undergo total colectomy in the observation period (Figure 1). Crude and adjusted HR's for the association between total colectomy and risk of developing inflammatory or autoimmune diseases are presented in Table 2 with the corresponding number of events in each group. In the adjusted analyses, the overall risk of any inflammatory or autoimmune disease was higher for patients with UC and total colectomy compared to patients with UC without total colectomy (aHR = 1.39 (95% CI: 1.24;1.57)). This increased risk was primarily driven by an increased risk of thyroid disease (aHR = 1.93 (95% CI: 1.38;2.69)), psoriasis vulgaris (HR = 1.67 (95% CI: 1.17;2.38), and ankylosing spondylitis (HR = 2.23 (95% CI: 1.43;3.47)).

When restricting the analyses to the time period where we had information on medication (1995-2018 and 2005-2018) the number of outcome events decreased. These results are presented in Table 3. The overall risk of developing any inflammatory or autoimmune disease (aHR = 1.34 (95% CI: 1.16;156) and aHR = 1.41 (95% CI: 1.09;1.83)) remained virtually unchanged for patients with UC and total colectomy, compared to our main result (aHR = 1.39 (95% CI: 1.24;1.57)).

In a sensitivity analysis excluding outcomes diagnosed within 6 months after total colectomy, there was still a positive association between total colectomy and risk of any inflammatory or autoimmune disease (aHR = 1.30 (95% CI: 1.15-1.47)).

When we restricted the analyses to only include outcomes with at least two diagnoses, the estimates were unchanged with an aHR of 1.31 (95% CI, 1.12-1.53) of any disease.

## DISCUSSION

With this study, we found that the risk of being diagnosed with an inflammatory or autoimmune disease was higher for patients with UC who underwent total colectomy compared to those who did not, indicating a potential role of the colonic flora in protecting the host from the development of autoimmune disease.

The association between total colectomy for UC and risk for inflammatory or autoimmune disease differed according to the type of autoimmune disease investigated; the risk was most pronounced for the development of type 2 diabetes mellitus, thyroid disease, psoriasis and ankylosing spondylitis and less for the development of type 1 diabetes mellitus, seropositive rheumatoid arthritis, multiple sclerosis and polymyalgia rheumatica, where the associations were statistically insignificant when adjusting for several potential confounders, albeit overall suggesting an increased risk associated with total colectomy. Differences in the magnitude of the association between total colectomy and risk of developing different types of inflammatory or autoimmune disease may reflect the multifactorial nature of these diseases, where the contribution of gut bacterial composition to the etiology of the diseases may differ.[17]

A total colectomy involves excision of the colon and terminal segment of the ileum, thus effectively removing the largest source of commensal bacteria in the human body.[18] The gut bacteria are an integral part of the epithelial barrier function that prevents pathogenic bacteria from infecting the host so the changes in diversity and composition of the gut flora that follows total colectomy,[19] together with small bowel bacterial overgrowth,[20] makes it theoretically plausible that a total colectomy also changes the individuals' risk of some diseases with an etiology that may in part be influenced by gut dysbiosis.

The underlying mechanisms by which the colonic microflora may affect the risk of inflammatory or autoimmune disease is largely unknown; molecular mimicry, where antibodies directed against specific bacterial antigens cross-reacts with host proteins to elicit an inflammatory reaction offers a potential explanation, but translational studies are scarce[6] and, to the best of our knowledge, there are no prospective epidemiological studies exploring an association between changes in colonic microbiome on the risk of inflammatory or autoimmune disease. Although our study supports a hypothesis of a crucial role of the gut microbiota in the development of such diseases, the underlying mechanisms cannot be elucidated from our study, and large, prospective studies are needed to examine biological causality.

The strengths of this study lie in the nationwide capture of all patients with UC and total colectomy, limiting selection bias, and in the use of national health registries with a documented high accuracy. The long follow-up periods of almost 15 years and 10 years for patients with and without total colectomy for UC is also unique and only possible due to the nationwide Danish health registries. Because we used Cox proportional hazards regression analysis, the differences in follow-up for UC patients with and without total colectomy were accounted for when calculating hazard ratios. The observation that our sensitivity analyses largely confirmed our main findings also add power to the study and confidence in the validity of the diagnoses.

