# Association between early life exposure to agriculture, biodiversity, and green space and risk of inflammatory bowel disease: a population-based cohort study

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## Summary

**Background** Specific pollutants and environmental exposures are implicated in modulating inflammatory bowel disease (IBD) risk. However, the role of environmental exposures, particularly during the early life period, towards IBD risk, has not been systematically evaluated.

**Methods** We conducted a nationwide population-based cohort study during the study period extending from January 1, 1995, to September 1, 2020, using cross-linked Danish registers, maps, and inventories to ascertain the impact of agricultural land use, biodiversity, green space, urban space, blue space, and normalized difference vegetation index during pregnancy and the first two years of life on IBD, Crohn's disease (CD), and ulcerative colitis (UC) risk, using adjusted Cox proportional hazards regression analyses. We adjusted for covariates sex, maternal age at delivery, calendar year of birth, municipal-level socioeconomic status, and first-degree relative with IBD.

**Findings** Of 1,438,487 individuals included in the study who were followed from age 2 years until a median (IQR) age of 14 (8–20) years, 3768 individuals were diagnosed with IBD. Exposure to the second, third and highest quartiles of agriculture land use during early life, relative to the lowest quartile, were associated with increased CD risk (aHR 1.12, 95% CI 1.01, 1.26, 1.19, 95% CI 1.05, 1.34 and, 1.24 95% CI 1.06, 1.46, respectively). There was no association of agriculture land use with UC risk. Conversely, exposure to the third quartile of biodiversity in early life, compared to the lowest quartile, were associated with a lower CD risk (aHR 0.86, 95% CI 0.75, 0.98). A protective effect of greenspace was noted in the highest quartile for CD (aHR 0.87, 95% CI 0.78, 0.98).

**Interpretation** In a nationwide cohort with long-term follow up data, early life environmental exposures were associated with modulation of CD risk, with a harmful effect of agriculture land use and protective effect of biodiversity and green space.

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**Keywords:** Crohn's disease; Epidemiology; Land use; Natural environment; Ulcerative colitis; Urban space

## Introduction

Inflammatory bowel disease (IBD), including subtypes Crohn's disease (CD) and ulcerative colitis (UC), is on the rise globally.[1,2] It is associated with substantial burden at the individual, healthcare system, and societal levels, and yet, IBD risk factors and underlying mechanisms remain elusive.[3,4] The emergence of IBD in the west in the early 1900s, and in recently industrialized countries in the late 1900s–2000s, has occurred in parallel with urbanization and loss of the natural environment.[5] Previous epidemiological studies have reported a protective effect of green space and blue space against IBD.[6,7] Clustering of CD in agricultural areas was reported in an ecologic analysis.[8] Sanmarco et al. have

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## Research in context

**Evidence before this study**

We searched PubMed and Embase for studies published from databases inception to November 16, 2023, that evaluated the association between environmental exposures and inflammatory bowel disease (IBD). We used the following search terms: "green space" OR "blue space" OR "biodiversity" OR "agriculture" OR "natural vegetation density index" OR "urban space" AND "inflammatory bowel disease" OR "Crohn's disease" OR "ulcerative colitis". We identified 296 studies through this search strategy. Of these, only a few studies reported that exposure to green space and blue space was protective against IBD, while clustering of Crohn's disease (CD) was reported in agricultural areas in an ecologic analysis. No study systematically evaluated the impact of environmental exposures during the early life period on IBD risk.

**Added value of this study**

To our knowledge, this is the first analysis leveraging a population-based birth cohort, cross-linked with national registers, maps, and inventories to report on the association between environmental exposures during the early life period and IBD risk. We found that higher exposure to agriculture land use was associated with increased CD risk, while exposure to higher biodiversity and green space was protective against CD. There was no clear impact of early life environmental exposures on ulcerative colitis risk.

**Implications of all the available evidence**

These data suggest that early life environmental exposures were associated with modulation of CD risk, with a harmful effect of agriculture land use and protective effect of biodiversity and green space.

reported on -cidal compounds leading to intestinal inflammation in animal models.[9] While these data provide important insights, the impact of relevant environmental exposures, such as agriculture land use and biodiversity during the early life period on IBD risk, remains unexplored. The biodiversity hypothesis suggests that exposure to natural environments enriches the human microbiome, supports immune health, and mitigates inflammatory diseases.[10,11] Considering the rapid decline of the natural environment due to the climate and biodiversity crises, there is an urgent need to understand the role of natural environmental exposures in health and disease.[12]

Further, the timing of the exposure is an important consideration. The early life period, defined as extending from the prenatal period to early childhood, is a critical window during which microbiome establishment and immune maturation occur; exposures during this period play a key role in modulating health and disease later in life.[13,14]

Therefore, we conducted a population-based cohort study to understand the role of early life environmental exposures on IBD risk.

# Methods

## Study design and participants

We conducted a nationwide cohort study for which the source population included all individuals born in Denmark during the period extending from January 1, 1995, to August 1, 2018, who were residents of Denmark for at least the first two years of life, and whose mothers were residents of Denmark during pregnancy. Study participants were identified using the Danish Civil Registration System (CRS), a register that prospectively records demographic, vital data, municipality of residence and linkage with first degree relatives for all residents of Denmark with continuous updates. The CRS is cross-linked to other registries through a unique personal identification number.

## Exposures

The following six environmental variables were considered as exposures of interest: agricultural land use, biodiversity, green space, normalized difference vegetation index (NDVI), blue space and urban space. We aggregated each exposure variable to the municipal level. Each offspring was assigned the exposure values for their municipality of residence during the exposure window extending from pregnancy through the first two years of life. Where an exposure value was measured at multiple time points, we assigned the latest value measured before the start of the exposure window. If a measurement was updated during the exposure window, we assigned the weighted average of the measurements before and after updating, weighing by the proportion of the exposure window spent before and after the update point (for example, for a participant whose exposure value was updated halfway through the exposure window, we assigned the mean of the value before and after the update). In the case of individuals changing municipality of residence during the exposure window (as was the case for 18% of subjects), we assigned the weighted average for the municipalities (for example, a participant spending 40% of their exposure window in municipality A and 60% in municipality B would be assigned the exposure value $0.4 \cdot \mathrm{Exp}_A + 0.6 \cdot \mathrm{Exp}_B$ where $\mathrm{Exp}_A$ and $\mathrm{Exp}_B$ are the exposure scores for municipality A and B).

Our data source for land cover is the CORINE Land Cover Inventory which uses a minimum mapping unit of 25 ha and 44 classes of land cover.[15] We categorized each as green space, urban space, agricultural land, or water bodies which were excluded from the municipal total (Supplementary Table S1). CORINE data has updates from 1990, 2000, 2006, 2012 and 2018.

Agricultural land use was measured as the proportion of the municipal land cover (discounting bodies of water) categorized as agricultural areas. Green space was defined as the proportion of land cover categorized as forest, natural green areas or urban green space. Blue space was defined as the proportion of the municipal area within 600 m distance of a body of water, corresponding to a-10-min walking distance.[16] Urban space was defined as the proportion of the municipal area covered by urban areas excluding green urban areas (such as parks, zoos and public gardens, but not private gardens or allotment gardens). By construction, urban, agricultural, and green space are proportions of municipal land use, and these sum to one, while blue space (defined as the proportion of the municipal area within walking distance of a body of water) may occur with any type of land use.

The NDVI, an indicator of vegetation health and density, is derived by comparing the reflectance of light in the near-infrared and red spectra. The index ranges from −1 to 1, with higher values indicating denser vegetation. As such, higher NDVI scores may indicate fields as well as green areas. We used the data organized and described by Engemann et al., computing NDVI values for Denmark in the peak growing season of May–July for the years 1990, 1998, 1999 to 2007, 2009, 2010, 2013 to 2015.[17] We computed NDVI scores as the mean normalized difference vegetation index for each municipality.

Biodiversity was defined by the bioscore, a measure of diversity of local flora and fauna, based on the incidence of red-listed species as well as indicators of rare habitat conditions, using the Danish Biodiversity Map, available for the year 2018.[18,19]

We examined temporal municipal-level trajectories for exposure variables that varied over time and found that green space, urban space, and agriculture land use remained relatively stable over the study period. There were periodic variations in NDVI, which are likely to be due to lower rainfall and dry vegetation (Supplementary Figure S1).

## Outcomes

Using the cross-linked Danish National Patient Registry, we identified all study participants who were diagnosed with Crohn's disease (CD) or ulcerative colitis (UC) based on a previously-used definition.[2]

## Covariates

We included the following covariates *a priori* in all models as these could be potential confounding variables: sex (female, male), maternal age at delivery (continuous), calendar year of birth, first-degree relative with IBD prior to the birth of the child (yes, no), and two municipal-level indicators of socioeconomic status: The proportion of the population aged 25–64 who have only a basic level (10 years) of education, and a socioeconomic index measuring relative municipal social spending needs.[20] The latter index is computed at the municipal level and encompasses items such as the burden of unemployment, the number of low-income citizens and citizens without education, the number of children in low-education families and the number of disabled and mentally ill citizens. Information on the other covariates were obtained from the cross-linked Danish National Patient Registry.

## Missing data

Observations missing exposure data were excluded from the analyses. For the covariates, rates of missingness (n = 138, 0%) were low enough that we handled missingness by imputing the mean value of the covariate.