While we were able to adjust for many potentially confounding factors in our study, e.g., sex, age, comorbidities and exposure to different types of medicine, the possibility of residual confounding remains. For example, patients who undergo total colectomy may be in more frequent contact with the health care system compared to patients with UC who do not undergo total colectomy and thus have quiescent and asymptomatic inflammatory or

autoimmune disease diagnosed. This would lead to an apparent higher risk for autoimmune disease compared to patients with UC without total colectomy, especially in the immediate postoperative period, where the majority of complications typically occur. However, the cumulative incidence of autoimmune disease after total colectomy for UC remained relatively constant over time, arguing against a substantial bias.
It is important to note, that although median follow-up was relatively long at approximately 15 and 10 years for patients with and without total colectomy, the risk of inflammatory or autoimmune disease extending beyond this period is uncertain at best, and the long-term risk of being diagnosed with these diseases should be explored further in future studies.

Patients who undergo total colectomy (especially with ileal pouch-anal anastomosis) for UC may have superimposed bacterial infections,[21] for which they receive antibiotics, that can disrupt the gut microbiome and potentially affect the risk of developing inflammatory or autoimmune disease. Therefore, we performed analyses adjusted for use of the most common types of antibiotics used for gastrointestinal infections in Denmark (metronidazole, quinolones, ceforuxime, piperacillin and beta-lactamase inhibitors). Still, we found that the risk of any inflammatory or autoimmune disease was increased for patients who underwent total colectomy, indicating that differences in exposure to antibiotics did not materially confound the estimates. As information on antibiotic dispensings were only available from 1995 onward, the adjusted estimates were not as robust as our main analyses, and this may explain the differences in our estimates, that all nonetheless suggested an increased risk associated with total colectomy. In-hospital treatment with intravenous steroids in cases of severe flares of UC is not registered in the Danish registries. Therefore, we could not ascertain the impact of steroid exposure as a confounder for the association with inflammatory or autoimmune disease, where especially the risk of type 2 diabetes mellitus could be related to steroid exposure in the immediate period after total colectomy, until patients are weaned off them.

As patients who undergo total colectomy typically have more severe disease than those who do not undergo total colectomy, our finding may also reflect an association between severe UC rather than total colectomy and risk of inflammatory or autoimmune disease,[22] and this is the most important limitation to our study. However, data contained in the DNPR are not sufficiently detailed to adequately determine disease severity. When we

adjusted for exposure to immunomodulatory medicine and biologics as a proxy for disease severity, the association was virtually unchanged. For the specific types of inflammatory or autoimmune disease, the few outcomes with loss of statistical power hindered meaningful comparative analyses. However, the very low number of patients exposed to either antibiotics, biologics, or immunomodulatory medicine 6 months prior to a diagnosis of inflammatory or autoimmune disease in our view argues against a biologically plausible association with disease severity or exposure to immunomodulatory drugs per se. Another important limitation is the imprecision of several of the associations, especially when adjusting for exposure to medicine, where many estimates included unity. Differences in the magnitude of the association between total colectomy and risk of developing different types of inflammatory or autoimmune disease may reflect the multifactorial nature of these diseases, where the contribution of gut bacterial composition to the etiology of the diseases may differ.[17] It may however also reflect that the associations are chance findings.

In conclusion, we found that patients who underwent total colectomy for UC had a higher overall risk of being diagnosed with inflammatory or autoimmune disease compared to patients with UC who did not undergo total colectomy. Further studies are needed to investigate a biologically causal relationship between total colectomy for UC and risk of inflammatory or autoimmune disease, and similar studies performed on other nationwide cohorts would also be of great value. Experimental studies in animal models of total colectomy could be undertaken to determine if the colonic flora or its absence, as suggested by the surgical experiment of colonic microbiomectomy, plays an essential role in immune homeostasis.

## TABLES

**Table 1: Baseline characteristics of the study population of patients diagnosed with ulcerative colitis (UC) from 1988 to 2015 (N=30,507)**

| | UC patients with colectomy N= 3,155 | UC patients without colectomy N=27,352 |
|---|---|---|
| **Duration of disease in years from time of UC diagnosis to end of follow-up (IQR)** | 15.03 (8.62-21.77) | 10.16 (5.80-16.94) |
| | | |
| **Age at time of UC diagnosis in years (mean (SD))** | 40.30 (18.29) | 46.44 (19.05) |
| | | |