## Statistical analyses

All individuals were followed from the age of 2 years, which is the end of the exposure period, until IBD diagnosis, emigration, death, or September 1, 2020, whichever occurred first. We analyzed the association between each exposure and the primary outcome IBD and secondary outcomes CD and UC using Cox proportional hazards regression analysis, categorizing each exposure into quartiles, and using the lowest quartile as reference, adjusting for covariates. We accounted for the potential correlation at the municipal level by clustering on municipality at birth.

Finally, to evaluate the robustness of our findings, we conducted sensitivity analyses. We repeated main analyses using varying definitions of blue space (distance of up to 300 and 1200 m to a body of water) and urban space (treating urban green space as urban space). We determined the impact of exposure to each environmental variable by restricting each exposure duration to pregnancy only and after extending the early life period definition to up to 5 years. Finally, given the skewed distributions of each exposure variable we repeated main analyses to compare the highest decile of each exposure with the lowest quartile for outcomes IBD, CD, and UC.

The proportional hazards assumption was evaluated using Schoenfeld residuals and was found to hold. We used R version 4.3.0 and the survival package in the statistical analyses.[21]

## Ethics statement

Research based on register-based data does not require ethical permission or informed consent in Denmark.

## Role of the funding source

The funders of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report.

## Results

### Cohort characteristics

Of 1,510,045 individuals born in Denmark during the study period, we included 1,438,487 individuals in the study. Exclusions are described in Supplementary Figure S2. Of these, 3768 individuals were diagnosed with IBD during the follow up period. IBD in a first-degree relative was more prevalent in the IBD group, compared to the non-IBD group (4.7% vs 1.2%). Other baseline variables were comparable between the two groups, with the IBD group being born earlier, since the earliest born individuals had more time to develop IBD (Table 1).

The distribution of each exposure is demonstrated in Fig. 1A. Correlation across all exposure variables is demonstrated in Fig. 1B. Agriculture land use has a strong negative correlation with urban areas, while green space and biodiversity have a strong positive correlation.

### IBD risk across quartiles of early life environmental exposures

Individuals with and without IBD were followed from age 2 years until a median (interquartile range, IQR) age of 17 (14, 20) and 14 (8, 20) years, respectively. Kaplan Meier curves of the cumulative incidences are reported in Supplementary Figure S3. The risk of IBD among individuals exposed to the third quartile of agriculture land use during early life, compared to the lowest quartile, was increased (aHR 1.13, 95% CI 1.01, 1.26). The risk of IBD among those exposed to the third quartiles of biodiversity in early life, compared to the lowest quartile, was lower (aHR 0.90, 95% CI 0.81, 0.99). With respect to environmental exposures green space, urban space, blue space, and NDVI, null associations were found across quartiles of each exposure and IBD risk (Fig. 2).

### CD and UC risk across quartiles of early life environmental exposures

Next, we determined the impact of early life environment on CD and UC risk separately. The risk of CD among individuals exposed to the second, third and highest quartiles of agriculture land use, relative to the lowest quartile, was increased (aHR 1.12, 95% CI 1.01, 1.26, 1.19, 95% CI 1.05, 1.34 and, 1.24 95% CI 1.06, 1.46, respectively). The risk of CD among those exposed to the third quartile of biodiversity, compared to the lowest quartile, was lower (aHR 0.86, 95% CI 0.75, 0.98) and those exposed to the highest quartile of green space, compared to the lowest quartile, was lower (aHR 0.87, 95% CI 0.78, 0.98). There was no association across

| Characteristic | Developed IBD during follow-up period N = 3768 | No IBD during follow-up period N = 1,434,719 | P-value[a] |
|---|---|---|---|
| **Year of birth** | | | <0.001 |
| 1995–99 | 2347 (62%) | 323,971 (23%) | |
| 2000–09 | 1352 (36%) | 623,428 (43%) | |
| 2010–18 | 69 (1.8%) | 487,320 (34%) | |
| **Maternal age in years** | | | <0.001 |
| Under 30 | 1942 (52%) | 672,711 (47%) | |
| 30–34 | 1279 (34%) | 504,184 (35%) | |
| ≥35 | 547 (15%) | 257,686 (18%) | |
| Unknown | 0 | 138 (0%) | |
| **Sex** | | | <0.001 |
| Female | 1941 (52%) | 698,332 (49%) | |
| Male | 1827 (48%) | 736,387 (51%) | |
| **IBD in first-degree relative** | 176 (4.7%) | 17,810 (1.2%) | <0.001 |
| **Municipal-level education score** | 0.34 (0.28, 0.39) | 0.34 (0.28, 0.40) | <0.001 |
| **Municipal-level socioeconomic index** | 1.00 (0.92, 1.07) | 1.00 (0.94, 1.06) | 0.2 |
| **Environmental exposures** | | | |
| Agricultural space[b] | 0.73 (0.54, 0.79) | 0.71 (0.37, 0.79) | <0.001 |
| Biodiversity[c] | 1.08 (0.75, 1.39) | 1.19 (0.79, 1.47) | 0.002 |
| Green space[c] | 0.13 (0.09, 0.18) | 0.14 (0.10, 0.18) | <0.001 |
| NDVI[d] | 0.43 (0.30, 0.48) | 0.44 (0.35, 0.50) | <0.001 |
| Blue space[b] | 0.10 (0.07, 0.18) | 0.10 (0.07, 0.19) | 0.071 |
| Urban space[b] | 0.13 (0.06, 0.35) | 0.14 (0.06, 0.42) | <0.001 |

IBD, inflammatory bowel disease; NDVI, normalized difference vegetation index. [a]Wilcoxon rank sum tests; Pearson's Chi-squared test. [b]Proportion of municipal area (range 0–1). [c]Municipal mean biodiversity score (range 0–20). [d]Municipal mean NDVI (range −1 to 1).

***Table 1:*** **Baseline characteristics of the study cohort by inflammatory bowel disease status.**

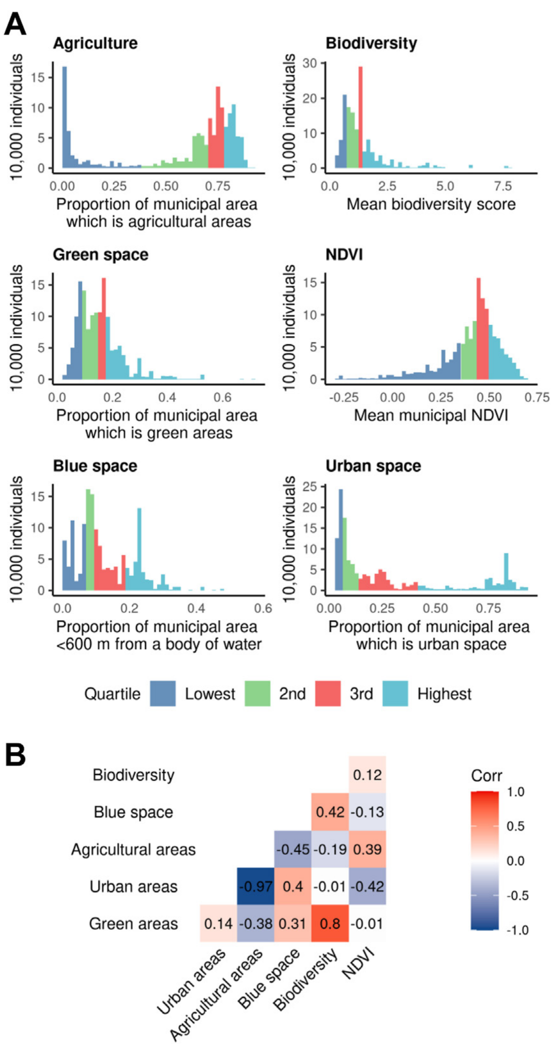


*Fig. 1:* **A**. Histograms of distributions of exposure variables by quartile and **B**. Correlation across variables. NDVI, normalized difference vegetation index.

quartiles of other environmental exposures and CD risk (Fig. 3A). While the third quartile of green space, compared to the lowest quartile, was associated with UC risk (aHR 1.14, 95% CI 1.01, 1.28), there was no association between the highest quartile of green space and UC, or between each of the remaining environmental exposures and UC (Fig. 3B).

### Sensitivity analyses

We conducted additional analyses to assess the robustness of our findings. First, we compared the highest decile for each exposure with the lowest quartile (Table 2). We found a protective effective of green space against IBD (0.84, 95% CI 0.75, 0.95) and CD (aHR 0.79, 95% CI 0.68, 0.92) and a protective effect of biodiversity against IBD (aHR 0.85, 95% CI 0.75, 0.97) and CD (aHR 0.80, 95% CI (0.66, 0.97). We noted a harmful effect of the highest decile of NDVI against CD (aHR 1.28, 95% 1.06, 1.55). We also noted a detrimental effect of middle quartile agricultural space on CD (aHR 1.16, 95% CI 1.02, 1.32). Next, we applied different definitions of blue space, using a distance of 300 m and 1200 m (in contrast to 600 m used in the main analysis) from a body of water. Estimates remained consistent across all analyses (Supplementary Table S2). Then, on moving urban green space from the green space category to the urban space category, estimates remained consistent (Supplementary Table S3). We also estimated the risk of IBD in the offspring with maternal exposure to environmental exposures during pregnancy and found that estimates were consistent with those in the main analysis (Supplementary Table S4). Similarly, when changing the definition of the early life period to include exposures until 5 years of age, estimates remained consistent (Supplementary Table S4).