| | | |
|---|---|---|
| **Age at time of UC diagnosis, N (%)** | | |
| **12-19** | 406 (12.9) | 1701 (6.2) |
| **20-39** | 1343 (42.6) | 10001 (36.6) |
| **40-59** | 825 (26.1) | 8308 (30.4) |
| **60-79** | 540 (17.1) | 6139 (22.4) |
| **>=80** | 41 (1.3) | 1203 (4.4) |
| | | |
| **Sex, N (%)** | | |
| **Female** | 1479 (46.9) | 14268 (52.2) |
| **Men** | 1676 (53.1) | 13084 (47.8) |
| | | |
| **Charlson comorbidity index (CCI)*, N (%)** | | |
| **0** | 2732 (86.6) | 21941 (80.2) |
| **1** | 242 (7.7) | 2871 (10.5) |
| **>=2** | 181 (5.7) | 2540 (9.3) |
| | | |
| **Year of UC diagnosis, N (%)** | | |
| **1988-1994** | 822 (26.1) | 3137 (11.5) |
| **1995-1999** | 657 (20.8) | 4547 (16.6) |
| **2000-2004** | 636 (20.2) | 5093 (18.6) |
| **2005-2009** | 552 (17.5) | 6269 (22.9) |
| **2010-2015** | 488 (15.5) | 8306 (30.4) |
| | | |
| **Year of colectomy, N (%)** | | |
| **1988-1994** | 470 (14.9) | - |
| **1995-1999** | 505 (16.0) | - |
| **2000-2004** | 594 (18.8) | - |
| **2005-2009** | 691 (21.9) | - |
| **2010-2015** | 895 (28.4) | - |
| | | |
| **Use of azathioprine/6-mercaptopurine within 6 months before autoimmune disease (outcome) #, N (%)** | | |
| **No** | 204 (94.9) | 1907 (96.8) |
| **Yes** | 11 (5.1) | 63 (3.2) |
| | | |
| **Use of antibiotics within 6 months before autoimmune disease (outcome) ‡, N (%)** | | |
| **No** | 201 (93.5) | 1909 (96.9) |
| **Yes** | 14 (6.5) | 61 (3.1) |
| | | |
| **Use of biological therapy within 6 months before autoimmune disease (outcome)¤, N (%)** | | |
| **No** | 202 (94.0) | 1927 (97.8) |
| **Yes** | 13 (6.1) | 43 (2.2) |
| | | |

**Notes**: * The Charlson Comorbidity Index was calculated based on hospital discharge diagnoses during a ten year period before baseline. #The use of azathioprine/6-mercaptopurine was based on the following ATC-codes: L04AX01 (AZA) and L01BB02 (6-mercaptopurine). ‡The use of antibiotics was based on the following ATC-codes: D06BX01, J01MA02, J01DC02, J01CR05. ¤The use of biological therapy was based on the procedure codes: BOHJ18A1, BOHJ18A3, BOHJ19H4, BOHJ18A4. For #, ‡ and ¤ a "yes" indicates at least one redeemed prescription/ at least one procedure code after UC diagnosis for the unexposed cohort or after the date of colectomy for the exposed cohort within 6 months before the first occurrence of any of the 10 listed autoimmune disease. £The pouch operations were defined from the following procedure codes: KJFH30, KJFH31, KJFH33, KJFH34, KJGB50, KJGB60, KJGB61 from 1996-2015.

Data on azathioprine/6-mercaptopurine and antibiotics were available in the registries from 1995-2018, and data on biological therapy were available from 2005-2018.

**Table 2: Cox regression analyses (crude and adjusted) - exclusion of outcomes within 30 days after colectomy.**

*Note: Due to lack of sufficient statistical power the number of observations less than 10 are not reported.*