## Discussion

In this analysis of a nationwide cohort of nearly 1.5 million individuals followed until a median age of 14 years, we report that early life exposure to agriculture land use is associated with an increased risk of CD, and exposure to biodiversity and green space are associated with a protective effect against IBD, after adjusting for relevant confounding variables. These associations are driven by an increase in CD risk, and there was no clear impact of early life environmental variables on UC risk. There was no association of NDVI, blue space, or urban space with CD or UC risk. Estimates were consistent across varying definitions of exposures and early life, indicative of the robustness of our findings. To our knowledge, this is the first report with a comprehensive analysis of early life environmental exposures pertaining to natural environment and land use in a population-based cohort.

Clustering of CD in agricultural areas has been reported in northern France in the EPIMAD registry.[8] Similarly, in British Columbia, Canada, spatial clustering of CD was reported in areas of agricultural petroleum oil pesticide application to orchards and grapes.[22] Petroleum oil, a fungicidal agent, alongside other pesticides, has been linked with rhinitis and antinuclear antibody positivity, suggesting a potentially proinflammatory effect of these agents.[23,24] In a comprehensive, integrated analysis of environmental chemicals from the United States Environmental Protection Agency toxicology database, Sanmarco et al. identified the top twenty chemicals that caused intestinal inflammation in zebrafish models; a majority of these chemicals were agricultural herbicidal, fungicidal and pesticidal compounds.[9] Our data lend further support to the hypothesis that agricultural -cidal agents may be associated with intestinal inflammation and CD risk.

| | IBD cases | 10,000 person-years at risk | HR (95% CI) unadjusted | HR (95% CI) adjusted |
|---|---|---|---|---|
| **Agriculture** | | | | |
| Lowest | 775 | 395.5 | 1 (Ref) | 1 (Ref) |
| 2nd | 885 | 415.6 | 1.08 (0.98 - 1.19) | 1.08 (0.97 - 1.21) |
| **3rd** | **1,113** | **454.2** | **1.11 (1.01 - 1.21)** | **1.13 (1.01 - 1.26)** |
| Highest | 995 | 443.6 | 1.07 (0.97 - 1.18) | 1.11 (0.98 - 1.26) |
| **Biodiversity** | | | | |
| Lowest | 966 | 419.6 | 1 (Ref) | 1 (Ref) |
| 2nd | 1,113 | 461.9 | 0.98 (0.90 - 1.07) | 0.98 (0.87 - 1.09) |
| **3rd** | **816** | **411.8** | **0.89 (0.81 - 0.98)** | **0.90 (0.81 - 0.99)** |
| Highest | 873 | 415.6 | 0.91 (0.83 - 1.00) | 0.92 (0.82 - 1.03) |
| **Green space** | | | | |
| Lowest | 1,062 | 453.3 | 1 (Ref) | 1 (Ref) |
| 2nd | 1,041 | 455.8 | 1.00 (0.92 - 1.09) | 1.03 (0.93 - 1.14) |
| 3rd | 719 | 357.3 | 1.02 (0.93 - 1.12) | 1.04 (0.95 - 1.14) |
| Highest | 946 | 442.5 | 0.92 (0.84 - 1.00) | 0.93 (0.85 - 1.03) |
| **NDVI** | | | | |
| Lowest | 1,095 | 392.0 | 1 (Ref) | 1 (Ref) |
| 2nd | 1,014 | 439.2 | 1.00 (0.92 - 1.09) | 1.00 (0.91 - 1.11) |
| 3rd | 1,010 | 460.1 | 1.00 (0.91 - 1.09) | 0.99 (0.90 - 1.10) |
| Highest | 649 | 417.6 | 1.07 (0.97 - 1.19) | 1.05 (0.94 - 1.17) |
| **Blue space** | | | | |
| Lowest | 1,062 | 455.4 | 1 (Ref) | 1 (Ref) |
| 2nd | 874 | 412.7 | 0.96 (0.88 - 1.05) | 0.96 (0.86 - 1.07) |
| 3rd | 990 | 430.0 | 1.00 (0.92 - 1.10) | 1.01 (0.91 - 1.12) |
| Highest | 842 | 410.8 | 0.93 (0.85 - 1.02) | 0.94 (0.84 - 1.05) |
| **Urban space** | | | | |
| Lowest | 1,004 | 456.4 | 1 (Ref) | 1 (Ref) |
| 2nd | 904 | 400.1 | 1.11 (1.01 - 1.21) | 1.09 (0.98 - 1.20) |
| 3rd | 1,089 | 464.6 | 1.03 (0.95 - 1.13) | 0.99 (0.85 - 1.16) |
| Highest | 771 | 387.8 | 0.99 (0.90 - 1.08) | 0.95 (0.82 - 1.11) |