| Autoimmune disease | UC patients with colectomy N=3,155 Number of events, (%) | UC patients without colectomy N=27,352 Number of events, (%) | Crude HR (95% CI) | Adjusted (model 1)* HR (95% CI) |
|---|---|---|---|---|
| Autoimmune disease# | 325 (11.37) | 2408 (9.39) | 1.19 (1.06,1.34) | 1.39 (1.24,1.57) |
| Diabetes type 1 | 62 (2.06) | 577 (2.13) | 1.13 (0.87,1.48) | 1.30 (0.99,1.70) |
| Diabetes type 2 | 161 (5.40) | 1369 (5.14) | 1.08 (0.91,1.27) | 1.29 (1.09,1.52) |
| Multiple sclerosis | 10 (0.32) | 72 (0.26) | 1.23 (0.63,2.40) | 1.24 (0.63,2.43) |
| Thyroid diseases | 44 (1.43) | 256 (0.94) | 1.74 (1.26,2.42) | 1.93 (1.38,2.69) |
| Seropositive rheumatoid arthritis | 41 (1.33) | 319 (1.18) | 1.17 (0.84,1.63) | 1.36 (0.97,1.90) |
| Psoriasis vulgaris | 38 (1.24) | 224 (0.82) | 1.52 (1.07,2.16) | 1.67 (1.17,2.38) |
| Ankylosing spondylitis | 26 (0.84) | 113 (0.41) | 2.18 (1.41,3.38) | 2.23 (1.43,3.47) |
| Systemic lupus erythematosus | - | 32 (0.12) | - | - |
| Polymyalgia rheumatic | 16 (0.52) | 202 (0.74) | 0.66 (0.39,1.10) | 0.92 (0.55,1.54) |
| Celiac disease | - | 67 (0.25) | - | - |

# First occurrence of any of the 10 listed autoimmune diseases.

* Model 1: Adjusted for age at time of UC diagnosis, sex, Charlson comorbidity index and year of UC diagnose (all the adjustment variables are categorical).

**Table 3: Cox regression analyses (crude and adjusted) - exclusion of outcomes within 30 days after colectomy.**

*Note: Due to lack of sufficient statistical power the number of observations less than 10 are not reported.*

| Autoimmune disease | UC patients with colectomy N=3,155 Number of events, (%) | UC patients without colectomy N=27,352 Number of events, (%) | Adjusted (model 2)¤ HR (95% CI) | UC patients with colectomy N=3,155 Number of events, (%) | UC patients without colectomy N=27,352 Number of events, (%) | Adjusted (model 3)# HR (95% CI) |
|---|---|---|---|---|---|---|
| Autoimmune disease# | 215 (10.18) | 1,970 (8.71) | 1.34 (1.16,1.56) | 71 (7.67) | 865 (6.42) | 1.41 (1.09,1.83) |
| Diabetes type 1 | 36 (1.61) | 465 (1.95) | 1.18 (0.83,1.68) | 13 (1.31) | 199 (1.39) | 1.56 (0.86,2.82) |
| Diabetes type 2 | 100 (4.52) | 1,131 (4.80) | 1.19 (0.96,1.47) | 39 (3.96) | 492 (3.49) | 1.64 (1.16,2.32) |
| Multiple sclerosis | - | 60 (0.25) | - | - | 26 (0.18) | - |
| Thyroid diseases | 30 (1.31) | 194 (0.81) | 2.31 (1.53,3.49) | - | 80 (0.55) | - |
| Seropositive rheumatoid arthritis | 27 (1.18) | 255 (1.06) | 1.35 (0.89,2.05) | - | 113 (0.78) | - |
| Psoriasis vulgaris | 28 (1.23) | 187 (0.78) | 1.66(1.09,2.54) | 10 (0.98) | 96 (0.66) | 0.99 (0.50,1.99) |
| Ankylosing spondylitis | 18 (0.78) | 102 (0.42) | 1.78 (1.04,3.03) | - | 46 (0.32) | - |
| Systemic lupus erythematosus | - | 27 (0.11) | - | - | 12 (0.08) | - |
| Polymyalgia rheumatic | 12 (0.52) | 169 (0.70) | 0.94 (0.52,1.72) | - | 77 (0.53) | - |
| Celiac disease | - | 58 (0.24) | - | - | 23 (0.16) | - |

# First occurrence of any of the 10 listed autoimmune diseases.

¤ Model 2: Adjusted for age at time of UC diagnosis, sex, Charlson comorbidity index, year of UC diagnosis, and the use of antibiotics at any time after UC diagnosis (unexposed) or date of colectomy (exposed (all the adjustment variables are categorical).

# Model 3: Adjusted for age at time of UC diagnosis, sex, Charlson comorbidity index, year of UC diagnosis, the use of azathioprine/6-mercaptopurin at any time after UC diagnosis (unexposed) or date of colectomy (exposed), the use of antibiotics at any time after UC diagnosis (unexposed) or date of colectomy (exposed) and the use of biological therapy at any time after UC diagnosis (unexposed) or date of colectomy (exposed).