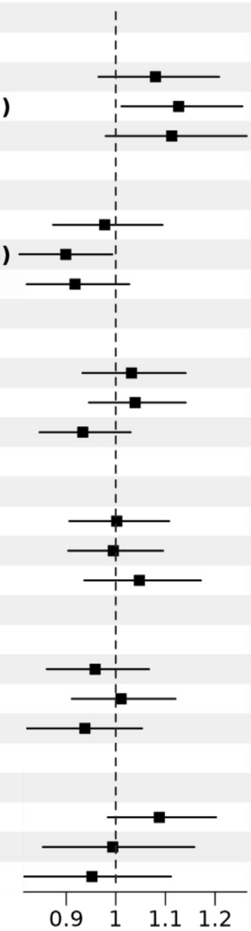


*Fig. 2:* Forest plot demonstrating the unadjusted and adjusted hazards ratios for the outcomes inflammatory bowel disease across quartiles of early life environmental exposure variables. The adjusted model is adjusted for sex (female, male), maternal age at delivery (continuous), calendar year of birth, municipal-level socioeconomic status, clustering on municipality of residence, and first-degree relative with IBD prior to the birth of the child (yes, no). Bold numbers indicate number of IBD cases, person-years at risk, and HRs for statistically significant adjusted HRs, p<0.05. CI, confidence interval; HR, hazard ratio; IBD, inflammatory bowel disease; NDVI, normalized difference vegetation index.

The consistent association of these chemicals with CD, but not with UC, is noteworthy and provides insights into differences between CD and UC pathogenesis. Certainly, differences between CD and UC epidemiology and risk factors have been previously reported.[2,13,25] CD is believed to have a longer preclinical course, in contrast to UC, which is considered to have a more abrupt onset.[26,27] Other potential explanations for our findings include loss of biodiversity, and thereby protective effect against CD, in regions with extensive agriculture. However, there is limited correlation between agriculture land use and biodiversity. In contrast, there is a positive correlation between NDVI and agricultural land use, which may explain the harmful effect of NDVI on CD in sensitivity analysis. The association between the third quartile for green space and UC is likely a chance finding, since this is the only association of an environmental exposures with UC and given the lack of a dose–response relationship.

Previous studies have reported on IBD risk with exposure to green space. Elten et al. measured the impact of average residential greenspace during pregnancy and childhood using the NDVI on IBD risk in Ontario, Canada.[6] They found a protective effect of green space against pediatric-onset IBD with a linear dose response relationship.[6] Similarly, Zhang et al.

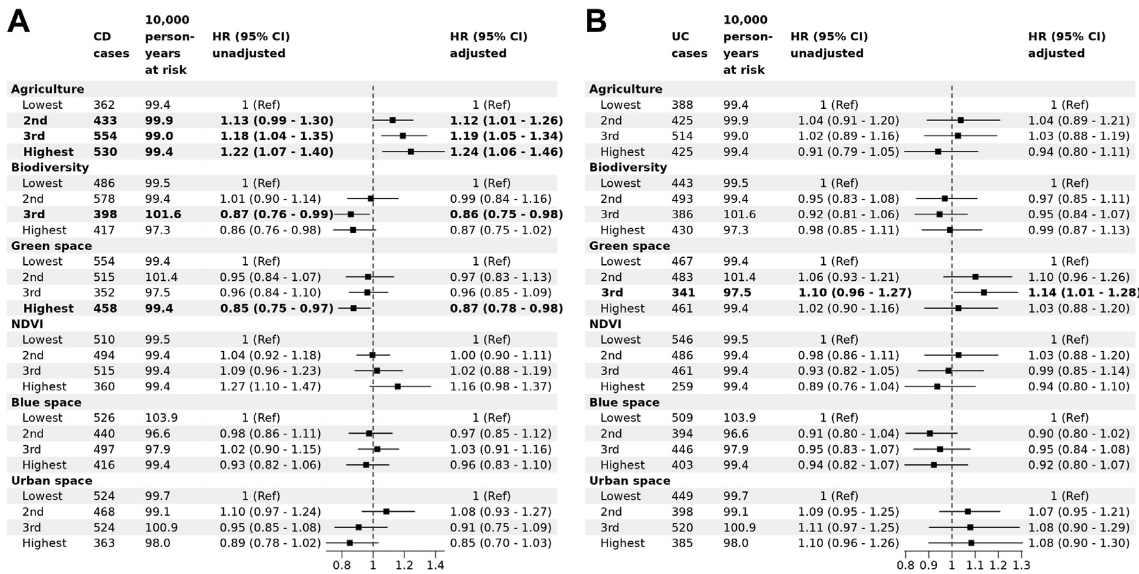


*Fig. 3:* Forest plot demonstrating the unadjusted and adjusted hazards ratios for outcomes (A) Crohn's disease and (B) Ulcerative colitis across quartiles of early life environmental exposure variables. A. The adjusted model is adjusted for sex (female, male), maternal age at delivery (continuous), calendar year of birth, municipal-level socioeconomic status, clustering on municipality of residence, and first-degree relative with IBD prior to the birth of the child (yes, no). CI, confidence interval; CD, Crohn's disease; HR, hazard ratio; NDVI, normalized difference vegetation index. B The adjusted model is adjusted for sex (female, male), maternal age at delivery (continuous), calendar year of birth, municipal-level socioeconomic status, clustering on municipality of residence, and first-degree relative with IBD prior to the birth of the child (yes, no). CI, confidence interval; HR, hazard ratio; NDVI, normalized difference vegetation index; UC, Ulcerative colitis. Bold numbers indicate number of CD / UC cases, person-years at risk, and HRs for statistically significant adjusted HRs, p<0.05.