Data on azathioprine/6-mercaptopurine and antibiotics were available in the registries from 1995-2018, and data on biological therapy were available from 2005-2018.

## Figure legends:

Figure 1. Cumulative incidence of any autoimmune disease after index date (total colectomy or corresponding matching date) for patients with UC in Denmark, 1988-2015.

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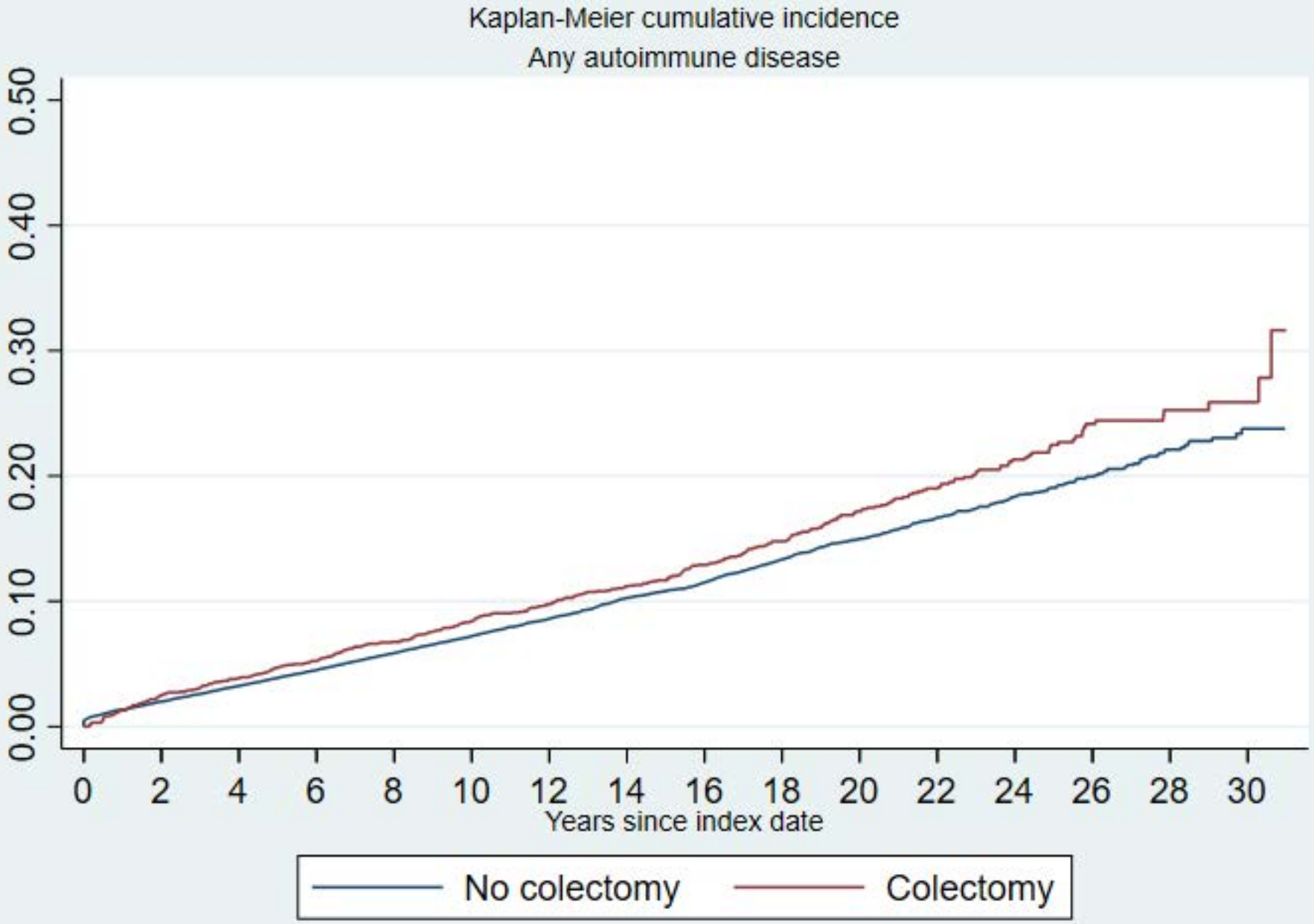
Kaplan-Meier cumulative incidence
Any autoimmune disease
0.00
0.10
0.20
0.30
0.40
0.50
0
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
Years since index date
No colectomy
Colectomy