| | Exposure category | 10,000 person-years at risk | IBD | | CD | | UC | |
|---|---|---|---|---|---|---|---|---|
| | | | Cases | Adjusted HR (95% CI) | Cases | Adjusted HR (95% CI) | Cases | Adjusted HR (95% CI) |
| Agriculture | Lowest quartile | 395.5 | 775 | 1 (REF) | 362 | 1 (REF) | 388 | 1 (REF) |
| | Middle | **1122.4** | 2571 | 1.10 (1.00–1.21) | **1285** | **1.16 (1.02–1.32)** | 1186 | 1.03 (0.89–1.17) |
| | Top decile | 191.0 | 422 | 1.03 (0.88–1.21) | 232 | 1.17 (0.96–1.43) | 178 | 0.89 (0.71–1.12) |
| Biodiversity | Lowest quartile | 419.6 | 966 | 1 (REF) | 486 | 1 (REF) | 443 | 1 (REF) |
| | Middle | 1123.8 | 2487 | 0.94 (0.86–1.04) | 1249 | 0.92 (0.81–1.05) | 1145 | 0.97 (0.87–1.09) |
| | **Top decile** | **165.5** | **315** | **0.85 (0.75–0.97)** | **144** | **0.80 (0.66–0.97)** | 164 | 0.95 (0.83–1.08) |
| Green space | Lowest quartile | 453.3 | 1062 | 1 (REF) | 554 | 1 (REF) | 467 | 1 (REF) |
| | Middle | 1071.8 | 2323 | 1.02 (0.94–1.11) | 1144 | 0.96 (0.85–1.07) | 1090 | 1.11 (0.99–1.25) |
| | **Top decile** | **183.8** | **383** | **0.84 (0.75–0.95)** | **181** | **0.79 (0.68–0.92)** | 195 | 0.94 (0.81–1.09) |
| NDVI | Lowest quartile | 392.0 | 1095 | 1 (REF) | 510 | 1 (REF) | 546 | 1 (REF) |
| | Middle | 1132.6 | 2356 | 1.00 (0.92–1.09) | 1188 | 1.02 (0.92–1.13) | 1082 | 1.00 (0.88–1.15) |
| | **Top decile** | **184.3** | 317 | 1.14 (0.97–1.33) | **181** | **1.28 (1.06–1.55)** | 124 | 1.02 (0.82–1.27) |
| Blue space | Lowest quartile | 455.4 | 1062 | 1 (REF) | 526 | 1 (REF) | 509 | 1 (REF) |
| | Middle | 1085.2 | 2340 | 0.97 (0.89–1.06) | 1164 | 0.98 (0.88–1.08) | 1077 | 0.93 (0.84–1.04) |
| | Top decile | 168.2 | 366 | 0.97 (0.85–1.11) | 189 | 1.06 (0.88–1.29) | 166 | 0.87 (0.74–1.04) |
| Urban space | Lowest quartile | 456.4 | 1004 | 1 (REF) | 524 | 1 (REF) | 449 | 1 (REF) |
| | Middle | 1144.1 | 2622 | 1.06 (0.96–1.18) | 1294 | 1.04 (0.89–1.20) | 1229 | 1.07 (0.95–1.21) |
| | Top decile | 108.4 | 142 | 1.00 (0.83–1.20) | 61 | 0.89 (0.63–1.26) | 74 | 1.06 (0.88–1.29) |

The adjusted model is adjusted for sex (female, male), maternal age at delivery (continuous), calendar year of birth, municipal-level socioeconomic status, clustering on municipality of residence, and first-degree relative with IBD prior to the birth of the child (yes, no). Bold numbers indicate person-years at risk, number of cases, and HRs for statistically significant HRs, p<0.05. CI, confidence interval; CD, Crohn's disease; HR, hazard ratio; NDVI, normalized difference vegetation index; UC, ulcerative colitis.

***Table 2:* Sensitivity analysis comparing the top decile for each environmental exposure to the bottom quartile for outcomes inflammatory bowel disease, Crohn's disease and ulcerative colitis.**

reported protective effects of green space, blue space, and natural environment, the latter based on land cover data, against IBD, among adults recruited to the UK Biobank.[7] Teich et al. found, in a case–control study, that self-reported access to a garden was lower among cases with pediatric-onset IBD, compared to matched siblings without IBD.[28] These data are in line with our findings of a protective effect of green space and higher biodiversity against CD, given the strong correlation between the two variables. Further, while we found no association between green space and IBD in the main analysis, we noted a protective effect of green space when comparing the top decile with the lowest quartile of green space. Broadly, these data, reporting on a protective impact of the natural environment on IBD risk, are consistent with those from our analysis. Given the correlation across variables pertaining to natural environment and that exposure to these variables occurs simultaneously (environmental mixture), the net effects of such "mixtures" are relevant. Pathways underlying these findings warrant further exploration in mechanistic studies of specific pollutants.[9] We are unable to differentiate between the impact of early life variables on pediatric-onset and adult-onset IBD given the relatively short follow up duration. Green space and biodiversity may have a direct causal effect on IBD, or indirect effect through diet, stress, physical activity, pollution, or other mechanism.[29] Certainly, green space exposure has been linked with enrichment of beneficial gut microbiome taxa.[30]

Our findings are consistent with observations of rising IBD in parallel with globalization, recent increase in IBD incidence in recently industrialized countries, and a higher incidence of IBD among immigrants to Europe and North America, relative to that in the emigration country.[1,5,31,32] Considering the rapid and alarming decline in the natural environment at the global level, particularly in disadvantaged communities and regions, these data emphasize the urgent need for environmental advocacy, policy change, and environmental justice.

The strengths of our study include the use of a nationwide population-based cohort, prospectively collected unbiased exposure and covariate data, and long term follow up through linked registers. Our study also has limitations. The distribution of environmental exposures in our study may not be representative of other populations. For example, Denmark is among the most intensively farmed countries with highly industrialized farming practices.[11] Denmark has a lower biodiversity relative to other European countries and relatively few individuals lived in high biodiversity areas. Exposure data were available for specific calendar years only and may not reflect accurately exposure at other time points. Place of residence was only available at the municipal level, which is a coarse measurement. For the land use variables, the CORINE inventory includes polygons of at least 25 ha, meaning that heterogeneity within each polygon cannot be assessed. Further, there are no data on how much time individuals spend in contact with nature or in other municipalities. Residence location, a complex exposure, is both a socioeconomic indicator and can be associated with lifestyle factors such as diet and smoking, which we did not have access to, and it is likely to affect risk of IBD in ways beyond the environmental exposures studied. Finally, as with all observational data, there is a risk of unmeasured and residual confounding.

In summary, in a nationwide cohort with prospective, long term follow up, we report that early life exposure to agriculture land use is associated increased risk, and exposure to biodiversity and green space is associated with a lower risk of CD. These data support the impact of early life factors towards IBD risk, with implications towards prevention. They also highlight the relevance of the natural environment towards health and disease and lend support to ongoing calls to prioritize environmental health and environmental justice.

### Contributors

MA: study concept and design, interpretation of data, drafting and critical revision of the manuscript for important intellectual content; AVH: study design; acquisition, analysis and interpretation of data, drafting and critical revision of the manuscript for important intellectual content; JFC: interpretation of data, critical revision of the manuscript for important intellectual content; TJ: interpretation of data, critical revision of the manuscript for important intellectual content; KHA: study concept and design, interpretation of data, critical revision of the manuscript for important intellectual content. The data were accessed and verified by AVH and KHA.

### Data sharing statement

The data underlying this article are available in the article and in its online supplementary material. The study is based on data from the Danish nationwide registers (https://sundhedsdatastyrelsen.dk). The register data is protected by the Danish Act on Processing of Personal Data and are accessed through application to and approval from the Danish Data Protection Agency and the Danish Health Data Authority.

### Declaration of interests

The corresponding author confirms on behalf of all authors that there have been no involvements that might raise the question of bias in the work reported or in the conclusions, implications, or opinions stated.

MA reports having consulted for Douglas Pharmaceuticals.

AVH reports no conflict of interest.

JFC reports receiving research grants from AbbVie, Janssen Pharmaceuticals and Takeda; receiving payment for lectures from AbbVie, Amgen, Allergan, Inc. Ferring Pharmaceuticals, Shire, and Takeda; receiving consulting fees from AbbVie, Amgen, Arena Pharmaceuticals, Boehringer Ingelheim, Bristol Myers Squibb, Celgene Corporation, Eli Lilly, Ferring Pharmaceuticals, Galmed Research, Glaxo Smith Kline, Geneva, Iterative Scopes, Janssen Pharmaceuticals, Kaleido Biosciences, Landos, Otsuka, Pfizer, Prometheus, Sanofi, Takeda, TiGenix,; and hold stock options in Intestinal Biotech Development.

TJ reports having consulted for Ferring.

KHA reports no conflict of interest.

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**Appendix A. Supplementary data**

Supplementary data related to this article can be found at https://doi.org/10.1016/j.eclinm.2024.102514.

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